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Genetic polymorphisms of diabetes-related genes, their interaction with diabetes status, and breast cancer incidence and mortality: The Long Island Breast Cancer Study Project

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Abstract

To examine 143 diabetes risk single nucleotide polymorphisms (SNPs), identified from genome-wide association studies, in association with breast cancer (BC) incidence and subsequent mortality. A population-based sample of Caucasian women with first primary invasive BC (*n* =

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

CONFLICTS OF INTEREST

None declared.

817) and controls (n = 1021) were interviewed to assess diabetes status. Using the National Death Index, women with BC were followed for >18 years during which 340 deaths occurred (139 BC deaths). Genotyping was done using DNA extracted from blood samples. We used unconditional logistic regression to estimate age-adjusted odds ratios and 95% confidence intervals (CIs) for BC incidence, and Cox regression to estimate age-adjusted hazard ratios and CIs for all-cause and BC-specific mortality. Twelve SNPs were associated with BC risk in additive genotype models, at α = 0.05. The top three significant SNPs included *SLC30A8*- rs4876369 (P = 0.0034), *HHEX*-rs11187146 (P = 0.0086), and *CDKN2A/CDKN2B*-rs1333049 (P = 0.0094). Diabetes status modified the associations between rs4876369 and rs2241745 and BC incidence, on the multiplicative interaction scale. Six SNPs were associated with all-cause (*CDKAL1*-rs981042, P = 0.0032; *HHEX*-rs1111875, P = 0.0361; and *INSR*-rs919275, P = 0.0488) or BC-specific (*CDKN2A/CDKN2B*-rs3218020, P = 0.0225; *CDKAL1*-rs981042, P = 0.0246; and *TCF2/HNF1B*-rs3094508, P = 0.0344) mortality in additive genotype models, at α = 0.05. Genetic polymorphisms that increase the risk of developing diabetes may also increase the risk of developing and dying from BC.

Keywords

breast cancer; diabetes; genetics; incidence; mortality; single nucleotide polymorphisms; survival

1 | INTRODUCTION

Over 11 million women aged ≥18 years were living with diagnosed diabetes in the United Sates (US) in 2015[1]. Diabetes is a known risk factor for heart disease, stroke, nephropathy, and neuropathy and cancers of the liver, pancreas, endometrium, and colon/rectum.[2] Accumulating epidemiologic evidence suggests that diabetes may also increase the risk of developing breast cancer (BC), the most frequently diagnosed cancer among women in the US.[3] A meta-analysis of five case-control and 15 cohort studies reported a 20% increased risk of developing BC among women with diabetes, compared to those without diabetes.[4]

Diabetes and BC are both complex multifactorial diseases that share several common predisposing risk factors, including age, overweight/obesity, alcohol consumption, and physical inactivity.[1,5,6] A number of biological mechanisms that focus on the underlying pathologic characteristics of diabetes have been proposed to explain a potential causal association between diabetes and carcinogenesis.[5] High circulating levels of glucose (ie, hyperglycemia), for example, may facilitate neoplastic proliferation due to high rates of glucose uptake required and adopted by many cancers.[5,7] Furthermore, high circulating levels of insulin (ie, hyperinsulinemia) may promote carcinogenesis directly through insulin receptor-mediated mitogenesis,[5,8,9] and indirectly through the reduced hepatic transcription of the insulin- like growth factor binding protein-1 (IGFBP-1) gene,[10] which results in increased circulating levels of free bioactive IGF-1, a potent mitogen of human breast MCF-7 cells.[11] Insulin resistance is also associated with reduced levels of sex hormone binding globulin (SHBG) and thus increases in bioavailable estrogen, although the directionality of association has not been established.[12]

Given the proliferative effects of insulin on BC cells, hyperinsulinemia is also hypothesized to increase risk of mortality following cancer.[5] To date, few studies[13] have investigated diabetes status in relation to mortality following BC. While studies consistently report a 40–50% increase in risk of all-cause mortality following BC,[13,14] results of BC-specific mortality have been mixed.[15–17] However, diabetes has also been associated with more advanced stage at BC presentation,[18,19] and for those with diabetes, BC treatment tends to be less aggressive and causes more adverse effects.[13,19]

Despite observational studies linking diabetes to BC, a few studies[20–22] have examined a small number (≤40) of diabetes risk single nucleotide polymorphisms (SNPs) in association with risk of incident BC and subsequent mortality. Given that diabetes is strongly influenced by genetic factors,[23] we examined the associations between 143 SNPs in or near 29 genes identified from genome-wide association studies of diabetes risk, and BC incidence and subsequent mortality among participants of a population-based study of BC.

2 | MATERIALS AND METHODS

2.1 | Study population

This study included 1838 Caucasian women from the Long Island Breast Cancer Study Project (LIBCSP), a population-based study initiated as a case-control study and then continued as a follow-up study. The LIBCSP study protocol was approved by the Institutional Review Boards of all participating institutions and written informed consent was obtained from participants prior to data collection.

2.2 | Case-control design

Details of the LIBCSP case-control design have been previously reported.[24] In brief, adult women with a first diagnosis of in situ or invasive BC during August 1, 1996 and July 31, 1997 were identified using a rapid reporting system established for the LIBCSP. Approximately 82.1% (n = 1,508) of eligible women with BC completed a comprehensive 100-min interviewer-administered questionnaire, on average within 3 months of diagnosis (25th percentile = 1.2 months, 75th percentile = 4.0 months). Seventy-three percent of BC participants provided blood samples, of which the majority were collected prior to chemotherapy (77.2%). Medical records were abstracted to obtain information on estrogen and progesterone receptor (ER and PR, respectively) status and first course of treatment. The mean age at BC diagnosis among the women included in this study was 59 (range = 25.1–91.9). The majority of women with BC were post-menopausal at diagnosis (68.1%). Most women were diagnosed with ER-positive (76.5%) or PR-positive (66.6%) BC, or both (61.9%).

