

Genetic polymorphisms of insulin-like growth factor 1 and insulin-like growth factor binding protein 3, xenoestrogen, phytoestrogen, and premenopausal breast cancer

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ABSTRACT

Background Previous studies suggest a combined effect of insulin-like growth factor 1 (IGF-1) and IGF binding protein 3 (IGFBP-3) gene polymorphisms, xenoestrogen, and phytoestrogen on the IGF-1 signalling pathway and serum concentrations in the IGF system, which are associated with premenopausal breast cancer (BCa) risk.

Methods Between 2010 and 2012, our study recruited 140 premenopausal BCa patients and 160 community-based premenopausal control subjects. Participants were surveyed about oral contraceptive (oc) use, dietary habits, and other BCa risk factors. TaqMan assays were used to determine IGF-1 rs1520220 and IGFBP-3 rs2854744 genotypes. Daily intakes of energy-adjusted soy isoflavones (EASIS) were calculated by the residual method. Multivariate logistic regression was applied to estimate the adjusted odds ratios (ORS) and 95% confidence intervals (CIS) of the IGF-1 rs1520220 and IGFBP-3 rs2854744 genotypes, oc use, and intake of EASIS. Stratified analyses were performed to detect the gene–environment combined effect, and multivariate logistic regression was used to estimate interaction coefficients (IORS) by the multiplicative model, with 95% CIS. The delta method was used to calculate interaction coefficients by the additive model [relative excess risk of interaction (RERI), attributable proportions of interaction (APIS)] and 95% CIS.

Results The IGF-1 and IGFBP-3 genotypes, oc use, and EASIS were not found to be associated with BCa risk (p > 0.05). Stratified analysis showed that the risk of BCa was markedly increased in women carrying the IGFBP-3C allele and using ocs compared with women either carrying the IGFBP-3C allele or using ocs (OR: 3.02; 95% CI: 1.04 to 8.79). The interaction coefficients IOR, RERI, and API were 4.89 (95% CI: 1.09 to 21.90), 2.42 (95% CI: -0.76 to 5.61), and 0.80 (95% CI: 0.46 to 1.67) respectively.

Conclusions The IGFBP-3 rs2854744 polymorphism and oc use might synergistically increase premenopausal Bca risk.

Key Words IGF-1, IGFBP-3, oral contraceptives, isoflavones, breast cancer

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INTRODUCTION

In China, the incidence of breast cancer (BCa) has increased in recent years. The average age of BCa onset in Chinese women is 47 years, which is 10 years younger than the average for Western women¹. Despite the aging of the Chinese population, about two thirds of BCa patients in China are, again in contrast to their Western counterparts, premenopausal at diagnosis¹. Thus, an understanding of the biologic mechanisms of premenopausal BCa and its particular risk factors in China is necessary. Insulin-like growth factor 1 (IGF-1) has long been known to contribute to premenopausal BCa risk^{2,3}. It accelerates cell division and inhibits apoptosis of BCa cells⁴. About 80% of IGF-1 is carried by IGF binding protein 3 (IGFBP-3), which inhibits cell growth and induces apoptosis in several cell lines by sequestration of IGF-1 or a mechanism independent of the mitogenic effects of IGF-1^{5,6}.

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About 38%–60% of the variation in serum levels of IGF-1 and IGFBP-3 is explained by genetics⁷. The G–C substitution in intron 3 of the *IGF1* gene [single nucleotide polymorphism (sNP) rs1520220] might influence circulating IGF-1 expression by altering the secondary structure of RNA or DNA^{8,9}. The A–C substitution at nucleotide –202 in the promoter region of the *IGFBP3* gene (sNP rs2854744) could theoretically result in reduced promoter activity and in turn lower the level of circulating IGFBP-3¹⁰. A number of population studies have observed effects of the foregoing two SNPs on serum IGF-1 and IGFBP-3^{11–14}.

The level of endogenous estrogen has reportedly been positively associated with IGF-1 expression¹⁵. Estradiol might elevate growth hormone–mediated expression of IGF-1 messenger RNA in normal mammary tissue by a factor of 4–6 and elevate IGF-1 bioactivity by downregulating IGFBP-3 synthesis¹⁶. It also plays a key role in the IGF-1 signalling transduction pathways that stimulate cell proliferation¹⁶.

