Null Results in Brief

No Association between *GPX1* Pro[198]Leu and Breast Cancer Risk

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Introduction

Polymorphisms in the antioxidant selenoprotein glutathione peroxidase (*GPX1*) have been proposed to be associated with lung cancer (1) and breast cancer (2). In particular, the *Leu* allele of the Pro[198]Leu polymorphism has been shown to have lower activity in MCF-7 cell lysates stimulated with sodium selenite (2). This polymorphism was strongly associated with risk of lung cancer (1); a significant association [odds ratio (OR), 1.9; 95% confidence interval (95% CI), 1.02-3.58] in a study of 79 breast cancer cases and 517 controls also has been observed (2). We assessed the association between *GPX1* polymorphisms and breast cancer risk in the prospective Nurses' Health Study.

Materials and Methods

Detailed information regarding the design of this nested case-control study (cases, n = 1,323; controls, n = 1,910) has been published previously (3). In brief, 32,826 women who were free of diagnosed breast cancer were followed for incident disease from time of blood sample collection in 1989 and 1990 up to May 31, 2000. The two single nucleotide polymorphisms studied [−1040 G/A (rs3448) and Pro[198]Leu (rs1050450)] were chosen as haplotype-tagging single nucleotide polymorphisms from the four single nucleotide polymorphisms in the National Cancer Institute's SNP500 database (http://snp500cancer.nci.nih.gov/home.cfm). Briefly, haplotype frequencies from 31 Caucasian samples were generated using the Phase program and haplotype-tagging single nucleotide polymorphisms were chosen using the BEST program (4). *GPX1* genotyping analysis was done by the Taqman Allelic Discrimination method (Applied Biosystems, Foster City, CA) using primers and probe sequences available from the SNP500 database. Genotype-specific ORs and 95% CIs were calculated using unconditional logistic regression and were adjusted for age, age at menopause, postmenopausal hormone use, body mass index at age 18 years, weight gain since age 18 years, benign breast disease, and family history of breast cancer. ORs in relation to tumor stage and size were calculated in cases only, with <4 nodes involved and tumor size ≤2 cm being the reference categories. In these analyses, we compared carriers of the rare allele with noncarriers. Haplotype frequencies in pooled cases and controls were estimated using PROC HAPLOTYPE and linkage disequilibrium was calculated using PROC ALLELE in the SAS System version 8.0 (SAS Institute, Cary, NC). Regression substitution (5) and unconditional logistic regression (adjusted as above) were used for haplotype association testing, with the G-Pro haplotype as the reference category. Power was calculated using STPLAN (http://calculators.stat.ucla.edu/powercalc/).

Results

Both polymorphisms were in Hardy-Weinberg equilibrium, and significant linkage disequilibrium existed between them (r = 0.4, D' = 1.00; P < 0.001). There was no difference in genotype frequency between our control group and that of Hu et al. (P = 0.26, Fisher's exact test). No association was observed between either *GPX1* genotype or the associated haplotypes and breast cancer risk (Table 1). No association was seen between *GPX1* alleles and advanced disease [≥4 involved nodes (n = 81) compared with <4 nodes (n = 967) involved: *−1040A* OR, 1.37; 95% CI, 0.87-2.16; *Leu*[198] OR, 0.75; 95% CI, 0.48-1.19]. Additionally, rare *GPX1* alleles were not significantly overrepresented among larger tumors [>2 cm (n = 245) compared with ≤2 cm (n = 964): *−1040A* OR, 1.09; 95% CI, 0.83-1.44; *Leu*[198] OR, 1.09; 95% CI, 0.83-1.45]. Lastly, no association was seen with any haplotype and tumor size or progression.

Conclusions

We did not observe an association between either the *−1040A* or the *Leu*[198] allele in the *GPX1* gene and breast cancer risk among Caucasian women. We had >99% power to detect a significant relative risk of 1.9 (as reported by Hu et al.) for homozygous carriers of the *Leu* allele compared with women homozygous for the *Pro* allele. It is possible that the association between this allele and breast cancer risk shown by Hu et al. could actually be explained by loss of heterozygosity due to the use of tumor DNA as opposed to peripheral blood from patients in their study. However, no association between

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Table 1. The relationship between *GPX1* genotype and breast cancer risk

	GPX1 genotype, *n* (%)		
	Pro[198]*Pro*	*Pro*[198]*Leu*	*Leu*[198]*Leu*
Cases	581 (47.27)	515 (41.9)	133 (10.82)
Controls	774 (47.51)	694 (42.60)	161 (9.88)
OR (95% CI)*	1.00 (reference)	0.91 (0.77-1.07)	1.07 (0.82-1.40)
	−1040 G/G	*−1040 G/A*	*−1040 A/A*
Cases	678 (54.59)	477 (38.41)	87 (7.00)
Controls	897 (54.63)	640 (38.97)	105 (6.39)
OR (95% CI)*	1.00 (reference)	1.01 (0.85-1.19)	1.15 (0.84-1.59)
	GPX1 haplotype,† *n* (%)		
	G-Pro	*A-Pro*	*G-Leu*
Cases (*n* = 1,293)	0.42	0.26	0.32
Controls (*n* = 1,695)	0.43	0.26	0.31
OR (95% CI)*	1.00 (reference)	1.09 (0.83-1.44)	1.00 (0.77-1.30)

*Estimated by unconditional logistic regression and adjusted for age, age at menopause, postmenopausal hormone use, age at menarche, weight gain since age 18 years, history of benign breast disease, and family history of breast cancer.
† Haplotype frequencies in case and control groups as estimated via PROC HAPLOTYPE, with all haplotypes and risk factors. In the model, the haplotype *A-Leu* was not observed in either group.

the *Leu*[198] allele and tumor progression or size was seen in the present study. Our study is >5 times larger than the study of Hu et al., suggesting that our result is less likely to be influenced by chance fluctuations in the case or control genotype frequency.

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