Women without BC were residents of the same two Long Island counties who were frequency-matched to the expected distribution of women with BC in 5-year age groups in 1996–1997. Women without BC who were 65 years of age and older were identified by Health Care Finance Administration (HCFA) rosters and those under 65 years of age were identified by random digit dialing. Approximately 62.7% (n = 1556) of eligible women without breast cancer completed the questionnaire and, of these, 73.3% provided blood

samples. The majority of LIBCSP participants were Caucasian (93%) and ranged in age from 20 to 98.

2.3 | Follow-up design

Details of the LIBCSP follow-up design and ascertainment of vital status have also been previously reported.[25] In brief, the National Death Index (NDI), a centralized database of death record information maintained by the National Center for Health Statistics,[26] was used to ascertain date and cause of death for the women with BC. International Statistical Classification of Diseases codes 9/10 174.9 and C-50.9 listed on the death certificate were used to identify BC-related deaths. Follow-up for mortality occurred from the date of diagnosis in 1996 or 1997 until December 31, 2014. The median follow-up was 17.6 years (range = 0.39–18.41 years) during which 340 deaths occurred, 139 of which were from BC.

2.4 | Diabetes status

Diabetes status was assessed by self-report as part of the case–control interview. Participants were asked whether they had ever been told by a physician that they had diabetes, sugar diabetes, or high blood sugar.[15] If participants responded in the affirmative, they were also asked the year in which the doctor first told them they had diabetes and whether they had been prescribed medication for their diabetes. Five women (three with BC and two without BC) were missing information on diabetes status. The prevalence of diabetes was 6.9% among women with BC, and 7.0% among women without BC.

2.5 | SNP selection and genotyping

We selected 158 polymorphisms in or near 29 genes for genotyping (Supplemental Table S1). These diabetes-related genes and SNPs were selected based on meta-analyses of genome wide association studies (GWAS),[27–30] which reported statistically significant associations between each SNP and diabetes risk. Genotyping was done in 2011 at the University of North Carolina at Chapel Hill using the Illumina GoldenGate assay (Illumina, Inc., San Diego, CA) on genomic DNA extracted from mononuclear cells in whole blood. Genome Studio software v. 2011.1 was used to review assay intensity data and genotype cluster images for all SNPs. Blind duplicates of 56 samples were genotyped to verify the reproducibility of genotype calls; concordance between duplicates was greater than 99.4% for all pairs.

2.6 | Statistical analysis

This study was restricted to the 1838 LIBCSP participants (817 women with invasive BC and 1021 women without BC) who self-identified as Caucasian and for whom genotyping data were available. We first tested all 158 SNPs for Hardy-Weinberg equilibrium (HWE) among the women without BC using Proc Allele in SAS/Genetics version 9.4 (SAS Institute Inc., Cary, NC) at $\alpha = 0.05$. Seven SNPs (rs1333040, rs10505312, rs11196199, rs7084875, rs6749108, rs7605725, rs4689382) exhibited significant departure from HWE and were not considered further. We next examined the minor allele frequencies of the remaining 151 SNPs in both women with and without BC; rs11708719 had a minor allele frequency <5% (4.55% in women with BC and 4.80% in women without BC). We examined the remaining

150 SNPs for linkage disequilibrium (LD) using the SNAP database based on HapMap.[31] The following seven SNPs were excluded due to LD: rs10010131, rs2046916, rs4712524, rs6744642, rs6769511, rs7748720, rs7923837. QC procedures resulted in 143 SNPs available for statistical analysis.

We used unconditional logistic regression in SAS software version 9.4 (SAS Institute, Inc., Cary, NC) to estimate age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) for the associations between the 143 SNPs and BC incidence. We used multivariable Cox regression in SAS to estimate age-adjusted hazard ratios (HRs) and 95% CIs of the associations between the 143 SNPs and all-cause and BC-specific mortality. For analyses of mortality, observations were censored at the end of follow-up in 2014, if alive. For analyses using BC-specific mortality as the outcome, non-BC deaths were censored at the time of death. For both BC incidence and mortality, we first examined each SNP using additive genotype models (ie, each variant copy is assumed to add to the expression of the phenotype). SNPs significantly associated with BC incidence or mortality at $\alpha = 0.05$ were subsequently examined using co-dominant genotype models (ie, each variant copy is assumed to have an effect on the phonotype) with common homozygous genotypes defined among the women without breast cancer as the referent group. Although we present results for SNPs associated with BC incidence or mortality at $\alpha = 0.05$, we also compared our results to a Bonferroni corrected α of 0.0003, to account for multiple comparisons given that we examined 143 SNPs.[32] SNPs associated with all-cause or breast cancer-specific mortality were also assessed for the proportional hazards assumption using Kaplan-Meier survival curves and Schoenfeld residuals;[33] there were no violations of the proportional hazards assumption. All logistic and Cox regression models were adjusted for age to account for frequency matching of women without BC to women with BC, but no other covariates, given that few factors are causally associated with genetic variants.[34]

For BC incidence, all-cause mortality, and BC-specific mortality, we created three “risk scores” by summing the “at-risk” alleles significantly associated with each outcome at $\alpha = 0.05$. The “at-risk” allele was defined as the less common (variant) allele, unless the variant was inversely associated with breast cancer in this population, in which case the more common allele was defined as the “at-risk” allele. We categorized the continuous summary scores into tertiles based on the distributions in women without BC.