The role of xenoestrogens [such as those used in hormone replacement therapy and oral contraceptives (ocs)] as potential modifiers of IGF-1 and IGFBP-3 levels has been studied. Premenopausal Chinese women have increasingly been using ocs since the one-child policy was enacted in the late 1970s, and ocs have become the main source of xenoestrogen exposure in premenopausal women. Intake of ocs is reported to raise the expression level and bioactivity of circulating estradiol in diverse ways^{17–19}. However, the effect of oc intake on serum IGF-1and IGFBP-3 expression has been evaluated only in population studies, which have produced inconsistent results^{20–23}.

Soy products, a major source of isoflavone, are a regular part of the diet in East Asia. Isoflavone, a type of phytoestrogen, has been found to play—depending on dose and endogenous estrogen level—both the estrogen and the antiestrogen role^{24,25}. Isoflavone has been demonstrated to be able to induce the IGF-1 signalling pathway *in vitro*, and the inhibition effect was identified as well^{26,27}. Evidence about the impact of dietary soy intake on serum IGF-1 and IGFBP-3 is also conflicting^{28,29}.

Given the foregoing evidence, a possible combined effect of IGF-1 and IGFBP-3 polymorphism, oc use, and isoflavone intake on serum concentrations of the components of the IGF system and on the IGF-1 signalling pathway is hypothesized to affect premenopausal BCa risk. To date, the number of studies that have investigated the combined effects of IGF-1 and IGFBP-3 genetic polymorphism, xenoestrogen, and phytoestrogen exposure is limited. We therefore set out to determine the gene–environment interactions between *IGF1* rs1520220, *IGFBP3* rs2854744, oc use, and soy isoflavone intake with respect to premenopausal BCa risk.

METHODS

Study Subjects

Between October 2010 and July 2012, the study sequentially recruited 140 premenopausal women with a new pathology-diagnosed primary Bca at the Sichuan Cancer Hospital. At the same time, 160 community-based premenopausal women undergoing routine physical examinations at the Chengdu Children and Women's Hospital were randomly selected as control subjects. Patients with metastatic Bca and control subjects with malignancies were excluded from the study. Women with reproductive endocrine disease, those using a subdermal implant or intravaginal contraception, and those with levonorgestrel-releasing intrauterine devices were also excluded.

Information on menopausal status was collected when the patients first visited the doctor or the control subjects underwent physical examination. Premenopausal women were defined as those who had a routine menstrual cycle and who had not undergone oophorectomy or hysterectomy. The study protocol was approved by the Institutional Review Board of Sichuan University. All participants provided written informed consent.

Data Collection

Information on demographic characteristics and BCa risk factors—including age, anthropometric indices (height, weight), reproductive factors (menopausal status, age at menarche, age at first delivery, number of live births, number of abortions, breastfeeding duration), lifestyle factors (smoking, alcohol consumption), and family and personal history of breast disease—was collected using a structured questionnaire. Subjects were also asked about oc use. Any woman who had used oc cumulatively for at least 6 months was considered to be a user.

A semiquantitative dietary questionnaire was used to collect long-term dietary habits (5 years or more). The questions asked about daily intake frequency and serving quantities (grams or millilitres) of 6 common food types from the Chinese Nutrition Society dietary guidelines. One of the types was soy products. The design, measurement methods, and reliability of the questionnaire have previously been assessed and reported in detail³⁰. In a pilot study, we investigated the soy products most commonly used by Sichuan women, including fresh and dried bean curd and soy milk. The daily intake of soy isoflavone was calculated as described in that study³¹. To guard against potential underreporting, the residual method was used to calculate the energy-adjusted daily intake of soy isoflavone (EASI)³¹. The regression model of daily isoflavone intake (y) on total daily energy intake (x) was fitted as y = 5.509 + 1000.004x + e, where e is the non-standardized residual. And the exposure to EASI was divided into high and low groups based on daily intake in the control group (7.84 mg).

Genotype Analysis

Whole blood (5 mL) was collected from participants by venipuncture into EDTA tubes and stored at -20°C. Using TIANAMP BLOOD DNA KITS (Tiangen, Beijing, P.R.C.), genomic DNA was extracted from the whole blood. The IGF-1 rs1520220 and IGFBP-3 rs2854744 genotypes were determined by TaqMan assay (purchased from Applied Biosystems, Foster City, CA, U.S.A.). All assays were performed using an Applied Biosystems 7500 thermal cycler. Duplicate detection was performed for 5% of samples at random, with the observed concordance rate being 100%.

