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Evaluation of associations between common variation in mitotic regulatory pathways and risk of overall and high grade breast cancer

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Abstract

Mitotic regulatory pathways ensure proper timing of mitotic entry, sister chromatid cohesion and separation, and cytokinesis. Disruption of this process results in inappropriate chromosome segregation and aneuploidy and appears to contribute to cancer. Specifically, dysregulation and somatic mutation of mitotic regulators has been observed in human cancers, and overexpression of mitotic regulators is common in aggressive and late stage tumors. However, the role of germline variation in mitotic pathways and risk of cancer is not well understood. We tested 1,084 haplotype-tagging and functional variants from 164 genes in mitotic regulatory pathways in 791 Caucasian women with breast cancer and 843 healthy controls for association with risk of overall and high grade breast cancer. Sixty-one single nucleotide polymorphisms (SNPs) from 40 genes were associated ($p < 0.05$) with risk of breast cancer in a log-additive model. In addition 60 SNPs were associated ($p < 0.05$) with risk of high grade breast cancer. However, none of these associations were significant after Bonferroni correction for multiple testing. In gene-level analyses, *CDC25C*, *SCC1/RAD21*, *TLK2*, and *SMC6L1* were associated ($p < 0.05$) with overall breast cancer risk, *CDC6*, *CDC27*, *SUMO3*, *RASSF1*, *KIF2*, and *CDC14A* were associated with high grade breast cancer risk, and *EIF3S10* and *CDC25A* were associated with both. Further investigation in breast and other cancers are needed to understand the influence of inherited variation in mitotic genes on tumor grade and cancer risk.

Keywords

breast cancer; genetics; mitotic; grade

Introduction

Aneuploidy is a common event in breast cancer, occurring in approximately 80% of all breast tumors [1], where it may play a role in tumor progression [2]. Whole chromosomal instability has also been detected as one of the earliest genetic alterations in premalignant lesions. One mechanism of chromosomal instability, and ultimately aneuploidy, is thought to be the abrogation of proper chromosome segregation during mitosis [3]. This process is regulated by checkpoints that ensure the maintenance of sister chromatid cohesion during

mitotic entry and progression, the timing of spindle assembly and attachment to centromeres, the fidelity of sister chromatid separation during the metaphase to anaphase transition and the appropriate cleavage of daughter cells during cytokinesis [4].

Multiple lines of evidence suggest a causal role for genetic mutations in these mitotic regulatory pathways in the development of chromosomal instability and cancer. Somatic mutations have been identified in genes involved in sister chromatid cohesion and in spindle assembly checkpoint genes in colon, breast and gastric cancer cell lines [4]. Mice with hypomorphic mutations in spindle assembly checkpoint genes such as *Mad2*, *Mad1*, *Bub1*, *BubR1*, and *Bub3* display elevated chromosome missegregation rates, and both *Mad2* and *BubR1* hypomorphic mice display increased rates of tumor development [3,5]. In addition, overexpression of numerous mitotic regulators has been observed in many aggressive and late stage tumors, suggesting that disrupted mitosis contributes to tumor progression. Evidence that overexpression of *Mad2* in transgenic mice promoted tumor formation confirmed this association [6]. Furthermore, recent studies have shown that delay or inhibition of cytokinesis by spindle poisons resulting in polyploidy cells can enhance tumor formation [7].

While the presence of somatic mutations in mitotic genes is well established, the influence of germline variation on risk of cancer has been less extensively investigated. However, a number of associations have been identified including those between SNPs in *LZTS1* and prostate cancer, *MAD1L1* and *MAD2L1* and lung cancer, *STK15* and uterine cancer [8–10], *NEK10* and breast cancer [11], and *AURKA* and several tumor types [12]. In addition, a promoter mutation in *BIRC5* has been consistently associated with risk of colorectal cancer, lung cancer, and urothelial cancer [13,14].