We examined diabetes status (yes vs no) as an effect modifier of the associations between SNPs found to be significantly associated and BC incidence or mortality. Multiplicative interactions were evaluated using likelihood ratio tests, comparing additive genotype models with diabetes-by-SNP cross-product terms against reduced models without the interaction terms. We did not examine interactions on the additive scale as we did not consider dominant genotype models.

3 | RESULTS

3.1 | Diabetes-related gene SNPs and breast cancer incidence

Twelve SNPs were significantly associated with the risk of developing BC in additive genotype models at $\alpha = 0.05$ (Figure 1), but none were statistically significant at the

Bonferroni-corrected alpha of 0.0003. The most significant SNPs included rs4876369 (P = 0.0034), for which the heterozygous (vs common homozygous) genotype was associated with an OR of BC of 1.41 (95% CI = 1.12–1.76), and rs11187146 (P = 0.0086) and rs1333049 (P = 0.0094), for which the variant (vs common) homozygous genotypes were associated with ORs of BC of 2.06 (95% CI = 1.03–4.10) and 1.40 (95% CI = 1.08–1.82), respectively, in co-dominant genotype models (Table 1). Variant (vs common) homozygous genotypes of rs10830963 (P = 0.0125), rs6794209 (P = 0.0201), rs290494 (P = 0.0412), and rs2241745 (P = 0.0458) were inversely associated with BC incidence (Table 1). The highest (vs lowest) category of the summary score of "at-risk" alleles comprised of the sum of the 12 statistically significant SNPs was associated with a 109% increase in risk of BC (OR = 2.09, 95% CI = 1.64–2.66, P < 0.0001).

Diabetes status modified two SNP-BC incidence associations: rs4876369 was associated with an OR of BC of 1.25 (95% CI = 1.02– 1.53) per allele increase among women without diabetes, and with an OR of BC of 4.30 (95% CI = 1.66–11.17) per allele increase among women with diabetes (P for multiplicative interaction = 0.0150) (Table 2). rs2241745 was associated with an OR of BC of 0.76 (95% CI = 0.61–0.94) per allele increase among women without diabetes, and with an OR of BC of 1.76 (95% CI = 0.86–3.58) per allele increase among women with diabetes (P for multiplicative interaction = 0.0283).

3.2 | Diabetes-related gene SNPs and all-cause and breast cancer-specific mortality

Three SNPs (rs981042, P = 0.0032; rs1111875, P = 0.0361; and rs919275, P = 0.0488) were significantly associated with all-cause mortality in additive genotype models at α = 0.05 (Figure 2), but none were statistically significant at the Bonferroni-corrected alpha of 0.0003. In co-dominant genotype models, the rs981042-CA (vs CC) genotype was associated with worse overall survival in the Kaplan-Meier survival curves (Figure 3) and with an all-cause mortality Cox model HR of 1.48 (95% CI = 1.09–2.01) (Table 3). The variant (vs common) homozygous genotypes of rs1111875 and rs919275 were associated with all-cause mortality Cox model HRs of 0.75 (95% CI = 0.53–1.06) and 0.73 (95% CI = 0.54–0.99), respectively (Table 3); however, in the Kaplan-Meier survival curves, the inverse association was only apparent for rs919275 (Figure 3). The highest (vs lowest) category of summary score of "at-risk" alleles comprised of the sum of the three statistically significant SNPs, was associated with a Cox model HR of all-cause mortality of 1.52 (95% CI = 1.14–2.04, P = 0.0004).

Three SNPs (rs3218020, P = 0.0225; rs981042, P = 0.0246; and rs3094508, P = 0.0344) were significantly associated with BC-specific mortality in additive genotype models (Figure 2). In co-dominant genotype models, the variant (vs common) homozygous genotypes of rs3218020 and rs3094508 were associated with improved BC- specific survival in the Kaplan-Meier survival curves (Figure 3) and with BC-specific mortality Cox model HRs of 0.57 (95% CI = 0.33–1.00) and 0.49 (95% CI = 0.26–0.90), respectively (Table 3). The rs981042- CA (vs CC) genotype was associated with worse BC-specific mortality in the Kaplan-Meier survival curves (Figure 3) and with a BC-specific mortality Cox model HR of 1.66 (95% CI = 1.05–2.61) (Table 3). The highest (vs lowest) category of the summary score of "at-risk" alleles comprised of the sum of the three statistically significant SNPs was

associated with a Cox model HR of BC-specific mortality of 2.26 (95% CI = 1.50–3.42, P = 0.0002).

Diabetes status did not modify the associations between rs981042, rs1111875, and rs919275 and all-cause mortality or between rs3218020, rs981042, and rs3094508 and breast cancer-specific mortality, on the multiplicative scale (Table 4).