Data Analysis

We used the chi-square test to check Hardy–Weinberg equilibrium for the control subjects. The chi-square test or Student t-test was used to assess intergroup distribution differences for BCa risk factors. Using multivariate logistic regression with adjustments for the potential confounders of age, body mass index, age at menarche, age at first delivery, and breastfeeding, we calculated odds ratios (ORS) with 95% confidence intervals (CIS) for the *IGF1* rs1520220 and *IGFBP3* rs2854744 polymorphisms, oc use, and EASI.

Stratified analyses were used to explore the combined effects of the *IGF1* rs1520220 and *IGFBP3* rs2854744 polymorphisms, oc use, and EASI. The wild-type homozygous *IGF1* or *IGFBP3* genotype combined with low exposure to ocs or EASI was treated as the reference. The adjusted ors and 95% cis for the genetic exposure group (oR_G), the environmental exposure group (oR_E), and the combined exposure group (oR_{GE}) were estimated from multivariate logistic regressions by controlling for the potential confounders already mentioned. The corresponding regression coefficients (β_G , β_E , β_{GE}) were calculated as $\ln(oR_G)$, $\ln(oR_E)$, and $\ln(oR_{GE})$.

The interaction coefficient by the multiplicative model—that is, the interactive odds ratio (IOR)—and its 95% cr were calculated and tested by multivariate logistic regression. The interaction coefficients by the additive model—that is, the relative excess risk of interaction (RERI) and attributable proportion of interaction (API)—with their 95% crs were estimated using the delta method as described by Hosmer and Lemeshow³² and by Assmann *et al.*³³. The calculation was performed using the Microsoft Excel (Redmond, WA, U.S.A.) procedure developed by Assmann *et al.*³³. The other analyses were performed using the SPSS Statistics software application (version 17.0: SPSS, Chicago, IL, U.S.A.). All *p* values were subjected to a two-tailed test with an alpha level of 0.05 for significance testing.

RESULTS

Distribution of BCa Risk Factors in Patients and Control Subjects

Mean age in the patient and control groups was 40.68 \pm 5.77 years and 43.16 \pm 6.94 years respectively. Several risk factors were distributed differently in the groups, including age, body mass index, age at menarche, age at first delivery, number of live births, and breastfeeding duration (\geq 3 months vs. <3 months, *p* < 0.05, Table I).

Association of IGF-1 and IGFBP-3 Genotypes, OC Use, and EASI with BCa

In the control group, the frequencies of the IGF-IC allele and the IGFBP-3C allele were 55.9% and 22.8%. In the control group, both sNPs conformed to Hardy–Weinberg equilibrium (p > 0.05). After adjusting for age, body mass index, age at menarche, age at first delivery, breastfeeding duration, IGF-1 and IGFBP-3 genotypes, and oc use, EASI was not significantly associated with BCa risk (Table II).

Combined Effects of *IGF1* and *IGFBP3* Genotypes, OC Use, and EASI

Stratified analyses were used to determine the combined effects of susceptible IGF-1 and IGFBP-3 genotypes, oc use, and EASI. We observed no combined effects of IGF-1 genetic polymorphism with either oc use or EASI. However, compared with either carrying the *IGFBP3* C allele or using ocs,

the combination of carrying the C allele and using ocs was associated with a markedly increased risk for BCa (OR: 3.02; 95% CI: 1.04 to 8.79; Table III).

IORs for IGFBP3 Genotypes and OC Use

By the multiplicative model (combined effect of *IGFBP3* genetic polymorphism and oc use), the IOR was 4.89 (95% cI: 1.09 to 21.90), which was significant (p < 0.05). A significant additive interaction of *IGFBP3* genetic polymorphism and oc use was also observed, with an API of 0.80 (95% cI: 0.46 to 1.67) and a RERI of 2.42 (95% cI: -0.76 to 5.61; Table III). The interaction coefficients of *IGF1* polymorphism and oc use, *IGF1* polymorphism and eASI, and *IGFBP3* polymorphism and EASI were nonsignificant.