We hypothesized that defects in the genes involved in mitosis including mitotic entry and progression, the spindle assembly checkpoint, and cytokinesis lead to an increased risk of breast cancer. We further postulated that variation in mitotic regulatory pathways is associated with high grade breast cancer risk given the relationship between histologic grade and mitotic index. To test these hypotheses, we investigated genes in these mitotic pathways using a haplotype-tagging approach to measure the association between mitotic genes and risk of overall and high grade breast cancer.

Materials and Methods

Mayo Clinic study subjects

The details of the Mayo Clinic breast cancer case-control study have previously been published [15]. Briefly, subjects were enrolled between February 1, 2001 and June 30, 2005. A total of 798 cases and 843 controls were ascertained at the time of genotyping. Cases were comprised of Caucasian women with invasive breast cancer diagnosed within 6 months of ascertainment with no prior history of cancer other than non-melanoma skin cancer. Controls were comprised of Caucasian women visiting the Mayo Clinic for general medical exams in the Department of Internal Medicine with no prior history of cancer. Participants provided written informed consent and a blood sample as a source of DNA.

Candidate Gene and SNP Selection

Genes that have been implicated in the literature in normal control of mitotic entry, the spindle assembly checkpoint, and cytokinesis were selected for this analysis. For each gene of interest, haplotype-tagging SNPs (htSNPs) were selected based on $r^2 > 0.80$ in publicly available European-American genotype data from HapMap release 21. SNPs with a minor allele frequency (MAF) > 0.05 located within 5kb upstream and downstream of the largest cDNA isoform were included (genome Build 35). Candidate functional SNPs meeting the

same criteria were also included. A total of 1,084 SNPs from 164 genes were successfully genotyped.

Histologic Grade

Histologic grade was determined using the Nottingham combined grading system. Tumors were classified as grade I (low), II (intermediate), and III (high), and only grade III (high) tumors were used in the high grade analyses.

Genotyping

A total of 1,741 samples (798 cases, 843 controls, and 100 duplicates) were genotyped on custom OPAs at Illumina Corporation (San Diego, CA, USA) using the Illumina GoldenGate assay. Only samples with call rates >95% were included in this analysis. Concordance between duplicate samples was 100%.

Statistical Analysis

The genotype data were used to determine allele frequencies in cases and controls separately. Departure from Hardy-Weinberg equilibrium (HWE) was assessed in controls using either a goodness of fit or exact test as appropriate. Single SNP analyses were conducted using unconditional logistic regression adjusting for age and region of residence in all models. In our primary analysis, we tested each SNP in a log-additive (ordinal) model with 1 degree of freedom. To obtain additional information, we also used a 2 degree of freedom test, calculating odds ratios and 95% confidence intervals separately for heterozygotes and rare homozygotes. Significance of association for each gene was estimated using a likelihood ratio test, where the full model contained all principal components that captured > 90% variance. Bonferroni correction was applied to adjust for multiple testing based on 164 genes and 1,084 SNPs.

Results

To address the hypothesis that inherited variation in mitotic signaling pathways influences risk of breast cancer, we tested 1,084 haplotype-tagging and functional variants from genes implicated in the regulation of mitotic entry (n=50), mitotic integrity and the mitotic checkpoint (n=62), and cytokinesis (n=35) (Supplementary Table 1). Sixty-one SNPs from 40 genes were associated with risk of breast cancer in a log-additive model ($p<0.05$) (Table 1, Supplementary Table 2). *NEK1*, *EIF3S10*, *HEI10*, *CCNB1*, *IP1*, *SMC6L1*, *FBXW7*, *RASGEF1A*, *CDC27*, *TLK2*, *CLIP170*, *AJUBA*, *PRC1*, and *CDC37L1* each contained multiple variants associated with breast cancer risk. To assess whether heterogeneity among histological tumor subtypes within the case population affected our results, we performed a subset analysis for estrogen receptor positive (ER-positive) breast cancers, which account for 83.9% of all cases. Results were very similar compared to those from the overall analysis, with 57 SNPs in 35 different genes significantly associated with risk of ER-positive breast cancer ($p<0.05$) (Supplementary Table 3). Thirty of these 35 genes (85.7%) contained SNPs that also had $p<0.05$ in the overall analysis. However, none of the SNPs associated with overall breast cancer or ER-positive breast cancer retained statistical significance after Bonferroni correction for 1,084 comparisons.