4 | DISCUSSION

In this population-based sample, of the 143 diabetes risk variants genotyped, 12 SNPs were associated with the risk of developing BC, three with all-cause mortality, and three with BC-specific mortality, in additive genotype models at an alpha of 0.05, but none were statistically significant at the Bonferroni-corrected alpha of 0.0003. The top three most significant SNPs associated with BC risk included: rs4876369, an intron variant of *SLC30A8* (solute carrier family 30 member 8), which encodes a zinc efflux transporter involved in the accumulation of zinc in intracellular vesicles;[35] rs11187146, an intergene variant in the *IDE-KIF11-HHEX* gene cluster at 10q23.33[36]; and rs1333049, an intergene variant in the cyclin-dependent kinase inhibitor 2A/B (*CDKN2A/B*) locus at 9p21.3, hypothesized to play a pivotal role in the development of cardiovascular disease by altering the dynamics of vascular cell proliferation.[37,38] ORs per allele increase for the 12 statistically significant SNPs ranged from 1.14 to 1.34 for those positively associated with BC and from 0.81 to 0.83 for those inversely associated with BC risk. Furthermore, diabetes status modified the association between two SNPs (*SLC30A*- rs4876369 and *IRS2*-rs2241745) and BC risk. In co-dominant genotype models, the variant homozygous genotypes were associated with ORs ranging from 1.33 to 2.06 for those positively associated and from 0.44 to 0.75 for those inversely associated with breast cancer risk, relative to the common homozygous genotypes. Among the four SNPs inversely associated with breast cancer was rs10830963, an intron of *MTNR1B* (melatonin receptor 1B), which encodes one of two high affinity forms of a receptor for melatonin, a pineal gland hormone that regulates glucose metabolism by affecting circadian insulin secretion.[39] Given that these 12 SNPs consist of intron and intergene variants, their functional impact on BC risk is not clear. These SNPs may be in LD with functionally relevant SNPs not included in our study or they may affect gene function by altering the stability, splicing, or localization of the mRNA.[40]

In a previous large study of type 2 diabetes susceptibility variants and the risk of developing BC in women of European ancestry, one SNP (*TCF7L2*-rs7903146) was positively associated and two (*FTO*- rs9939609 and *PRC1*-rs8042680) were inversely associated with BC risk.[20] In our study, we included rs7903146, an intron variant of *TCF7L2* (transcription factor 7 like 2); however, we found no association between this SNP and either risk of developing BC or mortality after BC. It is possible that our study lacked adequate power to detect this association, given that the study by Zhao et al. reported an OR estimate for the risk of developing BC of 1.04 (95% CI = 1.02– 1.06). A second study of type 2 diabetes risk alleles and BC incidence in Caucasian women reported positive associations for rs5945326 and rs1251809 and inverse associations for rs1111875 and rs10923931.[21] In contrast to the study by Hou et al., which reported an OR of 0.88 (95% CI = 0.78–0.99) for the C allele of

HHEX (hematopoietically expressed homeobox),[41] rs1111875 was not associated with risk of developing BC in our study. The rs1111875- A (vs G) allele was, however, inversely associated with all-cause mortality (HR = 0.84, 95% CI = 0.72–0.99) in our study.

For all-cause mortality, we observed that rs981042, an intron variant of *CDKAL1 (*CDK5 regulatory subunit associated protein 1 like 1), a gene of unknown function,[42] was associated with a HR per allele increase of 1.49. rs1111875, an intergene variant located near *HHEX*,[41] and rs919275, an intron variant of *INSR* (insulin receptor),[43] were associated with HRs of 0.84 and 0.86, respectively. For breast cancer-specific mortality, rs981042 and rs3218020, intron variants of *CDKAL1*[44] and *HNF1B*,[45] respectively, were associated with HRs per allele of 0.74 and 0.77. One previous study examined type 2 diabetes genetic variants in association with BC survival among 6000 Chinese women.[46] The study by Bao et al. examined a gene risk score based on 33 GWAS-identified diabetes risk variants. In their study, there was no association between the gene risk score and subsequent survival among women with breast cancer; however, among women with ER- negative breast cancer, a higher gene risk score was associated with worse overall survival and this association was modified by a history of diabetes.[46] In our study, the majority of women with BC were diagnosed with ER-positive BC, which limited our ability to examine effect modification by ER status. Individually, rs7403531 and rs391300 were positively associated with all-cause mortality in their study.[46] In contrast to our study, in their study rs4430796 was inversely associated with all-cause mortality. rs2028299 and rs1359790, SNPs not included in our study, were also significantly associated with BC recurrence/mortality.[46]

Our study had several strengths, including a larger number of SNPs examined than studies published to date, a genetically homogenous population, and the use of existing resources from a population-based study of BC. Additionally, Mendelian randomization, the random assortment of alleles at the time of gamete formation, minimizes the potential for the association between diabetes risk variants and BC to be confounded by environmental factors.[34] While several larger studies have examined diabetes risk variants in association with breast cancer risk, to our knowledge, ours is the first study to examine diabetes risk variants in association with BC-specific mortality among US Caucasian women. However, our study was limited by the comparatively smaller sample than studies published to date, which may have limited our ability to detect associations between the SNPs examined here and BC risk/mortality. Second, the large number of statistical tests could have resulted in spurious results; however, we used a targeted approach in selecting diabetes variants of interest and also interpreted our results using a Bonferroni corrected threshold. Third, we relied on GWAS published before November 2007, and thus potentially interesting variants identified since then may have been missed. Furthermore, many of the diabetes SNPs we found to be significantly related to BC risk are intron variants; their role in BC carcinogenesis remains to be clarified. Last, in examining effect modification by diabetes status, we relied on self-reported diabetes and were not able to distinguish between type 1 and type 2 diabetes; however, in the LIBCSP, the majority of women who reported taking diabetes medications (85%) listed medications that are used to treat type 2 diabetes[15] and type 2 diabetes is the most common type, accounting for 95% of prevalent cases.[1] Furthermore, both type 1 and type 2 diabetes share common predisposing genetic factors as

well as the metabolic sequalae hypothesized to influence breast carcinogenesis and progression.[47]

5 | CONCLUSION

In summary, genetic polymorphisms that increase the risk of developing diabetes may also increase the risk of developing and dying from BC. Our study helps further clarify the association between diabetes and BC risk and may highlight important biological mechanisms of breast carcinogenesis and progression. The prevalence of diabetes is expected to increase by 54% to more than 54.9 million Americans between 2015 and 2030. Thus, a better understanding of how diabetes impacts breast cancer risk may be important for reducing the high burden of BC in the US.[3]

