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Assessing Interactions between Common Genetic Variant on 2q35 and Hormone Receptor Status with Breast Cancer Risk: Evidence Based on 26 Studies

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Abstract

Genome-wide association studies have identified 2q35-rs13387042 as a new breast cancer (BC) susceptibility locus in populations of European descent. Since then, the relationship between 2q35-rs13387042 and breast cancer has been reported in various ethnic groups; however, these studies have yielded inconsistent results. To investigate this inconsistency, we performed a meta-analysis of 26 studies involving a total of 101,529 cases and 167,363 controls for 2q35-rs13387042 polymorphism to evaluate its effect on genetic susceptibility for breast cancer. An overall random effects odds ratio of 1.14 (95% CI: 1.11–1.16, $P < 10^{-5}$) was found for rs13387042-A variant. Significant results were also observed using dominant (OR = 1.14, 95% CI: 1.12–1.17, $P < 10^{-5}$), recessive (OR = 1.17, 95% CI: 1.13–1.21, $P < 10^{-5}$) and co-dominant genetic model (heterozygous: OR = 1.15, 95% CI: 1.12–1.19, $P < 10^{-5}$; homozygous: OR = 1.20, 95% CI: 1.15–1.24, $P < 10^{-5}$). There was strong evidence of heterogeneity, which largely disappeared after stratification by ethnicity. Significant associations were found in East Asians, and White populations when stratified by ethnicity; while no significant associations were observed in Africans and other ethnic populations. An association was observed for both ER-positive (OR = 1.17, 95% 1.15–1.19; $P < 10^{-5}$) and ER-negative disease (OR = 1.08, 95% CI: 1.04–1.13; $P < 10^{-4}$) and both progesterone receptor (PR)-positive (OR = 1.18, 95% CI: 1.15–1.21; $P < 10^{-5}$) and PR-negative disease (OR = 1.10, 95% CI: 1.05–1.15; $P < 10^{-4}$). In conclusion, this meta-analysis demonstrated that the A allele of 2q35-rs13387042 is a risk factor associated with increased breast cancer susceptibility.

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Introduction

Breast cancer is the most common cancer and the leading cause of cancer death among women worldwide, accounting for 23% of the total cancer cases and 14% of the cancer deaths in 2008 [1]. The mechanism of breast carcinogenesis is still not fully understood. It has been suggested that environmental and genetic factors may affect the individual's susceptibility to cancer [2]. High-penetrance breast cancer susceptibility genes, such as BRCA1 and BRCA2, explain only a small fraction of breast cancers in the general population because of their low mutation rates [3]. Over the past decades, the candidate approach has ever been successfully employed to identify BC susceptibility, such as ATM and XRCC1 of DNA repair genes have been confirmed to be associated with BC risk [4–6]. However, most of the genetic variants identified by candidate-gene studies have not been replicated [7]. Recently, several genome-wide association studies (GWAS) have been conducted and identified genetic susceptibility loci that are associated with breast cancer risk [8–11]. The rs13387042 polymorphism at chromosome 2q35 has been identified as a new hotspot for breast cancer susceptibility by a

recent GWA study [12]. Associations between the 2q35-rs13387042 polymorphism and breast cancer have been independently replicated by subsequent studies; however, a proportion of them have produced contrary results. Growing evidence suggests substantial heterogeneity by tumor subtype, defined by hormone receptor status, for association with the polymorphism [9,12]. Because estrogen receptor (ER) and progesterone receptor (PR) statuses are the major markers of breast cancer subtypes, these observations suggest that inherited risk variants of these subtypes may vary. The lack of concordance across many of these studies reflects limitation in the studies, such as small sample size, ethnic difference, and study design. With the increased studies in recent years among East Asians, Africans and some other ethnic populations, there is a need to reconcile this inconsistency and to clarify the problems in previous studies. We therefore performed a meta-analysis of the published studies to clarify this inconsistency and to establish a comprehensive picture of the relationship between 2q35 rs13387042 polymorphism and breast cancer.

Materials and Methods

Literature search strategy and inclusion criteria

Papers published before the end of January 2013 were identified through a search of Pubmed, SCOPUS, ISI web of knowledge, Embase and Cochrane databases Search term combinations were keywords relating to the chromosome 2q35 (e.g., "chromosome 2q35", and "rs13387042") in combination with words related to breast cancer (e.g., "breast cancer", "breast carcinoma", "malignant breast neoplasm") and polymorphism or variation. The titles and abstracts of potential articles were screened to determine their relevance, and any clearly irrelevant studies were excluded. The full texts of the remaining articles were read to determine whether they contained information on the topic of interest. In addition, all reference lists from the main reports and relevant reviews were hand searched for additional eligible studies not indexed by Medline.

Inclusion criteria and data extraction

Eligible studies had to meet all of the following criteria: (1) original papers containing independent data which have been published in peer-reviewed journal, (2) case–control or cohort studies, (3) genotype distribution information or odds ratio (OR) with its 95% confidence interval (CI) and *P*-value, (4) genotype distribution of control group must be consistent with Hardy–Weinberg equilibrium (HWE).

For each qualified study, the following information was extracted independently and entered into separate databases by two authors: first author's surname, publication date, ethnicity, source of control subjects, genotyping method, age, tumor stage, histopathological subtype, ER status, PR status, total number of cases and controls, and genotype frequency in cases and controls. The results were compared, and disagreements were discussed among all authors and resolved with consensus. For studies including subjects of different ethnic groups, data were extracted separately according to ethnicity. If multiple published reports from the same study population were available, we included only the one with largest sample size and the most detailed information. Meanwhile, different case–control groups in one study were considered as independent studies.

