Null Results in Brief

Transforming Growth Factor β Receptor Type I and Transforming Growth Factor β1 Polymorphisms Are Not Associated with Postmenopausal Breast Cancer

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Introduction

A polymorphic allele in transforming growth factor β receptor 1 (*TGFβR1*) is hypothesized to increase risk of cancer (1). The allele, designated as *TGFβR1*6A*, results from the deletion of three alanines within a nine-alanine stretch in exon 1, and *in vitro* studies have shown that *TGFβR1*6A* is an impaired mediator of TGF-β antiproliferative signals compared with wild-type (*TGFβR1*9A*; refs. 2, 3). Most previous studies (2, 4, 5), including a meta-analysis (6), have found an association between *TGFβR1*6A* and increased risk of breast cancer, but the majority of these studies were hospital based (2, 5, 6). One study (5) further suggested that *TGFβR1*6A* interacts with the *TGFβ1 T29C* polymorphism. The *TGFβ1 29C* allele leads to significantly higher serum levels of TGF-β1 (7, 8), is hypothesized to reduce breast cancer risk (8), and has been examined in many previous breast cancer studies (5, 8-15). We used a nested case-control study within the Cancer Prevention Study II Nutrition Cohort (16) to examine whether the *TGFβR1*6A* allele influenced risk of postmenopausal breast cancer either alone or in combination with *TGFβ1 T29C*.

Materials and Methods

This analysis includes 502 postmenopausal breast cancer cases and 505 controls drawn from a subgroup of the cohort who donated blood samples (n = 21,965 women). Controls were matched on age, race/ethnicity, and date of blood collection; cases and controls had no previous history of cancer (other than nonmelanoma skin cancer). The cohort and case control study are detailed elsewhere (16, 17).

Genotyping was done at Applied Biosystems (Foster City, CA) using DNA purified from buffy coat. The *TGFβ1 T29C* assay was done using TaqMan as previously described (12). The *TGFβR1* assay has also previously been described (1). Following PCR amplification, *TGFβR1* alleles were visualized using an ABI Prism sequence analyzer. A 115-bp peak corresponded to the *9A allele and a 107-bp peak corresponded to the *6A allele. Laboratory personnel were blinded to case-control status and 10% blind duplicates were included in the assays. The concordance rate was 100% for *TGFβ1* and 98% for *TGFβR1*. Overall success rate for the genotyping assays was >96% for both loci and there were no deviations from Hardy-Weinberg equilibrium.

Unconditional logistic regression was used to examine the association between *TGFβ1* and *TGFβR1* and breast cancer while controlling for matching factors. We examined whether the association with the variants of interest differed by stage at diagnosis using general summary stage to classify cases as *in situ*, localized, or regional/distant metastasis. We evaluated whether known breast cancer risk factors were confounders in the logistic models but their inclusion in the models did not appreciably change our effect estimates, and thus we did not include them in the final models.

To examine possible interactions between *6A (*TGFβR1*) and *T29C* (*TGFβ1*) alleles, individuals were classified into low, intermediate, or high TGF-β signalers based on previously published criteria shown in Table 1 (5).

Results

Cases were mostly Caucasian (99%) with median age of 68 years (range, 46-83 years) at diagnosis. Table 1 shows the genotype frequencies, odds ratios (OR), and 95% confidence intervals (95% CI) for the association between breast cancer and the *TGFβR1*6A* and *TGFβ1 T29C* overall and by stage. Because the *TGFβR1*6A* allele is uncommon (allele frequency among controls, 10.7%), we combined women homozygous and heterozygous for the *6A allele and compared them with *9A homozygotes. We found no association with breast cancer and *TGFβR1*6A* (OR, 0.95; 95% CI, 0.69-1.30, for *6A* carriers versus *9A/9A*) or with *TGFβ1 C/C* compared with *T/T* (OR, 0.89; 95% CI, 0.61-1.31), and no differences in analyses stratified by stage.

