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Common genetic variants in the microRNA biogenesis pathway are not associated with breast cancer risk in Asian women

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Abstract

Background—Although the role of microRNA in cancer development and progression has been well established, the association between genetic variants in microRNA biogenesis pathway genes and breast cancer risk has been yet unclear.

Methods—We analyzed data from two genome-wide association studies conducted in East Asian women including 5,066 cases and 4,337 controls. Among the SNPs which were directly genotyped or imputed, we selected 237 SNPs in 32 genes involved in microRNA biogenesis pathway and its regulation.

Results—Although 8 SNPs were nominally associated with breast cancer risk in combined samples ($P<0.05$), none of them were significant after adjustment for multiple comparisons.

Conclusions—The common genetic variants in microRNA biogenesis pathway genes may not be associated with breast cancer risk.

Impact—This study suggests no association between the polymorphisms in microRNA biogenesis pathway genes and breast cancer risk. Studies with large sample size and more genetic variants should be warranted to adequately evaluate the potential association.

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Conflicts of interests: None

Keywords

breast cancer; genetic susceptibility; microRNA biogenesis pathway; single nucleotide polymorphism

Introduction

MicroRNAs are a major class of non-coding RNAs that post-transcriptionally modulate gene expression in a sequence specific manner. The role of microRNAs in human cancer pathogenesis has been well established by the identification of aberrant expression of microRNAs in many types of cancer (1). There is increasing evidence that the genetic variants in microRNA genes, in their biogenesis pathway genes and binding sites of target mRNA are associated with cancer risk and survival (2). However, the association between the polymorphisms of microRNA biogenesis pathway genes and breast cancer risk is uncertain. The previous study focusing on this pathway was conducted in small sample size and the coverage of genes of interests was limited (3). More recently, the results on the effects of genetic variants in microRNA biogenesis pathway genes were inconsistent even in the same cancer type (4). We comprehensively evaluated the common variants in microRNA biogenesis pathway genes by analyzing data from the two previous genome-wide association studies (GWASs) conducted in women of Asian ancestry consisted of 5,066 breast cancer cases and 4,337 controls.

Materials and Methods

Detailed methods for the Seoul Breast Cancer Study and the Shanghai Breast Cancer Study have been published elsewhere (5, 6) and described in brief in Supplementary Table S1 (3). This study included 5,066 histologically confirmed breast cancer cases and 4,337 controls [(2,190/2,052) Koreans and (2,876/2,285) Chinese]. The protocol was approved by the institutional review board and all participants provided written informed consent.

The thirty five genes (ADAR, ADARB1, DDX5, DDX17, DDX20, DGCR8, DICER1, DROSHA, EIF2C1, EIF2C2, GEMIN4, HNRNPA1, ILF2, ILF3, KHSRP, LIN28A, NANOG, PAPD4, PIWIL1, RAN, SMAD1, SMAD2, SMAD3, SMAD4, SMAD6, SMAD7, SNIP1, SRRT, SRSF1, TARBP,TRIM32, TRIM71, XPO5, XRN2, and *ZCCHC11*) were selected on the basis of their biological role in microRNA biogenesis as determined by the combination of literature review (7) and the bioinformatics tool the Gene Ontology (Supplementary Table S2). Using the data from the HapMap Project and web-based SNP selection tool (8), we selected haplotype tagging SNPs defined by the linkage disequilibrium with the pairwise $r^2 \geq 0.60$, in the region from 10 kb upstream and 5 kb downstream of the largest cDNA of each gene with minor allele frequency (MAF) of ≥ 0.05 in Asian populations. For three genes (*TARBP*, *DDX5* and *SRSF1*) among the 35 genes, no SNP was included because of low MAF or low imputation quality (RSQR < 0.3, *DDX5* rs1991401 and *SRSF1* rs2233911). A total of 247 SNPs (26 genotyped and 221 imputed) located in 32 genes were included. For individual study, odds ratios (OR) and 95% confidence intervals (CI) were estimated using logistic regression implemented in PLINK (genotyped SNPs) and MACH2DAT (imputed SNPs) with adjustment for age. We carried out meta-analysis using

a weighted z-score method under the fixed effect model implemented in METAL (9). Cochran's *Q* statistics were used to assess the heterogeneity across the studies.