Statistical methods

The meta-analysis examined the association between the rs13387042 polymorphism and the risk of BC, for the: (i) allele contrast, (ii) dominant, (iii) recessive, and (iv) co-dominant models [13]. Crude ORs with 95% CIs were calculated using raw data, according to the method of Woolf B [14]. Cochran's Q-statistic test was performed to assess possible heterogeneity in the combined studies [15]. Both fixed-effects (Mantel–Haenszel method) [16] and random-effects (DerSimonian–Laird method) [17] models were performed to calculate the pooled ORs. Owing to a priori assumptions about the likelihood of heterogeneity between primary studies, the random-effects model, which usually is more conservative, was chosen. Sub-group analyses and meta-regression were used to explore heterogeneity [18]. Ethnicity, study design (GWAS vs. candidate gene study), ER status (ER-positive vs. ER-negative), PR status (PR-positive vs. PR-negative) and invasiveness (invasive vs. in situ) were prespecified as characteristics for assessment of heterogeneity. Ethnic group was defined as White (i.e., people of European origin), East Asian (e.g. Chinese, Japanese, and Korean), African and others (e.g., Jew and Hawaiian). One-way sensitivity analysis was performed to assess the stability of the results, namely, a single study in the meta-analysis was deleted each time to reflect the influence of the individual data set to the pooled OR. Funnel plots and the Egger's test were used to examine the influence of publication bias (linear regression analysis) [19]. All P values are two-sided at the $P = 0.05$ level. All of the statistical tests used in this meta-analysis were performed by STATA version 10.0 (Stata Corporation, College Station, TX).

Results

Characteristics of included studies

The combined search yielded 113 references. 87 articles were excluded because they clearly did not meet the criteria or overlapping references (Figure S1). Finally, a total of 26 eligible association studies were included involving 101,529 breast cancer cases and 167,363 controls [12,20–44]. Of the cases, 80% were White, 12% were East Asian, 7% were African descent, and 1% were of other ethnic origins. The main study characteristics were summarized in Table 1.

Association of 2q35-rs13387042 with breast cancer

There was a wide variation in the A allele frequency of the rs13387042 polymorphism among the controls across different ethnicities, ranging from 0.05 to 0.75 (Table 1). For East Asian controls, the A allele frequency was 0.12 (95% CI: 0.08–0.16), which was lower than that in White controls (0.51; 95% CI: 0.48–0.53) and African controls (0.72; 95% CI: 0.65–0.79).

The main results of this meta-analysis were listed in Table 2 and Table S1. In the overall analysis, the rs13387042 polymorphism was significantly associated with elevated breast cancer risk with a per-allele OR of 1.14 [95% CI: 1.11–1.16, $P(Z)<10^{-5}$, $P(Q)<10^{-5}$; Figure 1], with corresponding results under dominant and recessive genetic models of 1.14 [95% CI: 1.12–1.17, $P(Z)<10^{-5}$, $P(Q) = 0.11$] and 1.17 [95%CI: 1.13–1.21, $P(Z)<10^{-5}$, $P(Q)<10^{-5}$]. Significant associations were also found for co-dominant genetic model [heterozygous: OR = 1.15, 95% CI: 1.12–1.19, $P(Z)<10^{-5}$, $P(Q)<10^{-4}$; homozygous: OR = 1.20, 95% CI: 1.15–1.24, $P(Z)<10^{-5}$, $P(Q)<10^{-5}$]. In the stratified analysis by ethnicity, significantly increased risks were found among East Asians [A allele: OR = 1.12, 95% CI: 1.03–1.21, $P(Z) = 0.004$, $P(Q) = 0.18$; dominant model: OR = 1.10, 95% CI: 1.03–1.18, $P(Z) = 0.003$, $P(Q) = 0.27$; recessive model: OR = 1.09, 95% CI: 1.02–1.19, $P(Z) = 0.01$, $P(Q) = 0.63$; heterozygous: OR = 1.11, 95% CI: 1.04–1.20, $P(Z) = 0.001$, $P(Q) = 0.33$; homozygous: OR = 1.10, 95% CI: 1.02–1.19, $P(Z)<10^{-4}$, $P(Q) = 0.25$] and White populations [A allele: OR = 1.14, 95% CI: 1.12–1.17, $P(Z)<10^{-5}$, $P(Q) = 0.02$; dominant model: OR = 1.16, 95% CI: 1.13–1.18, $P(Z)<10^{-5}$, $P(Q) = 0.59$; recessive model: OR = 1.20, 95% CI: 1.14–1.24, $P(Z)<10^{-5}$, $P(Q)<10^{-4}$; heterozygous: OR = 1.15, 95% CI: 1.13–1.18, $P(Z)<10^{-5}$, $P(Q) = 0.002$; homozygous: OR = 1.21, 95% CI: 1.15–1.25, $P(Z)<10^{-5}$, $P(Q)<10^{-4}$]. However, no significant associations were detected among African [A allele: OR = 1.07, 95% CI: 0.99–1.16, $P(Z) = 0.17$, $P(Q) = 0.03$] and other ethnic populations [A allele: OR = 1.24, 95% CI: 0.59–2.61, $P(Z) = 0.57$, $P(Q)<10^{-4}$]. Subsidiary analyses of study design yielded a per-allele OR for GWAS of 1.16 [95% CI: 1.14–1.19, $P(Q)<10^{-5}$] and for candidate gene study of 1.11 [95% CI: 1.08–1.15, $P(Q)<10^{-5}$].

We further performed analyses to test for differences in the associations of the polymorphism with breast cancer risk with respect to different prognostic factors. Specifically, we compared estrogen receptor–positive (ER+) case subjects with ER-negative (ER−) case subjects, and in a similar fashion progesterone receptor-positive (PR+) case subjects with receptor–negative (PR−) case subjects. Stratification of tumors by ER status

Table 1. Characteristics of the studies included in the meta-analysis.