DISCUSSION

In this case–control study, we found that *IGFBP3* A-202C and oc use synergistically increased the risk for premenopausal BCa. Women carrying the C allele who had a history of oc use had a markedly increased risk of BCa; alone, however, neither the *IGFBP3* C allele nor oc use alone had any influence on BCa risk.

Population studies have found that the A allele of rs2854744 is positively associated with circulating IGFBP-3, with a distinct dose-response relationship³⁴⁻³⁶. However, research findings in women carrying the IGF1 rs1520220C allele are inconsistent with respect to whether their serum IGF-1 concentration is relatively high or low^{6,11,12}. Conflicting results have also been reported concerning a possible association between those two SNPS and BCa risk. A study in Europe with 8760 subjects reported that women carrying the Callele of IGF-1 had a risk of Bca that was higher by a factor of 1.41; women carrying the A allele of IGFBP-3 had a risk that was 87% lower¹². A case-control study from Shanghai (2503 women) showed that the IGFBP-3C allele conferred a risk that was higher by a factor of 1.6³⁷. However, several studies, including the present one, found no association of those SNPS with BCa risk^{7,14,35,38}. The negative findings might be a result of limited sample size. This potential association should be further explored with studies having larger sample sizes.

Because China's one-child policy has been strictly enforced since the 1980s, Chinese women born in the 1960s or later are more likely than their counterparts in former generations to have used oc during their childbearing years. According to a national sampling survey of 40,000 women, the proportion of oc use in China between 1988 and 2001 ranged from 2.9% to 6.6% for women 15–49 years of age³⁹. A pooled analysis of 53,297 women with and 100,239 women without Bca from 54 epidemiologic studies found that the risk of Bca was 24% higher in women currently using oc than in those who had never used oc⁴⁰. Recently, a study in African American women reported that oc use was associated with risks for estrogen receptor–positive and –negative Bca that were higher by factors of 1.46 and 1.57 respectively¹⁷.

The mechanism by which oc use might affect Bca risk is complicated. Oral contraceptives consisting of estrogen and progesterone combined or progesterone alone have been found to potentially increase circulating estradiol

TABLE I	Breast ca	ancer (BCa)	risk f	factors	for	cases	and	controls
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Factor	Cases	Controls	Statistic	<i>p</i> Value
Mean age (years)				
At diagnosis	40.68±5.77	43.16±6.94	3.34	0.001 ^a
At menarche	13.91±1.71	13.46±1.35	-2.51	0.013 ^a
At first delivery	22.90±2.69	24.71±2.63	5.80	< 0.001 ^a
Mean body mass index (kg/m²)	22.43±2.69	21.70±2.44	-2.45	0.015 ^a
Intake of soy isoflavones (mg/day) ^b	0.88±0.48	0.87±0.53	-0.17	0.87 ^a
Smoking [<i>n</i> (%)]				
No	139 (99.3)	156 (97.5)	1.22	0.39 ^c
Yes	1 (0.7)	4 (2.5)		
Alcohol drinking [n (%)]				
No	133 (95.0)	149 (93.1)	0.47	0.63 ^d
Yes	7 (5.0)	11 (6.9)		
Oral contraceptive use ^e [<i>n</i> (%)]				
No	114 (81.4)	140 (87.5)	2.12	0.15 ^d
Yes	26 (18.6)	20 (12.5)		
Live births [n (%)]				
<2	100 (71.4)	139 (86.9)	11.00	0.001 ^d
≥2	40 (28.6)	21 (13.1)		
Abortions [n (%)]				
0	27 (19.0)	29 (18.1)	1.56	0.46 ^d
1–2	71 (50.0)	92 (57.5)		
≥3	42 (31.0)	39 (24.4)		
Breastfeeding [n (%)]				
≥3 Months	122 (87.1)	110 (68.8)	14.41	< 0.001 ^d
<3 Months	18 (12.9)	50 (31.2)		
Benign breast disease [<i>n</i> (%)]				
No	112 (80.0)	123 (76.9)	0.43	0.58 ^d
Yes	28 (20.0)	37 (23.1)		
Family history of BCa [n (%)]				
No	136 (97.1)	153 (95.6)	0.49	0.55 ^d
Yes	4 (2.9)	7 (4.4)		

^a By Student t-test.

^b Because of skewed distribution in the case and control groups, log values were used.