Results

Of the 247 SNPs, 8 SNPs (*DDX20* rs3754025, *EIF2C2* rs7834784 and rs6578126, *PIWIL1* rs10848087, *DICER1*rs12432281, *SMAD6* rs17264185 and rs7170982, and *XRN2* rs10485627) were associated with breast cancer risk ($P_{meta} < 0.050$, Table 1). The most significant association was observed in intronic SNP rs7834784 in *EIF2C2* with $OR_{per\text{-}allele}$ as 1.15 (95% CI, 1.02-1.29; $P_{meta} = 0.021$). Study specific estimates of both studies were generally similar. However, these nominally significant associations were not significant when accounting for multiple comparisons. The results of all the SNPs evaluated were presented in Supplementary Table S3.

Discussion

In this study, SNPs in microRNA biogenesis pathway genes were not associated with breast cancer risk. We could not exclude the possibility that the lack of association could be come from the differences in subject characteristics, the interactions of environmental modifiers, and other uncontrolled bias between both studies. We also could not replicate the SNPs shown in previous report (3) although highly correlated proxies were included aside from SNPs in *DDX20*. Considering the sample size of the previous report, it could be a chance finding and at least need to be further investigated in other populations. The strengths of this study are its large sample size and comprehensive search of genes of interests and the high coverage of known SNPs. With the current sample size, for a SNP with MAF as 0.26 (average value of all tested SNPs), the minimum log-additive OR that we can detect at 99% power is 1.22, considering the Bonferroni-corrected significance level (*P* = 1E-04). Future genomic scan including several kinds of structural variations, copy number variants, new variants identified by high-resolution sequencing with larger sample size, as well as subtype specific analysis, could elucidate further information on the potential association.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Table 1

Association between the SNPs with a $P_{meta} < 0.050$ in the genes of interests and breast cancer risk

SNP	Genomic location[a] /Position (bp)[a]	Gene/ Predicted function	Alleles[b]	Seoul Breast Cancer Study (2,190 cases and 2,052 controls)				Shanghai Breast Cancer Study (2,876 cases and 2,285 controls)				Combined (5,066 cases and 4,337 controls)		
				EAF[c]	Per-allele OR (95% CI)[d]	RSQR e	P_{trend}	EAF[c]	Per-allele OR (95% CI)[d]	RSQR e	Ptrend	Per-allele OR (95% CI)	p_{meta}	p_{het} [f]
rs3754025	1p13.2/ 112104760	*DDX20*/ intronic	A/T	0.10	0.94 (0.81-1.08)	1.00	0.37	0.07	0.86 (0.74-1.00)	1.00	0.05	0.90 (0.81-1.00)	0.05	0.45
rs7834784	8q24.3/ 141693661	*EIF2C2*/ intronic	C/T	0.08	1.29 (1.08-1.54)	0.80	0.01	0.09	1.06 (0.91-1.23)	0.80	0.48	1.15 (1.02-1.29)	0.02	0.10
rs6578126	8q24.3/ 141704788	*EIF2C2*/ intronic	A/G	0.06	0.77 (0.64-0.93)	0.99	0.01	0.05	0.98 (0.82-1.16)	0.99	0.81	0.79 (0.77-1.00)	0.04	0.07
rs10848087	12q24.33/ 129405118	*PIWIL1*/ coding	A/G	0.09	0.82 (0.70-0.96)	0.91	0.01	0.10	0.96 (0.84-1.10)	0.90	0.56	0.90 (0.81-0.99)	0.04	0.13
rs12432281	14q32.13/ 94674881	*DICER1*/ 5UTR	A/G	0.30	0.94 (0.83-1.06)	0.58	0.30	0.34	0.89 (0.80-0.99)	0.58	0.03	0.91 (0.84-0.98)	0.02	0.51
rs17264185	15q22.31/ 64784141	*SMAD6*/ intronic	A/G	0.83	1.12 (0.99-1.26)	0.89	0.08	0.85	1.09 (0.97-1.21)	1.00	0.14	1.10 (1.01-1.19)	0.02	0.74
rs7170982	15q22.31/ 64843093	*SMAD6*/ intronic	G/T	0.35	1.09 (1.00-1.20)	0.96	0.06	0.32	1.05 (0.96-1.14)	0.98	0.29	1.07 (1.00-1.14)	0.04	0.52
rs10485627	20p11.22/ 21266977	*XRN2*/ intronic	A/C	0.82	0.88 (0.79-0.99)	1.00	0.03	0.79	0.93 (0.85-1.03)	0.98	0.17	1.10 (0.85-0.98)	0.01	0.45

Abbreviations: EAF, effect allele frequency; OR, odds ratio; CI, confidence interval

[a] Location based on NCBI Human Genome Build 36.3. Hg18

[b] Effect/reference alleles.

[c] Effect allele frequency in controls.

[d] Adjusted for age.

[e] Imputation R-square

[f] P for heterogeneity between both studies was calculated using a Cochran's *Q* test.