Histologic grading in breast cancer is comprised of the evaluation of three features: tubule formation, nuclear pleomorphism, and mitotic count. Tumor grade is associated with tumor aggressiveness and in part reflects the degree to which tumors retain normal mitotic control. High grade breast cancers are associated with poor prognosis compared to lower grade breast cancers, with mitotic index being the most important prognostic factor among the three features comprising histologic grade [16]. To determine whether genetic variation in

mitotic genes is associated with high grade breast cancer, we performed a subset analysis comparing women with high grade breast cancers (n=206) to normal controls (n=843). Sixty SNPs from 34 genes were associated with an increased risk of high grade breast cancer ($p<0.05$) (Table 2, Supplementary Table 4). Twenty-one of these variants were also associated with overall breast cancer, and the directions of association were consistent in both analyses. Additionally, the magnitude of effect was stronger for 19 of the SNPs in the high grade analysis compared to the overall analysis. However, as in the overall analysis, no SNPs were significant after Bonferroni correction for 1,084 comparisons. Three genes- *CDC27*, *ROCK1*, and *SEPT6* contained six SNPs that were associated with high grade breast cancer risk but not overall breast cancer risk. An additional 17 genes represented by 24 SNPs were exclusively associated with high grade breast cancer risk. Fifteen of these genes are involved in regulation of mitotic entry or the mitotic checkpoint.

We next performed an exploratory analysis of breast cancer risk at the gene level using principal components analysis to identify genes associated with breast cancer risk as well as genes not captured by single-SNP analyses alone (Table 3). *CDC25A*, *CDC25C*, *EIF3S10*, *SCC1/RAD21*, *TLK2*, and *SMC6L1* were associated ($p<0.05$) with overall risk of breast cancer. After Bonferroni correction for 164 gene tests, none of these associations retained statistical significance. Only two of the six genes associated with risk in the overall analysis, *EIF3S10* and *CDC25A*, were also associated with risk of high grade breast cancer. *CDC6*, *CDC27*, *SUMO3*, *RASSF1*, *KIF2*, and *CDC14A* were associated with high grade breast cancer only.

Discussion

Improper chromosome segregation during mitosis can lead to aneuploidy, which promotes tumor development and progression. To better understand the contribution of mitotic genes to cancer predisposition we investigated whether inherited genetic variation in mitotic genes contributes to the development of breast cancer. Although our analysis of tagging SNPs from genes involved in mitotic entry, the mitotic checkpoint, and cytokinesis did not identify significant associations when accounting for multiple testing, exploratory analyses revealed interesting patterns of association among the genes in this biologically relevant pathway, particularly with respect to histologic grade of breast cancer. Forty genes were associated ($p<0.05$) with risk of breast cancer in the Mayo Clinic population. When we restricted our analysis to the subset of women with high grade breast cancer, SNPs in 34 genes displayed associations among women with high grade disease. Of these, 20 variants in 12 genes demonstrated larger effects in high grade cancer and an additional 24 variants in 17 genes were not associated with risk of overall disease.

Interestingly, only genes implicated in mitotic entry or the mitotic checkpoint were suggestive of gene-level associations. Four of the 50 mitotic entry genes (8.0%) and 8 of the 64 mitotic checkpoint genes (8.1%) tested in this study were associated with either overall or high grade breast cancer risk at the gene-level. Additionally, several important genes encoding proteins involved in cohesin-removal, a process necessary for the proper separation of sister chromatids during the metaphase-anaphase transition, were represented by SNPs associated with risk of high grade breast cancer: *SMC6*, *SCC1/RAD21*, *ESPL1*, *PTTG1*, and *CDC14A*. In this process, separase (ESPL1) is released from securin (PTTG1) upon activation of the anaphase-promoting complex (APC), and the *SCC1/RAD21* subunit of cohesin is cleaved by separase through a SMC5-SMC6-mediated mechanism [17]. Release of separase in this process also activates CDC14 in a positive-feedback loop, in which CDC14 dephosphorylates securin for continual ubiquitination by the APC [18]. Failure of this mechanism could result in non-disjunction and aneuploidy. Furthermore, while the functional association between the mitotic checkpoint and histologic grade has not