Study	Year	Ethnicity	Genotyping method	No. of cases/controls	Control source	RAF in cases/controls	Study design
Stacey [12]	2008	European	SNP Array	4420/17365	GP	0.54/0.50	GWAS
Milne [20]	2009	European, Asian	SNP Array, iPLEX	31511/35969	GP, HP	0.55/0.51	GWAS
Zheng [21]	2009	African	Massarray	810/1784	GP	0.77/0.74	Candidate gene
Antoniou [22]	2009	European, American	TaqMan, iPLEX	7805/6675	GP	0.53/0.51	Candidate gene
Reeves [23]	2010	British	TaqMan	10306/10393	GP	0.54/0.50	Candidate gene
Hemminki [24]	2010	European	iPLEX	1415/1830	GP	0.57/0.54	Candidate gene
Zheng [25]	2010	Chinese	SNP Array	3039/3082	GP	0.11/0.11	Candidate gene
Barnholtz-Sloan [26]	2010	American	GoldenGate	1230/1117	GP	0.55/0.53	Candidate gene
Teraoka [27]	2011	European, American	Golden Gate	704/1386	GP	0.55/0.52	Candidate gene
Fletcher [28]	2011	British	SNP Array, GoldenGate	7643/7443	GP	0.53/0.52	GWAS
Campa [29]	2011	American, European, African, Asian, Hawaiian	SNP Array, TaqMan	8314/11589	GP	0.52/0.49	GWAS
Jiang [30]	2011	Chinese	SNaPshot	492/510	GP	0.12/0.10	Candidate gene
Li [31]	2011	European	SNP Array	1557/4584	GP	0.48/0.47	Candidate gene
Chen [32]	2011	African	SNP Array	3016/2745	GP	0.73/0.72	Candidate gene
Slattery [33]	2011	American	TaqMan	1733/2041	GP	0.53/0.52	Candidate gene
Stevens [34]	2011	European, American, Australian	iPLEX	2977/4976	GP	0.53/0.51	Candidate gene
Hutter [35]	2011	African	SNP Array	316/7484	GP	0.69/0.70	Candidate gene
Dai [36]	2012	Chinese	TaqMan	1771/1851	GP	0.13/0.11	Candidate gene
He [37]	2012	European	TaqMan	3683/34174	GP	0.55/0.50	Candidate gene
Shan [38]	2012	Tunisian	TaqMan	640/367	GP	0.58/0.55	Candidate gene
Kim [39]	2012	Korean	SNP Array, TaqMan	2257/2052	GP	0.10/0.10	GWAS
Huo [40]	2012	African	GoldenGate	1509/1383	GP	0.77/0.75	Candidate gene
Lin [41]	2012	Chinese	SNP Array	88/69	GP	0.15/0.06	Candidate gene
Harlid [42]	2012	European	MassARRAY	3393/4837	GP	0.53/0.50	Candidate gene
Sueta [43]	2012	Japanese	TaqMan	697/1394	HP	0.10/0.10	Candidate gene
Rinella [44]	2013	Jewish	KASPar	203/263	GP	0.66/0.52	Candidate gene

GP: general population, HP: hospital patient, RAF: risk allele frequency.
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indicated that rs13387042 had a stronger association with ER-positive [per-allele OR = 1.17, 95% 1.15–1.19; P(Z)<10^{-5}; P(Q) = 0.47] than ER-negative tumors [per-allele OR = 1.08, 95% CI: 1.04–1.13; P(Z)<10^{-4}; P(Q) = 0.18] (Figure 2). Similarly, a stronger association was also observed for the polymorphism with PR-positive tumors [per-allele OR = 1.18, 95% CI: 1.15–1.21; P(Z)<10^{-5}; P(Q) = 0.57] compared with PR-negative tumors [per-allele OR = 1.10, 95% CI: 1.05–1.15; P(Z)<10^{-4}; P(Q) = 0.16] (Figure 3).

The effect of the polymorphism was assessed for over-all breast cancer risk. No association was established between the polymorphism and tumor invasiveness. The data on tumor invasiveness were available in three studies, which included 20442 breast cancer patients. For 2q35-rs13387042, there appeared to be a similar per-allele OR for in situ cancer [per-allele OR = 1.17, 95% CI: 1.04–1.13; P(Z)<10^{-5}; P(Q) = 0.27] as compared to invasive cancer [per-allele OR = 1.19, 95% CI: 1.15–1.22; P(Z)<10^{-5}; P(Q) = 0.51].

Significant heterogeneity was present among the 44 data sets from 26 studies of the rs13387042 polymorphism (P<0.05). In meta-regression analysis, sample size (P = 0.21), source of controls (P = 0.15) and genotyping method (P = 0.73) did not significantly explain such heterogeneity. By contrast, ethnicity (P = 0.004) was significantly correlated with the magnitude of the genetic effect.

Sensitivity analyses and publication bias

Influence analysis was performed to assess the influence of each individual study on the pooled OR by sequential removal of individual studies. The results suggested that no individual study significantly affected the pooled OR, thus suggesting that the results of this meta-analysis are stable (data not shown). The shape of the funnel plot did not indicate any evidence of obvious asymmetry (Figure S2), thus suggesting no publication bias among the studies included. The statistical results still did not show preferential publication of positive findings in smaller studies (Begg's test, P = 0.27; Egger's test, P = 0.63).

Discussion

GWAS have led to the identification of multiple new genetic variants associated with breast cancer risk. Most of these breast cancer GWAS and replication studies have been conducted in European populations [11,12,24,28] and to a lesser extent in East Asians [25,30,36]. However, there are significant differences in allele frequencies and the prevalence of breast cancer among different populations. It is, therefore, important to quantitatively assess the effects of the GWAS-identified markers in different ethnic populations and explore potential heterogeneity of published data. To the best of our knowledge, this is the first

Table 2. Meta-analysis of the 2q35-rs13387042 polymorphism on breast cancer risk.