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Abbreviations:

BC	breast cancer
CDKAL1	CDK5 regulatory subunit associated protein 1 like 1
CDKN2A/B	cyclin-dependent kinase inhibitor 2A/B
CI	confidence interval
ER	estrogen receptor
GWAS	genome wide association study
HCFA	health care finance administration
HHEX	hematopoietically expressed homeobox
HR	hazard ratio
HWE	Hardy-Weinberg equilibrium
IGFBP-1	insulin-like growth factor binding protein-1
INSR	insulin receptor

LIBCSP	Long Island Breast Cancer Study Project
MTNR1B	melatonin receptor 1B
NDI	National Death Index
OR	odds ratio
PR	progesterone receptor
SHBG	sex hormone binding globulin
SLC30A8	solute carrier family 30 member 8
SNP	single nucleotide polymorphism
TCF7L2	transcription factor 7 like 2
US	United States

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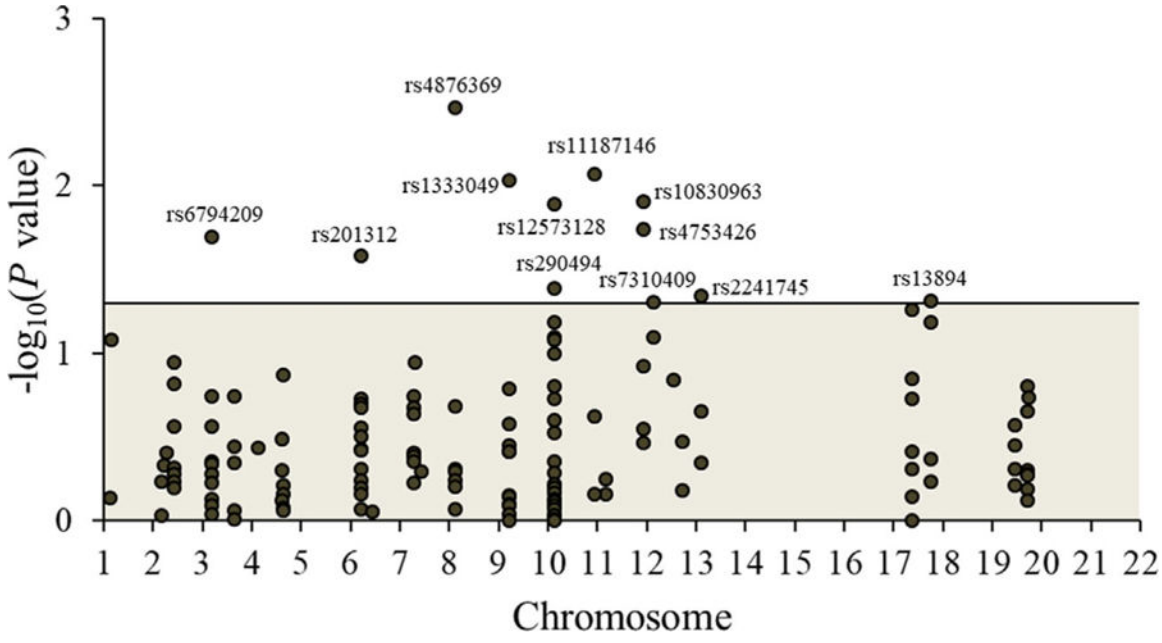

FIGURE 1.

Manhattan plot showing the significance of additive association between all 143 diabetes-related SNPs and *incident breast cancer* among 817 women with invasive breast cancer cases and 1021 age-matched women without breast cancer in the Long Island Breast Cancer Study Project. The 12 SNPs that reached significance at alpha of 0.05 (those above the horizontal line) are listed in Table 1. [Color figure can be viewed at wileyonlinelibrary.com]

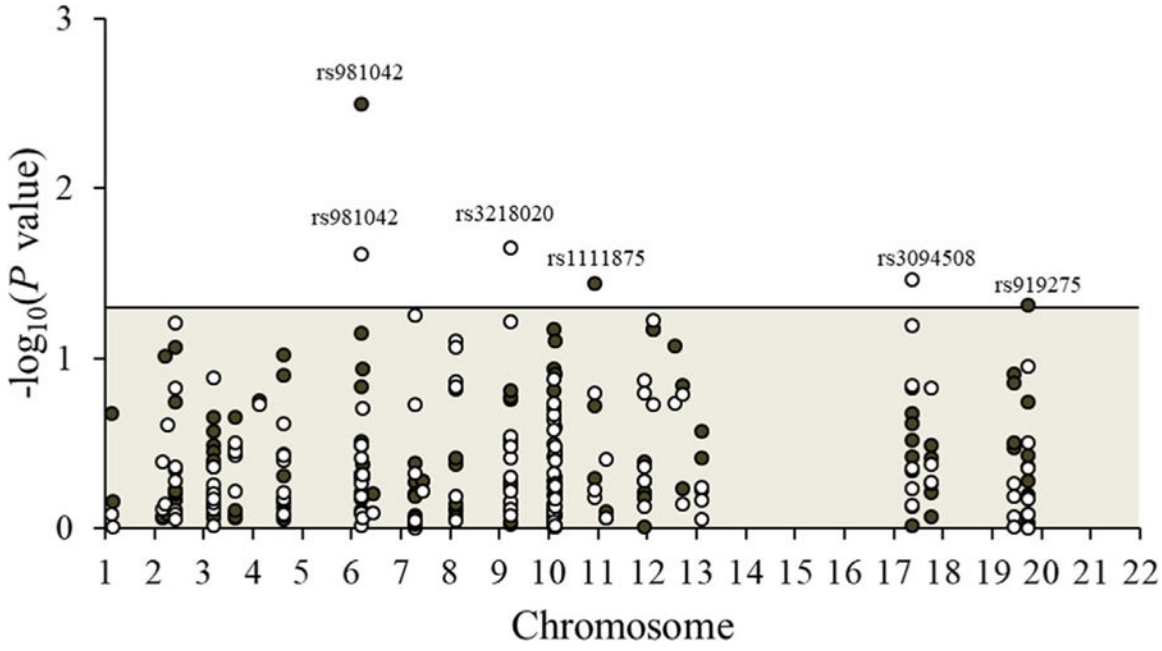

FIGURE 2.