been well studied, a study of invasive ductal breast carcinomas has shown that dysregulation of components of the APC [19] is correlated with higher histologic grade in these tumors.

A strength of this study is that we performed a comprehensive evaluation of variation in mitotic genes and risk of breast cancer. While none of the reported associations retain statistical significance after adjusting for multiple testing, the results of these exploratory analyses warrant further investigation in larger, independent studies of breast cancer. Because of the large number of genes and SNPs assessed in this relatively small sample, we had limited statistical power to detect significant associations. Furthermore, the strong biological rationale used to select these genes suggests that the observed associations, while not statistically significant, may be genuine. In addition, we contribute to the relatively sparse literature on the relationship between genetic variation and breast cancer grade, which has important prognostic implications for women with breast cancer.

In summary, we report the first epidemiological study to comprehensively evaluate genetic variation in genes involved in mitotic regulatory pathways and breast cancer risk. We examined both individual SNP associations and gene-level associations and identified genes that are suggestive of association with risk of both overall and high grade disease. Since aneuploidy and chromosomal instability are commonly observed in human cancers, further investigation of the influence of inherited variation in mitotic genes on tumor grade and cancer risk in additional studies of both breast cancer and other cancer types is merited.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

1. Tirkkonen M, Tanner M, Karhu R, Kallioniemi A, Isola J, Kallioniemi OP. Molecular cytogenetics of primary breast cancer by CGH. *Genes Chromosomes Cancer*. 1998; 21 (3):177–184. [PubMed: 9523192]
2. Mendelin J, Grayson M, Wallis T, Visscher DW. Analysis of chromosome aneuploidy in breast carcinoma progression by using fluorescence in situ hybridization. *Lab Invest*. 1999; 79 (4):387–393. [PubMed: 10211991]
3. Thompson SL, Bakhoum SF, Compton DA. Mechanisms of chromosomal instability. *Curr Biol*. 2010; 20(6):R285–295. [PubMed: 20334839]
4. Molinari M. Cell cycle checkpoints and their inactivation in human cancer. *Cell Prolif*. 2000; 33 (5): 261–274. [PubMed: 11063129]
5. Baker DJ, Jin F, Jeganathan KB, van Deursen JM. Whole chromosome instability caused by Bub1 insufficiency drives tumorigenesis through tumor suppressor gene loss of heterozygosity. *Cancer Cell*. 2009; 16 (6):475–486. [PubMed: 19962666]
6. Sotillo R, Hernando E, Diaz-Rodriguez E, Teruya-Feldstein J, Cordon-Cardo C, Lowe SW, Benezra R. Mad2 overexpression promotes aneuploidy and tumorigenesis in mice. *Cancer Cell*. 2007; 11 (1): 9–23. [PubMed: 17189715]
7. Fujiwara T, Bandi M, Nitta M, Ivanova EV, Bronson RT, Pellman D. Cytokinesis failure generating tetraploids promotes tumorigenesis in p53-null cells. *Nature*. 2005; 437 (7061):1043–1047. [PubMed: 16222300]