Sub-group analysis	No. of data sets	No. of cases/controls	A allele				Dominant model				Recessive model			
			OR (95%CI)	P(Z)	P(Q)[a]	P(Q)[b]	OR (95%CI)	P(Z)	P(Q)[a]	P(Q)[b]	OR (95%CI)	P(Z)	P(Q)[a]	P(Q)[b]
Total	44	101529/167363	1.14 (1.11–1.16)	$<10^{-5}$	$<10^{-5}$		1.14 (1.12–1.17)	$<10^{-5}$	0.11		1.17 (1.13–1.21)	$<10^{-5}$	$<10^{-5}$	
Ethnicity						0.04				0.02				0.03
White	26	82814/140849	1.14 (1.12–1.17)	$<10^{-5}$	0.02		1.16 (1.13–1.18)	$<10^{-5}$	0.59		1.20 (1.14–1.24)	$<10^{-5}$	$<10^{-4}$	
East Asian	9	11681/11773	1.12 (1.03–1.21)	0.004	0.18		1.10 (1.03–1.18)	0.003	0.27		1.09 (1.02–1.19)	0.01	0.63	
African	7	6692/14193	1.07 (0.99–1.16)	0.07	0.13		1.07 (0.99–1.17)	0.09	0.12		1.06 (0.94–1.21)	0.37	0.22	
Other	2	342/548	1.24 (0.59–2.61)	0.57	$<10^{-4}$		1.07 (0.66–1.76)	0.77	0.06		1.45 (0.89–2.09)	0.52	0.004	
Study design						0.20				0.11				0.07
GWAS	5	54145/74418	1.16 (1.14–1.19)	$<10^{-5}$	0.38		1.16 (1.14–1.19)	$<10^{-5}$	0.25		1.18 (1.15–1.21)	$<10^{-5}$	0.16	
Candidate gene	39	47384/92945	1.11 (1.08–1.15)	$<10^{-5}$	0.001		1.12 (1.09–1.16)	$<10^{-5}$	0.003		1.15 (1.11–1.19)	$<10^{-5}$	$<10^{-4}$	

[a] Cochran's chi-square Q statistic test used to assess the heterogeneity in subgroups.
[b] Cochran's chi-square Q statistic test used to assess the heterogeneity between subgroups.
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comprehensive meta-analysis which comprises a total of 101,529 cases and 167,363 controls from 26 studies, examining the association of 2q35 rs13387042 polymorphism with breast cancer risk.

Our results demonstrated that the A allele of the 2q35-rs13387042 polymorphism is a risk factor for developing breast cancer. In the stratified analysis by ethnicity, significant associations were found in East Asians and Whites for the polymorphism in all genetic models. However, no significant associations were detected among African and other ethnic populations. There are some points should be concerned for such inconsistent results. Firstly, ethnic differences may attribute to these different results, since the distributions of the 2q35-rs13387042 polymorphism were different between various ethnic populations. For instance, the frequencies of risk-A allele differs from 6% in Chinese population [41], 51% in Whites [12,23,28], to 72% in African descents [21,32,35]. On the other hand, a polymorphism may be in close linkage with another nearby causal variant in one ethnic population but not in another. 2q35-rs13387042 polymorphism may be in close linkage with different nearby causal variants in different populations. Furthermore, study design or small sample size or some environmental factors may affect the results. Most of these studies did not consider most of the important environmental factors. It is possible that variation at this locus has modest effects on breast cancer, but environmental factors may predominate in the progress of breast cancer, and mask the effects of this variation. Specific environmental factors like lifestyle and hormone replacement therapy that have been already well studied in recent decades [2,45]. The unconsidered factors mixed together may cover the role of 2q35-rs13387042 polymorphism. Thus, even if the variation has a causal effect on breast cancer, it may take a long time to be observed. Therefore, it is not surprising that inconsistent results for 2q35-rs13387042 polymorphism were found in breast cancer susceptibility.

The original publication on 2q35-rs13387042 [12] reported that the associated risk was confined to ER-positive breast cancer. We found that the association with rs13387042 was apparent for both ER-positive and ER-negative disease. However, the association appeared to be slightly stronger for ER-positive disease. This tendency to be more strongly associated with the risk of ER-positive breast cancer has been observed for other clearly established susceptibility SNPs, notably FGFR2-rs2981582, 8q-rs13281615, and 5p-rs10941679 [12,46,47], perhaps reflecting the fact that they were initially identified by GWASs for which most of the case patients in the hypothesis-generating phases had ER-positive disease. In addition, we also found that the association appeared to be much stronger for PR-positive than the PR-negative breast cancer. It is unclear whether PR status has an effect on breast carcinogenesis independent of ER status. About 65% of ER-positive breast cancers are also PR-positive, and there is a high correlation between ER and PR expression [48,49]. In addition, the per-allele odds ratio estimates were very similar for invasive and in situ disease.