Manhattan plot showing the significance of additive association between all 143 diabetes-related SNPs and *all-cause* (black circles) and *breast cancer-specific* (white circles) *mortality* among 817 women with invasive breast cancer in the Long Island Breast Cancer Study Project. The six SNPs that reached significance at alpha of 0.05 (those above the horizontal line) are listed in Table 3. [Color figure can be viewed at wileyonlinelibrary.com]

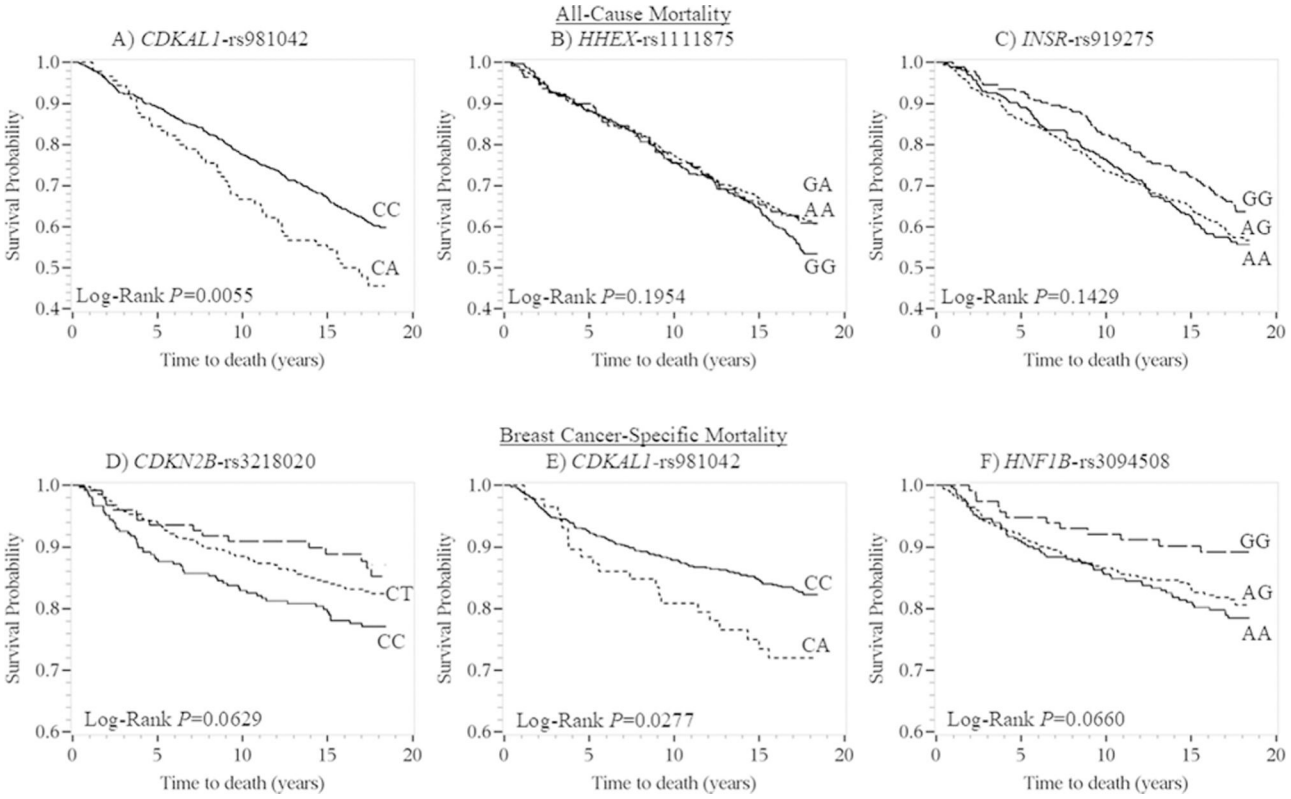

FIGURE 3.

Kaplan-Meier survival plots for all-cause for (A) CDKAL1-rs981042, (B) HHEX-rs1111875, and (C) INSR-rs919275; and breast cancer-specific mortality for (D) CDKN2B-rs3218020, (E) CDKAL1-rs981042, and (F) HNF1B-rs3094508 stratified by genotype among LIBCSP women diagnosed with invasive breast cancer in 1996–1997 (n = 817). The x-axis shows times to death in years; the y-axis shows proportion of participants alive

TABLE 1

Co-dominant genotype model age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) for diabetes-related SNPs and breast cancer (BC) risk in the LIBCSP (n = 1838)