8. Hawkins GA, Mychaleckyj JC, Zheng SL, Faith DA, Kelly B, Isaacs SD, Wiley KE, Chang BL, Ewing CM, Bujnovszky P, Bleecker ER, Walsh PC, Meyers DA, Isaacs WB, Xu J. Germline sequence variants of the LZTS1 gene are associated with prostate cancer risk. *Cancer Genet Cytogenet.* 2002; 137 (1):1–7. [PubMed: 12377406]
9. Guo Y, Zhang X, Yang M, Miao X, Shi Y, Yao J, Tan W, Sun T, Zhao D, Yu D, Liu J, Lin D. Functional evaluation of missense variations in the human MAD1L1 and MAD2L1 genes and their impact on susceptibility to lung cancer. *J Med Genet.* 2010; 47 (9):616–622. [PubMed: 20516147]
10. Milam MR, Gu J, Yang H, Celestino J, Wu W, Horwitz IB, Lacour RA, Westin SN, Gershenson DM, Wu X, Lu KH. STK15 F31I polymorphism is associated with increased uterine cancer risk: a pilot study. *Gynecol Oncol.* 2007; 107 (1):71–74. [PubMed: 17599395]
11. Ahmed S, Thomas G, Ghousaini M, Healey CS, Humphreys MK, Platte R, Morrison J, Maranian M, Pooley KA, Luben R, Eccles D, Evans DG, Fletcher O, Johnson N, dos Santos Silva I, Peto J, Stratton MR, Rahman N, Jacobs K, Prentice R, Anderson GL, Rajkovic A, Curb JD, Ziegler RG, Berg CD, Buys SS, McCarty CA, Feigelson HS, Calle EE, Thun MJ, Diver WR, Bojesen S, Nordestgaard BG, Flyger H, Dork T, Schurmann P, Hillemanns P, Karstens JH, Bogdanova NV, Antonenkova NN, Zalutsky IV, Bermisheva M, Fedorova S, Khusnutdinova E, Kang D, Yoo KY, Noh DY, Ahn SH, Devilee P, van Asperen CJ, Tollenaar RA, Seynaeve C, Garcia-Closas M, Lissowska J, Brinton L, Peplonska B, Nevanlinna H, Heikkinen T, Aittomaki K, Blomqvist C, Hopper JL, Southey MC, Smith L, Spurdle AB, Schmidt MK, Broeks A, van Hien RR, Cornelissen S, Milne RL, Ribas G, Gonzalez-Neira A, Benitez J, Schmutzler RK, Burwinkel B, Bartram CR, Meindl A, Brauch H, Justenhoven C, Hamann U, Chang-Claude J, Hein R, Wang-Gohrke S, Lindblom A, Margolin S, Mannermaa A, Kosma VM, Kataja V, Olson JE, Wang X, Fredericksen Z, Giles GG, Severi G, Baglietto L, English DR, Hankinson SE, Cox DG, Kraft P, Vatten LJ, Hveem K, Kumle M, Sigurdson A, Doody M, Bhatti P, Alexander BH, Hooning MJ, van den Ouweland AM, Oldenburg RA, Schutte M, Hall P, Czene K, Liu J, Li Y, Cox A, Elliott G, Brock I, Reed MW, Shen CY, Yu JC, Hsu GC, Chen ST, Anton-Culver H, Ziogas A, Andrulis IL, Knight JA, Beesley J, Goode EL, Couch F, Chenevix-Trench G, Hoover RN, Ponder BA, Hunter DJ, Pharoah PD, Dunning AM, Chanock SJ, Easton DF. Newly discovered breast cancer susceptibility loci on 3p24 and 17q23.2. *Nat Genet.* 2009; 41 (5):585–590. [PubMed: 19330027]