A number of factors predict breast cancer, however, detailed pathogenesis mechanisms of breast cancer remain a matter of speculation. 2q35-rs13387042 is located in a 90-kb region of high linkage disequilibrium that contains neither known genes nor non coding RNAs [12,29]. The causal variant (or variants) in this region has (have) not been determined, and it is possible that one or more SNPs may confer a higher risk than 2q35-rs13387042. Thus, functional studies in this region are likely to lead to a better understanding of mechanisms of carcinogenesis and progression of breast cancer. However, the ORs we obtained were small with narrow CIs. This indicates that when considered alone as a genetic
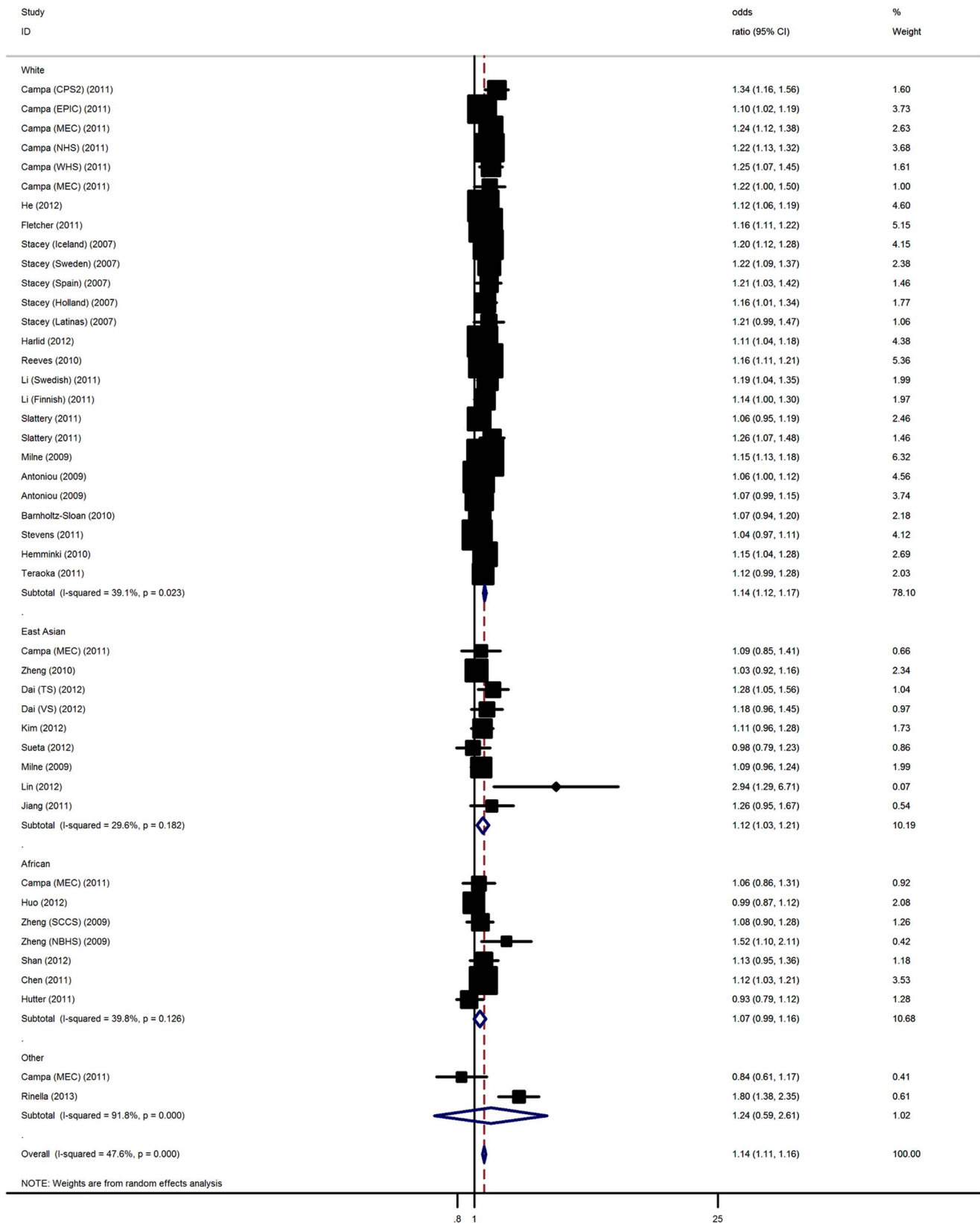
Figure 1. Forest plot from the meta-analysis of breast cancer risk and 2q35-rs13387042 polymorphism.
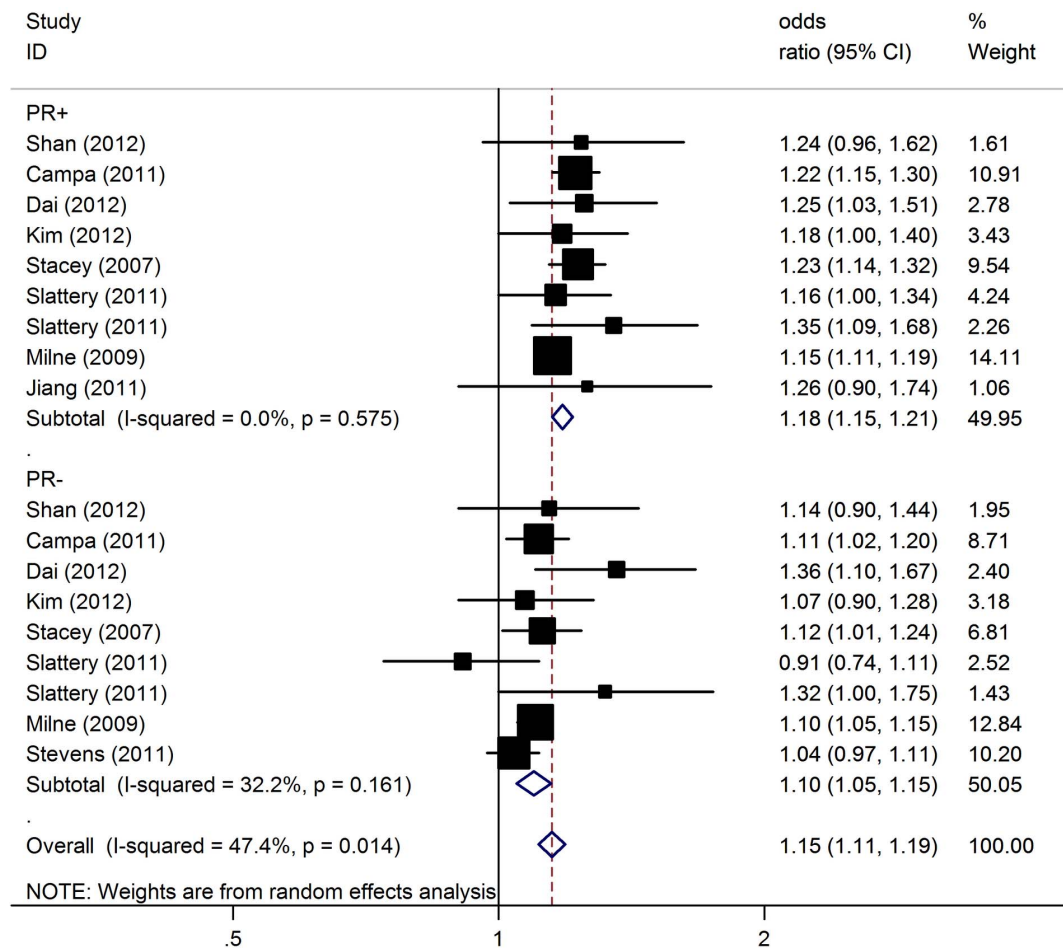doi:10.1371/journal.pone.0069056.g001
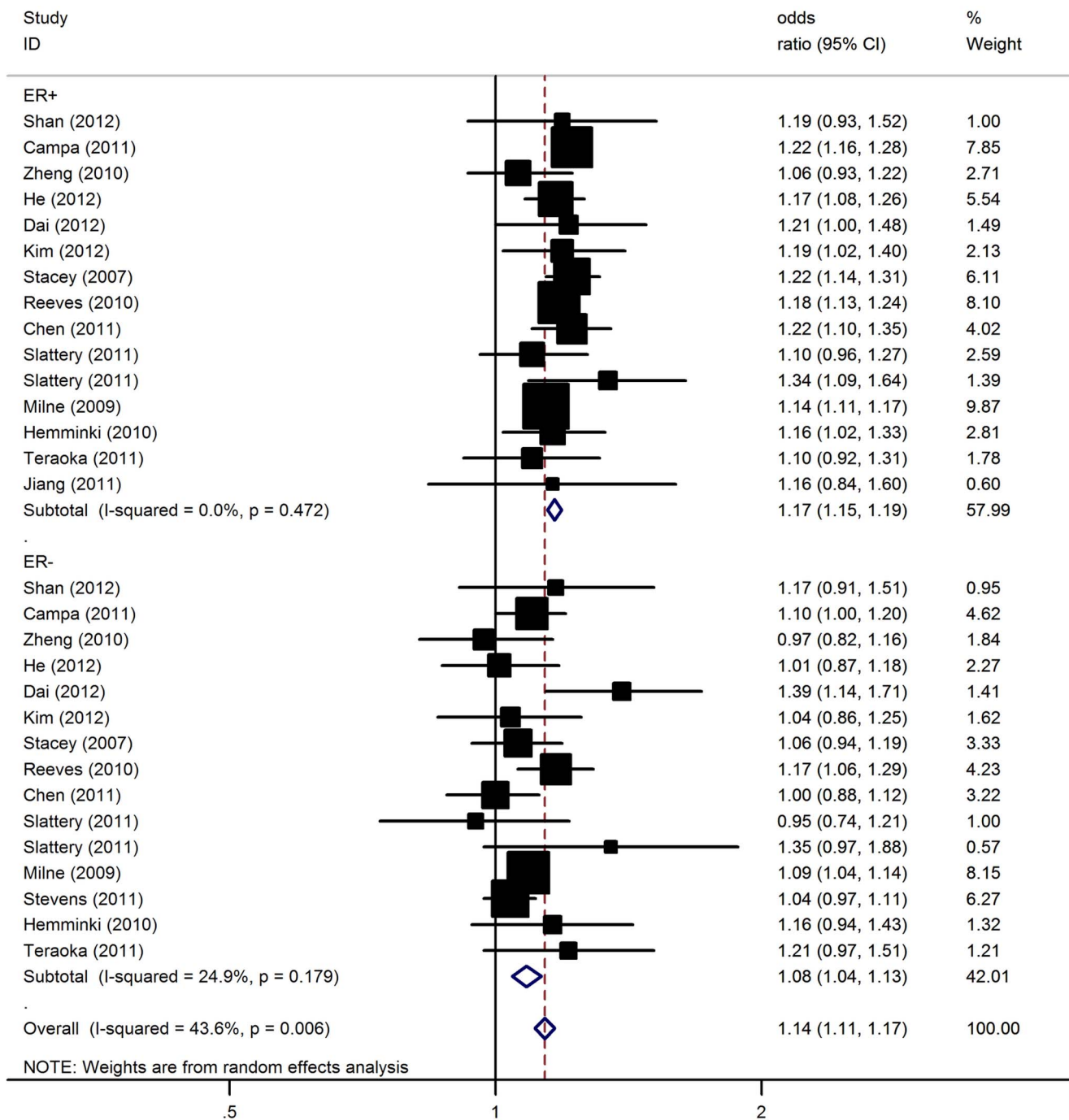
Figure 2. Association between 2q35-rs13387042 and breast cancer risk by ER status.
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factor, the 2q35-rs13387042 polymorphism has a very small but detectable effect on susceptibility to breast cancer. This could be regarded simply as a weak genetic effect that has an additive effect when combined with other susceptibility loci.