Gene	rs ID location	Genotype	817 BC/1021 no BC	OR (95% CI)	P^a
SLC30A8	rs4876369	AA	588/802	1.00	
	Intron	AG	206/198	1.41 (1.12–1.76)	
		GG	16/16	1.37 (0.68–2.77)	0.0034
HHEX-EXOC6	rs11187146	GG	582/777	1.00	
	Intergene	GC	210/229	1.24 (1.00–1.54)	
		CC	21/14	2.06 (1.03–4.10)	0.0086
CDKN2A-CDKN2B	rs1333049	CC	209/319	1.00	
	Intergene	CG	404/483	1.28 (1.03–1.59)	
		GG	201/216	1.40 (1.08–1.82)	0.0094
MTNR1B	rs10830963	CC	458/507	1.00	
	Intron	CG	301/426	0.78 (0.64–0.95)	
		GG	52/81	0.75 (0.52–1.09)	0.0125
TCF7L2	rs12573128	AA	537/725	1.00	
	Intron	AG	236/263	1.19 (0.97–1.47)	
		GG	35/28	1.74 (1.04–2.91)	0.0129
MTNR1B	rs4753426	CC	173/249	1.00	
	Upstream 2KB	CT	407/528	1.09 (0.86–1.38)	
		TT	231/239	1.37 (1.05–1.79)	0.0184
IGF2BP2	rs6794209	CC	576/683	1.00	
	Intron	CT	221/290	0.91 (0.74–1.12)	
		TT	17/43	0.44 (0.25–0.79)	0.0201
CDKAL1	rs201312	AA	509/681	1.00	
	Intron	AG	262/303	1.17 (0.96–1.43)	
		GG	39/32	1.60 (0.98–2.59)	0.0263
TCF7L2	rs290494	TT	601/714	1.00	
	Intron	TG	192/269	0.86 (0.69–1.07)	
		GG	18/34	0.60 (0.33–1.07)	0.0412

Gene	rs ID location	Genotype	817 BC/1021 no BC	OR (95% CI)	P^a
IRS2	rs2241745	AA	635/755	1.00	
	Intron	AG	168/246	0.81 (0.64–1.01)	
		GG	9/14	0.72 (0.31–1.67)	0.0458
SAT2, SHBG	rs13894	CC	700/912	1.00	
	Intron	CT	111/105	1.36 (1.02–1.81)	
		TT	<5/<5	–	0.0492
HNF1A	rs7310409	GG	258/349	1.00	
	Intron	GA	378/489	1.05 (0.85–1.30)	
		AA	175/177	1.33 (1.02–1.73)	0.0496
Sum of “at-risk” alleles[b]		0–9	172/312	1.00	
		10–11	258/375	1.22 (0.96–1.56)	
		12–24	361/314	2.09 (1.64–2.66)	<0.0001

Long Island Breast Cancer Study Project (LIBCSP) women without breast cancer were age-matched to women diagnosed with breast cancer between August 1, 1996 and July 31, 1997.

[a] Additive genotype model *P* values.

[b] The “at-risk” allele was defined as follows: rs4876369: G allele; rs11187146: C allele; rs1333049: G allele; rs10830963: C allele; rs12573128: G allele; rs4753426: T allele; rs6794209: C allele; rs201312: G allele; rs290494: T allele; rs2241745: A allele; rs13894: A allele; rs7310409: A allele.

TABLE 2

Additive genotype model age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) for diabetes-related SNP genotypes and breast cancer risk, overall and stratified by diabetes status in the LIBCSP (n = 1,833).

				Diabetes status		
			Overall	No (n = 1706)	Yes (n = 127)	
Gene	**rs ID**	**Alleles**	**OR (95% CI)**	**OR (95% CI)**	**OR (95% CI)**	$P_{Interaction}$ [a]
SLC30A8	rs4876369	A/G	1.34 (1.10–1.63)	1.25 (1.02–1.53)	4.30 (1.66–11.17)	0.0150
HHEX-EXOC6	rs11187146	G/C	1.29 (1.07–1.56)	1.36 (1.12–1.66)	0.72 (0.35–1.46)	0.0925
CDKN2A-CDKN2B	rs1333049	C/G	1.19 (1.04–1.35)	1.17 (1.03–1.34)	1.24 (0.75–2.03)	0.7644
MTNR1B	rs10830963	C/G	0.83 (0.71–0.96)	0.80 (0.69–0.94)	1.35 (0.76–2.41)	0.1008
TCF7L2	rs12573128	A/G	1.24 (1.05–1.47)	1.23 (1.03–1.47)	1.28 (0.66–2.48)	0.9562
MTNR1B	rs4753426	C/T	1.17 (1.03–1.34)	1.21 (1.05–1.39)	0.79 (0.48–1.31)	0.1194
IGF2BP2	rs6794209	C/T	0.81 (0.68–0.97)	0.80 (0.66–0.95)	1.00 (0.53–1.89)	0.3237
CDKAL1	rs201312	A/G	1.21 (1.02–1.42)	1.20 (1.01–1.42)	1.19 (0.61–2.30)	0.9777
TCF7L2	rs290494	T/G	0.83 (0.69–0.99)	0.82 (0.68–0.99)	0.88 (0.46–1.70)	0.7418
IRS2	rs2241745	A/G	0.81 (0.66–1.00)	0.76 (0.61–0.94)	1.76 (0.86–3.58)	0.0283
SAT2, SHBG	rs13894	C/T	1.32 (1.00–1.75)	1.32 (0.99–1.76)	1.23 (0.40–3.81)	0.9506
HNF1A	rs7310409	G/A	1.14 (1.00–1.30)	1.14 (0.99–1.31)	1.17 (0.73–1.85)	0.9377
Sum of “at-risk” alleles [b]			1.19 (1.13–1.24)	1.19 (1.14–1.25)	1.06 (0.88–1.28)	0.2253

Long Island Breast Cancer Study Project (LIBCSP) women without breast cancer were age-matched to women diagnosed with breast cancer between August 1, 1996 and July 31, 1997.

[a] Multiplicative interaction P values.