12. Ewart-Toland A, Dai Q, Gao YT, Nagase H, Dunlop MG, Farrington SM, Barnetson RA, Anton-Culver H, Peel D, Ziogas A, Lin D, Miao X, Sun T, Ostrander EA, Stanford JL, Langlois M, Chan JM, Yuan J, Harris CC, Bowman ED, Clayman GL, Lippman SM, Lee JJ, Zheng W, Balmain A. Aurora-A/STK15 T+91A is a general low penetrance cancer susceptibility gene: a meta-analysis of multiple cancer types. *Carcinogenesis.* 2005; 26 (8):1368–1373. [PubMed: 15802297]
13. Gazouli M, Tzanakis N, Rallis G, Theodoropoulos G, Papaconstantinou I, Kostakis A, Anagnostou NP, Nikiteas N. Survivin -31G/C promoter polymorphism and sporadic colorectal cancer. *Int J Colorectal Dis.* 2009; 24 (2):145–150. [PubMed: 18946675]
14. Jang JS, Kim KM, Kang KH, Choi JE, Lee WK, Kim CH, Kang YM, Kam S, Kim IS, Jun JE, Jung TH, Park JY. Polymorphisms in the survivin gene and the risk of lung cancer. *Lung Cancer.* 2008; 60 (1):31–39. [PubMed: 17961802]
15. Kelemen LE, Wang X, Fredericksen ZS, Pankratz VS, Pharoah PD, Ahmed S, Dunning AM, Easton DF, Vierkant RA, Cerhan JR, Goode EL, Olson JE, Couch FJ. Genetic variation in the chromosome 17q23 amplicon and breast cancer risk. *Cancer Epidemiol Biomarkers Prev.* 2009; 18 (6):1864–1868. [PubMed: 19454617]
16. Ignatiadis M, Sotiriou C. Understanding the molecular basis of histologic grade. *Pathobiology.* 2008; 75 (2):104–111. [PubMed: 18544965]
17. Outwin EA, Irmisch A, Murray JM, O'Connell MJ. Smc5-Smc6-dependent removal of cohesin from mitotic chromosomes. *Mol Cell Biol.* 2009; 29 (16):4363–4375. [PubMed: 19528228]
18. Holt LJ, Krutchinsky AN, Morgan DO. Positive feedback sharpens the anaphase switch. *Nature.* 2008; 454 (7202):353–357. [PubMed: 18552837]
19. Park KH, Choi SE, Eom M, Kang Y. Downregulation of the anaphase-promoting complex (APC)7 in invasive ductal carcinomas of the breast and its clinicopathologic relationships. *Breast Cancer Res.* 2005; 7 (2):R238–247. [PubMed: 15743504]