Compared with the previous meta-analysis [50], the present study is much larger, with almost sixty times as many cases as the earlier meta-analysis. In addition, we also performed analyses to test for differences in the associations of the polymorphism with breast cancer risk with respect to different hormone receptor status. Furthermore, we explored potential sources of heterogeneity across studies.

Limitations also inevitably existed in this meta-analysis. First, our meta-analysis is based on unadjusted estimates, whereas a more precise analysis could be performed if individual data were available, which would allow for an adjustment estimate. To be made, however, this approach requires the authors of all of the published studies to share their data. Second, no statistically significant association between the polymorphism and breast cancer appeared in other ethnic populations in racial subgroup analysis. However, the other ethnic population reports in the subgroup analysis include a mixture of populations from very distant countries, so the result must be interpreted with caution.

Figure 3. Association between 2q35-rs13387042 and breast cancer risk by PR status.
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Finally, the subgroup meta-analyses considering interactions between rs13387042 polymorphism and hormone receptor status, as well as tumor invasiveness were performed on the basis of a fraction of all the possible data to be pooled, so selection bias may have occurred and our results may be overinflated. Nevertheless, the total number of subjects included in this part of the analysis comprises the largest sample size so far.

Despite these limitations, this meta-analysis suggests that 2q35-rs13387042 polymorphism was significantly associated with increased risk of breast cancer, particularly in East Asian and white populations. As studies among other ethnic populations are currently limited, further studies including a wider spectrum of subjects to investigate the role of this variant in other populations will be needed.