[b] The “at-risk” allele was defined as follows: rs4876369: G allele; rs11187146: C allele; rs1333049: G allele; rs10830963: C allele; rs12573128: G allele; rs4753426: T allele; rs6794209: C allele; rs201312: G allele; rs290494: T allele; rs2241745: A allele; rs13894: A allele; rs7310409: A allele.

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TABLE 3

Co-dominant genotype model age-adjusted hazard ratios (HRs) with 95% confidence intervals (CIs) for diabetes-related SNPs and mortality following breast cancer in the LIBCSP (n = 817).

	rs ID		All-cause mortality (*n* deaths = 340)			
Gene	**Location**	**Genotype**	**Deaths**	**Censored**	**HR (95% CI)**	***P*[a]**
CDKAL1	rs981042	CC	286	434	1.00	
	Intron	CA	49	41	1.48 (1.09–2.01)	
		AA	<5	<5	–	0.0032
HHEX-EXOC6	rs1111875	GG	137	158	1.00	
	Intergene	GA	157	249	0.79 (0.62–0.99)	
		AA	43	67	0.75 (0.53–1.06)	0.0361
INSR	rs919275	AA	112	142	1.00	
	Intron	AG	159	212	0.92 (0.72–1.18)	
		GG	66	117	0.73 (0.54–0.99)	0.0488
Sum of “at-risk” alleles[b]		0–2	159	266	1.00	
		3	112	140	1.27 (1.00–1.62)	
		4–6	63	65	1.52 (1.14–2.04)	0.0004
			Breast cancer-specific mortality (*n* deaths = 139)			
Gene	**rs ID**	**Genotype**	**Deaths**	**Censored**	**HR (95% CI)**	***P*[a]**
CDKN2A, CDKN2B	rs3218020	CC	58	211	1.00	
	Intron	CT	65	344	0.72 (0.51–1.03)	
		TT	16	110	0.57 (0.33–1.00)	0.0225
CDKAL1	rs981042	CC	116	600	1.00	
	Intron	CA	22	66	1.66 (1.05–2.61)	
		AA	<5	<5	–	0.0246
TCF2, HNF1B	rs3094508	AA	62	254	1.00	
	Intron	AG	65	306	0.89 (0.63–1.27)	
		GG	12	106	0.49 (0.26–0.90)	0.0344
Sum of “at-risk” alleles[c]		0–2	52	326	1.00	
		3	47	236	1.25 (0.84–1.85)	

	rs ID		All-cause mortality (*n* deaths = 340)			
Gene	**Location**	**Genotype**	**Deaths**	**Censored**	**HR (95% CI)**	***P*[a]**
		4–6	40	99	2.26 (1.50–3.42)	0.0002

Long Island Breast Cancer Study Project (LIBCSP) women diagnosed with breast cancer between August 1, 1996 and July 31, 1997 and followed-up for vital status through December 31, 2014.

[a] Additive genotype model *P* values.

[b] The “at-risk” allele was defined as follows: rs981042: A allele; rs1111875: G allele; rs919275: A allele.

[c] The “at-risk” allele was defined as follows: rs3218020: C allele; rs981042: A allele; rs3094508: A allele.

TABLE 4

Additive genotype model age-adjusted hazard ratios (HRs) and 95% confidence intervals (CIs) for diabetes-related SNP genotypes and mortality following breast cancer, overall and stratified by diabetes status in the LIBCSP ($n = 817$)

			All-Cause Mortality			
			Diabetes Status			
			Overall	No ($n = 758$)	Yes ($n = 56$)	
Gene	**rs ID**	**Alleles**	**HR (95% CI)**	**HR (95% CI)**	**HR (95% CI)**	$P_{Interaction}$ [a]
CDKAL1	rs981042	C/A	1.49 (1.14–1.94)	1.46 (1.08–1.96)	1.33 (0.75–2.35)	0.9699
HHEX-EXOC6	rs1111875	G/A	0.84 (0.72–0.99)	0.85 (0.72–1.01)	0.90 (0.56–1.44)	0.7751
INSR	rs919275	A/G	0.86 (0.75–1.00)	0.89 (0.76–1.04)	0.61 (0.39–0.97)	0.1618
Sum of "at-risk" alleles [b]			1.21 (1.09–1.34)	1.17 (1.05–1.31)	1.33 (1.01–1.76)	0.3962
			Breast cancer-specific mortality			
			Diabetes Status			
			Overall	No ($n = 758$)	Yes ($n = 56$)	
		Alleles	**HR (95% CI)**	**HR (95% CI)**	**HR (95% CI)**	$P_{Interaction}$ [a]
CDKN2A, CDKN2B	rs3218020	C/T	0.74 (0.58–0.96)	0.76 (0.58–0.99)	0.58 (0.25–1.39)	0.4963
CDKAL1	rs981042	C/A	1.61 (1.06–2.44)	1.58 (1.00–2.48)	1.61 (0.60–4.28)	0.8581
TCF2, HNF1B	rs3094508	A/G	0.77 (0.60–0.98)	0.76 (0.59–0.99)	0.90 (0.41–1.96)	0.6659
Sum of "at-risk" alleles [c]			1.49 (1.21–1.85)	1.37 (1.15–1.64)	1.42 (0.82–2.47)	0.8127

Long Island Breast Cancer Study Project (LIBCSP) women diagnosed with breast cancer between August 1, 1996 and July 31, 1997 and followed-up for vital status through December 31, 2014

[a] Multiplicative interaction *P* values.

[b] The "at-risk" allele was defined as follows: rs981042: A allele; rs1111875: G allele; rs919275: A allele.

[c] The "at-risk" allele was defined as follows: rs3218020: C allele; rs981042: A allele; rs3094508: A allele.