Because the *TGFβ1 T* allele is associated with lower circulating levels of TGF-β1 than the *C* allele, and the *TGFβR1*6A* allele is a compromised form of wild-type *TGFβR1*, a previous study combined these variants to classify individuals as high, intermediate, or low signalers (5). We found no statistically significant differences across these groups (P_{trend} = 0.70).

Conclusions

We found no evidence of association of breast cancer with *TGFβR1*6A* nor with *TGFβ1 T29C* allele, either alone or in combination. Our study had ~80% power to detect an OR of >1.5 for allele frequencies of 11% as observed for *TGFβR1*6A* among our controls (α = 0.05, β = 0.20). Previous studies have reported ORs in the range of 1.5 to 1.6 (2, 4, 5); thus, we had

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Table 1. ORs and 95% CIs for breast cancer by TGFβR1 and TGFβ1 among all cases and controls and by stage in the Cancer Prevention Study II Nutrition Cohort

Genotype	Cases, % (*N* = 502)	Controls, % (*N* = 505)	OR* (95% CI)	Stage		
				In situ (cases = 109)	Localized (cases = 310)	Regional/distant (cases = 71)
				OR (95% CI)	OR (95% CI)	OR (95% CI)
*TGFβR1*6A*						
9A/9A	387 (77)†	384 (76)	1.00 (—)	1.00 (—)	1.00 (—)	1.00 (—)
9A/6A or 6A/6A	94 (19)	100 (20)	0.95 (0.69-1.30)	0.90 (0.53-1.55)	0.99 (0.69-1.42)	0.84 (0.43-1.63)
TGFβ1 T29C						
T/T	182 (36)	181 (36)	1.00 (—)	1.00 (—)	1.00 (—)	1.00 (—)
T/C	233 (46)	221 (44)	1.05 (0.79-1.38)	1.15 (0.72-1.83)	0.93 (0.68-1.28)	1.65 (0.90-3.01)
C/C	70 (14)	79 (16)	0.89 (0.61-1.31)	1.05 (0.56-1.98)	0.71 (0.45-1.12)	1.51 (0.69-3.29)
			P_{trend} = 0.71	P_{trend} = 0.76	P_{trend} = 0.18	P_{trend} = 0.20
TGFβR1 and TGFβ1 signal‡						
High signalers C/C 9A/9A	55	61	1.00 (—)			
Intermediate signalers C/T 9A/9A, T/T 9A/9A, C/C 9A/6A, and C/C 6A/6A	331	322	1.14 (0.76-1.69)			
Low signalers C/T 9A/6A, T/T 9A/6A, C/T 6A/6A, and T/T 6A/6A	79	79	1.12 (0.69-1.81)			
			P_{trend} = 0.70			

*ORs adjusted for matching factors.
†Percentages do not sum to 100 due to missing genotype data.
‡Classification based on previous study [Kaklamani et al. (5)].

adequate power to observe an association of similar magnitude in our population.

A meta-analysis of *TGFβR1*6A* that included 1,420 cases of breast cancer reported a summary OR of 1.38 (95% CI, 1.14-1.67) among **6A* carriers compared with **9A/*9A* (6). However, there are important methodologic limitations in previous studies included in the meta-analysis. None were population based and most used external controls and did not account for differences in age and ethnicity between case and control groups (2, 4-6, 10).

TGFβ1 and breast cancer has been studied more extensively than *TGFβR1*, and the results have been mixed (5, 8-15). Only one previous study (5) has looked at these two polymorphisms jointly and found increased risk of breast cancer among the group that they defined as low signalers compared with high signalers (OR, 1.69; 95% CI, 1.08-2.66). By their definition, high signalers are those with *TGFβ1* (*C/C*) and *TGFβR1* (*9A/9A*). We did not observe an association between breast cancer and low signaling in our study (OR, 1.12; 95% CI, 0.69-1.81).

In summary, although both the variants examined in this study have been shown to affect TGF-β signaling, we found no evidence that they are associated with postmenopausal breast cancer, either alone or in combination.

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