Supporting Information

Figure S1 Flow chart of literature search.
(TIF)

Figure S2 Begg's funnel plot of 2q35-rs13387042 polymorphism and breast cancer risk (allele contrast).
(TIF)

Table S1 Meta-analysis of the 2q35-rs13387042 polymorphism on breast cancer risk using co-dominant model.
(DOCX)

Checklist S1   PRISMA 2009 Checklist.
(DOC)

Author Contributions

Conceived and designed the experiments: TH JH QYW JS. Performed the experiments: TH JH WLL QQY KLN. Analyzed the data: TH JH WLL QQY KLN. Contributed reagents/materials/analysis tools: WLL QQY KLN. Wrote the paper: TH JH QYW JS.

References

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, et al. (2011) Global cancer statistics. CA Cancer J Clin 61:69–90
2. Lichtenstein P, Holm NV, Verkasalo PK, Iliadou A, Kaprio J, et al. (2000) Environmental and heritable factors in the causation of cancer-analyses of cohorts of twins from Sweden, Denmark, and Finland. N Engl J Med 343:78–85
3. Walsh T, Casadei S, Coats KH, Swisher E, Stray SM, et al. (2006) Spectrum of mutations in BRCA1, BRCA2, CHEK2, and TP53 in families at high risk of breast cancer. JAMA 295:1379–1388.
4. Sun T, Miao X, Wang J, Tan W, Zhou Y, et al. (2004) Functional Phe31Ile polymorphism in Aurora A and risk of breast carcinoma. Carcinogenesis 25:2225–2230

5. Renwick A, Thompson D, Seal S, Kelly P, Chagtai T, et al. (2006) ATM mutations that cause ataxia-telangiectasia are breast cancer susceptibility alleles. Nat Genet 38:873–875

6. Liu L, Yuan P, Wu C, Zhang X, Guo H, et al. (2011) A functional −77T[C polymorphism in XRCC1 is associated with risk of breast cancer. Breast Cancer Res Treat 125:479–487

7. Zhang B, Beeghly-Fadiel A, Long J, Zheng W (2011) Genetic variants associated with breast-cancer risk: comprehensive research synopsis, meta-analysis, and epidemiological evidence. Lancet Oncol 12:477–488

8. Easton DF, Pooley KA, Dunning AM, Pharoah PD, Thompson D, et al. (2007) Genome-wide association study identifies novel breast cancer susceptibility loci. Nature 447:1087–1093

9. Hunter DJ, Kraft P, Jacobs KB, Cox DG, Yeager M, et al. (2007) A genome-wide association study identifies alleles in FGFR2 associated with risk of sporadic postmenopausal breast cancer. Nat Genet 39:870–874

10. Zheng W, Long J, Gao YT, Li C, Zheng Y, et al. (2009) Genome-wide association study identifies a new breast cancer susceptibility locus at 6q25.1. Nat Genet 41:324–328

11. Thomas G, Jacobs KB, Kraft P, Yeager M, Wacholder S, et al. (2009) A multistage genome-wide association study in breast cancer identifies two new risk alleles at 1p11.2 and 14q24.1 (RAD51L1). Nat Genet 41:579–584

12. Stacey SN, Manolescu A, Sulem P, Rafnar T, Gudmundsson J, et al. (2007) Common variants on chromosomes 2q35 and 16q12 confer susceptibility to estrogen receptor-positive breast cancer. Nat Genet 39:865–869

13. Palomaki GE, Melillo S, Bradley LA (2010) Association between 9p21 genomic markers and heart disease: a meta-analysis. JAMA 303:648–656

14. Woolf B (1955) On estimating the relation between blood group and disease. Ann Hum Genet 19: 251–253

15. Cochran WG (1954) The combination of estimates from different experiments. Biometrics 10: 101–129

16. Mantel N, Haenszel W (1959) Statistical aspects of the analysis of data from retrospective studies of disease. J Natl Cancer Inst 22: 719–748

17. DerSimonian R, Laird N (1986) Meta-analysis in clinical trials. Control Clin Trials 7:177–188

18. van Houwelingen HC, Arends LR, Stijnen T (2002) Advanced methods in meta-analysis: multivariate approach and meta-regression. Stat Med 21:589–624

19. Egger M, Davey Smith G, Schneider M, Minder C (1997) Bias in meta-analysis detected by a simple, graphical test. BMJ 315: 629–634

20. Milne RL, Benítez J, Nevanlinna H, Heikkinen T, Aittomäki K, et al. (2009) Risk of estrogen receptor-positive and -negative breast cancer and single-nucleotide polymorphism 2q35-rs13387042. J Natl Cancer Inst 101:1012–1018

21. Zheng W, Cai Q, Signorello LB, Long J, Hargreaves MK, et al. (2009) Evaluation of 11 breast cancer susceptibility loci in African-American women. Cancer Epidemiol Biomarkers Prev 18:2761–2764

22. Antoniou AC, Sinilnikova OM, McGuffog L, Healey S, Nevanlinna H, et al. (2009) Common variants in LSP1, 2q35 and 8q24 and breast cancer risk for BRCA1 and BRCA2 mutation carriers. Hum Mol Genet 18:4442–4456

23. Reeves GK, Travis RC, Green J, Bull D, Tipper S, et al. (2010) Incidence of breast cancer and its subtypes in relation to individual and multiple low-penetrance genetic susceptibility loci. JAMA 304:426–434

24. Hemminki K, Müller-Myhsok B, Lichtner P, Engel C, Chen B, et al. (2010) Low-risk variants FGFR2, TNRC9 and LSP1 in German familial breast cancer patients. Int J Cancer 126:2858–2862

25. Zheng W, Wen W, Gao YT, Shyr Y, Zheng Y, et al. (2010) Genetic and clinical predictors for breast cancer risk assessment and stratification among Chinese women. J Natl Cancer Inst 102:972–981

26. Barnholtz-Sloan JS, Shetty PB, Guan X, Nyante SJ, Luo J, et al. (2010) FGFR2 and other loci identified in genome-wide association studies are associated with breast cancer in African-American and younger women. Carcinogenesis 31:1417–1423