20. Olson JE, Wang X, Pankratz VS, Fredericksen ZS, Vachon CM, Vierkant RA, Cerhan JR, Couch FJ. Centrosome-related genes, genetic variation, and risk of breast cancer. *Breast Cancer Res Treat.* 2011; 125 (1):221–228. [PubMed: 20508983]
21. Olson JE, Wang X, Goode EL, Pankratz VS, Fredericksen ZS, Vierkant RA, Pharoah PD, Cerhan JR, Couch FJ. Variation in genes required for normal mitosis and risk of breast cancer. *Breast Cancer Res Treat.* 2010; 119 (2):423–430. [PubMed: 19377877]
22. Wang X, Fredericksen ZS, Vierkant RA, Kosel ML, Pankratz VS, Cerhan JR, Justenhoven C, Brauch H, Olson JE, Couch FJ. Association of genetic variation in mitotic kinases with breast cancer risk. *Breast Cancer Res Treat.* 2010; 119 (2):453–462. [PubMed: 19404734]

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Table 1
Single SNP associations with overall risk of breast cancer (n=791 cases, 843 controls) (p<0.01)

Log-additive model							2-d.f. model			
Gene	SNP	MAF	Chromosome	OR	95% CI	P-value	Heterozygous		Homozygous	
							OR	95% CI	OR	95% CI
Mitotic entry										
CDC25C ^a	rs11567998	0.020	5	0.39	(0.20 – 0.74)	0.0044	0.39	(0.20 – 0.74)	NA	NA
EIF3S10 ^b	rs10787899	0.41	10	1.27	(1.11 – 1.46)	0.00065	1.19	(0.95 – 1.49)	1.65	(1.24 – 2.18)
	rs3824830	0.38	10	1.22	(1.06 – 1.41)	0.0050	1.18	(0.95 – 1.46)	1.52	(1.14 – 2.03)
Mitotic checkpoint										
SCC1/RAD21	rs4579555	0.097	8	1.42	(1.15 – 1.77)	0.0014	1.43	(1.12 – 1.83)	1.98	(0.88 – 4.43)
TLK2 ^c	rs733025	0.36	17	0.81	(0.70 – 0.94)	0.0058	0.78	(0.63 – 0.96)	0.69	(0.49 – 0.96)
	rs6504112	0.42	17	0.83	(0.72 – 0.95)	0.0076	0.82	(0.66 – 1.01)	0.69	(0.51 – 0.92)
	rs2245092	0.37	17	0.81	(0.70 – 0.94)	0.0050	0.8	(0.65 – 0.99)	0.66	(0.48 – 0.92)
Cytokinesis										
DOCK7	rs1748197	0.35	1	0.81	(0.70 – 0.94)	0.0060	0.71	(0.58 – 0.87)	0.76	(0.54 – 1.07)
ROCK1 ^d	rs17202375	0.079	18	1.4	(1.10 – 1.79)	0.0066	1.43	(1.10 – 1.85)	1.61	(0.50 – 5.15)

^aOlson, et al. 2010 [20]

^bOlson, et al. 2010 [21]

^cKeleman, et al. 2008 [15]

^dWang, et al. 2010 [22]

Table 2

Single SNP associations with risk of high grade breast cancer (n=206 cases, 843 controls) (p<0.01)

Log-additive model				2-d.f. model				
				Heterozygous		Homozygous		
Gene	SNP	MAF	Chr	OR	95% CI	P-value	OR	95% CI
Mitotic entry								
EIF3S10	rs10787900 *	0.24	10	0.63	(0.47 – 0.84)	0.0015	0.65	(0.46 – 0.92)
Mitotic checkpoint								
CDC14A	rs10875295	0.45	1	1.55	(1.23 – 1.94)	0.00016	1.42	(0.95 – 2.12)
	rs508020 *	0.46	1	1.46	(1.17 – 1.83)	0.00096	1.19	(0.80 – 1.79)
	rs3754131 *	0.40	1	0.73	(0.58 – 0.92)	0.0086	0.70	(0.50 – 0.98)
KATNB1	rs12047007 *	0.36	1	1.35	(1.07 – 1.68)	0.0097	1.20	(0.84 – 1.70)
	rs12708992 *	0.068	16	2.00	(1.37 – 2.94)	0.00037	1.88	(1.25 – 2.82)
	rs12922275 *	0.051	16	1.88	(1.21 – 2.92)	0.00450	1.93	(1.23 – 3.01)
CDC27	rs764792 *	0.40	17	1.43	(1.14 – 1.80)	0.0021	1.40	(0.97 – 2.03)
Cytokinesis								
CDC37L1	rs367147	0.47	9	1.36	(1.09 – 1.70)	0.0075	1.00	(0.67 – 1.48)
TEX14	rs4793954 *	0.29	17	1.36	(1.08 – 1.71)	0.0091	1.38	(0.98 – 1.93)

* SNPs associated with high grade breast cancer only, not significant in overall analysis

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Table 3

Gene level associations with risk of breast cancer (p<0.05)

Gene	Chromosome	No. SNPs genotyped	No. principal components	Overall p-value	High grade p-value
Mitotic entry					
CDC25C	5	10	4	0.0064	0.074
EIF3S10	10	4	3	0.026	0.0086
CDC25A	3	2	2	0.047	0.025
CDC6	17	3	1	0.56	0.026
Mitotic integrity/checkpoint					
SCC1/RAD21	8	6	3	0.022	0.17
TLK2	17	5	2	0.027	0.066
SMC6L1	2	11	5	0.038	0.096
CDC27	17	8	5	0.46	0.0046
SUMO-3	21	7	5	0.10	0.018
RASSF1	3	2	2	0.10	0.036
KIF2A	5	6	4	0.39	0.043
CDC14A	1	19	7	0.69	0.047