27. Teraoka SN, Bernstein JL, Reiner AS, Haile RW, Bernstein L, et al. (2011) Single nucleotide polymorphisms associated with risk for contralateral breast cancer in the Women's Environment, Cancer, and Radiation Epidemiology (WECARE) Study. Breast Cancer Res 13:R114

28. Fletcher O, Johnson N, Orr N, Hosking FJ, Gibson LJ, et al. (2011) Novel breast cancer susceptibility locus at 9q31.2: results of a genome-wide association study. J Natl Cancer Inst 103:425–435

29. Campa D, Kaaks R, Le Marchand L, Haiman CA, Travis RC, et al. (2011) Interactions between genetic variants and breast cancer risk factors in the breast and prostate cancer cohort consortium. J Natl Cancer Inst 103:1252–1263

30. Jiang Y, Han J, Liu J, Zhang G, Wang L, et al. (2011) Risk of genome-wide association study newly identified genetic variants for breast cancer in Chinese women of Heilongjiang Province. Breast Cancer Res Treat 128:251–257

31. Li J, Humphreys K, Heikkinen T, Aittomäki K, Blomqvist C, et al. (2011) A combined analysis of genome-wide association studies in breast cancer. Breast Cancer Res Treat 126:717–727

32. Chen F, Chen GK, Millikan RC, John EM, Ambrosone CB, et al. (2011) Fine-mapping of breast cancer susceptibility loci characterizes genetic risk in African Americans. Hum Mol Genet 20:4491–4503

33. Slattery ML, Baumgartner KB, Giuliano AR, Byers T, Herrick JS, et al. (2011) Replication of five GWAS-identified loci and breast cancer risk among Hispanic and non-Hispanic white women living in the Southwestern United States. Breast Cancer Res Treat 129:531–539

34. Stevens KN, Vachon CM, Lee AM, Slager S, Lesnick T, et al. (2011) Common breast cancer susceptibility loci are associated with triple-negative breast cancer. Cancer Res 71:6240–6429

35. Hutter CM, Young AM, Ochs-Balcom HM, Carty CL, Wang T, et al. (2011) Replication of breast cancer GWAS susceptibility loci in the Women's Health Initiative African American SHARe Study. Cancer Epidemiol Biomarkers Prev 20:1950–1959

36. Dai J, Hu Z, Jiang Y, Shen H, Dong J, et al. (2012) Breast cancer risk assessment with five independent genetic variants and two risk factors in Chinese women. Breast Cancer Res 14:R17

37. He C, Chasman DI, Dreyfus J, Hwang SJ, Ruiter R, et al. (2012) Reproductive aging-associated common genetic variants and the risk of breast cancer. Breast Cancer Res 14:R54

38. Shan J, Mahfoudh W, Dsouza SP, Hassen E, Bouaouina N, et al. (2012) Genome-Wide Association Studies (GWAS) breast cancer susceptibility loci in Arabs: susceptibility and prognostic implications in Tunisians. Breast Cancer Res Treat 135:715–724

39. Kim HC, Lee JY, Sung H, Choi JY, Park SK, et al. (2012) A genome-wide association study identifies a breast cancer risk variant in ERBB4 at 2q34: results from the Seoul Breast Cancer Study. Breast Cancer Res 14:R56

40. Huo D, Zheng Y, Ogundiran TO, Adebamowo C, Nathanson KL, et al. (2012) Evaluation of 19 susceptibility loci of breast cancer in women of African ancestry. Carcinogenesis 33:835–840

41. Lin CY, Ho CM, Bau DT, Yang SF, Liu SH, et al. (2012) Evaluation of breast cancer susceptibility loci on 2q35, 3p24, 17q23 and FGFR2 genes in Taiwanese women with breast cancer. Anticancer Res 32:475–482

42. Harlid S, Ivarsson MI, Butt S, Grzybowska E, Eyfjörd JE, et al. (2012) Combined effect of low-penetrant SNPs on breast cancer risk. Br J Cancer 106:389–396

43. Sueta A, Ito H, Kawase T, Hirose K, Hosono S, et al. (2012) A genetic risk predictor for breast cancer using a combination of low-penetrance polymorphisms in a Japanese population. Breast Cancer Res Treat 132:711–721

44. Rinella ES, Shao Y, Yackowski L, Pramanik S, Oratz R, et al. (2013) Genetic variants associated with breast cancer risk for Ashkenazi Jewish women with strong family histories but no identifiable BRCA1/2 mutation. Hum Genet doi: 10.1007/s00439-013-1269-4

45. Parkin DM, Bray F, Ferlay J, Pisani P (2005) Global cancer statistics, 2002. CA Cancer J Clin 55:74–108

46. Garcia-Closas M, Hall P, Nevanlinna H, Pooley K, Morrison J, et al. (2008) Heterogeneity of breast cancer associations with five susceptibility loci by clinical and pathological characteristics. PLoS Genet 4:e1000054

47. Stacey SN, Manolescu A, Sulem P, Thorlacius S, Gudjonsson SA, et al. (2008) Common variants on chromosome 5p12 confer susceptibility to estrogen receptor-positive breast cancer. Nat Genet 40:703–706

48. Nielsen TO, Hsu FD, Jensen K, Cheang M, Karaca G, et al. (2004) Immunohistochemical and clinical characterization of the basal-like subtype of invasive breast carcinoma. Clin Cancer Res 10:5367–5374

49. Yang XR, Pfeiffer RM, Garcia-Closas M, Rimm DL, Lissowska J, et al. (2007) Hormonal markers in breast cancer: coexpression, relationship with pathologic characteristics, and risk factor associations in a population-based study. Cancer Res 67:10608–10617

50. Peng SH, Lu BJ, Ruan WJ, Zhu YM, Sheng HQ, et al. (2011) Genetic polymorphisms and breast cancer risk: evidence from meta-analyses, pooled analyses, and genome-wide association studies. Breast Cancer Res Treat 127:309–324