^c By Fisher exact test.

^d By chi-square test.

^e Defined as 6 months' cumulative use.

levels^{17–19}. It has been hypothesized that oc use increases the BCa risk because of estrogen- or progesterone-induced proliferation of BCa cells, including interaction with the IGF-1 pathway⁴¹. Despite the insignificant association between ос use and вса risk found in the present study, a combined effect of oc use and IGFBP3 polymorphism was detected. Stratified analysis and traditional logistic regression both showed that, compared with women only carrying the C allele or only using oc, women both carrying the IGFBP3 Callele and using ocs had a significantly higher Bcarisk. It has been widely reported that the IGFBP3 rs2854744 C allele is associated with lower IGFBP-3 expression, which in turn increases the free IGF-1 concentration and enhances its bioactivity¹⁶. The free IGF-1 and estradiol might act synergistically to promote Akt protein synthesis and activity, which is the key means by which the IGF-1 signalling pathway suppresses apoptosis⁴². Estradiol has been found

to work synergistically with IGF-1 to promote proliferation in BCa cell lines, which is blocked by IGF-1 antibody⁴³.

Thus, our results appear to be biologically plausible. We found a strong synergistic effect between oc use and *IGFBP3* polymorphism, with an IOR greater than 4. Because the additive model might be better able to explain the biologic interaction, we also estimated the RERI and API interaction coefficients by the additive model. With statistical significance, the API indicated that the risk for premenopausal BCa declined by 80% after removal of the combined effect.

Soy products are common in Asian diets, in which they are the main source of isoflavone. The protective effect of dietary soy intake against BCa has been demonstrated by a number of studies in Asia. A systematic review from Japan showed that soy intake is associated with a lower risk of female BCa (OR: 0.5-0.67)⁴⁴. A meta-analysis from China

Risk factor	Cases [<i>n</i> (%)]	Controls [<i>n</i> (%)]	OR	95% Cl ^a	
IGF1 rs1520220					
GG ^b	38 (27.2)	36 (22.5)	Ref	erence	
GC	58 (41.4)	69 (43.1)	0.76	0.40 to 1.46	
CC	44 (31.4)	55 (34.4)	0.73	0.37 to 1.43	
GC+CC	102 (72.8)	124 (77.5)	1.34	0.74 to 2.41	
IGFBP3 rs2854744					
AA ^b	85 (60.7)	96 (60.0)	Ref	erence	
CA	46 (32.9)	55 (34.4)	1.10	0.39 to 3.10	
CC	9 (6.4)	9 (5.6)	1.26	0.73 to 2.20	
CC+CA	55 (39.3)	64 (40.0)	1.24	0.73 to 2.09	
Oral contraceptive use					
No	114 (81.4)	140 (87.5)	Reference		
Yes	26 (18.6)	20 (12.5)	1.41	0.69 to 2.87	
Daily EASI (mg) ^c					
Low (<7.84)	80 (57.1)	80 (50.0)	Ref	erence	
High (≥7.84)	60 (42.9)	80 (50.0)	0.88	0.53 to 1.49	

TABLE II	Associations	between	study variables
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а Adjusted for age, body mass index, age at menarche, age at first delivery, number of live births, and breastfeeding.

b Wild-type homozygous.

Categorized as higher or lower than the mean of the intake by control subjects (7.84 mg/day). С

OR = odds ratio; CI = confidence interval; IGF-1 = insulin-like growth factor 1; IGFBP-3 = insulin-like growth factor binding protein 3; EASI = energy-adjusted intake of soy isoflavone.

Risk factor	IGF-1 type	Cases [<i>n</i> (%)]	Controls [<i>n</i> (%)]	OR	95% Cl ^a	IGFBP-3 type	Cases [n (%)]	Controls [<i>n</i> (%)]	OR	95% Cl ^a
Oral contraceptive use										
No	GG ^b	84 (60.0)	110 (68.8)	Ref	erence	AA ^b	75 (53.6)	83 (51.9)	Ref	erence
	GC+CC	30 (21.4)	30 (18.8)	1.31	0.68 to 2.51	CC+CA	10 (7.1)	13 (8.1)	0.66	0.24 to 1.79
Yes	GG	18 (12.9)	14 (8.8)	1.37	0.59 to 3.21	AA	39 (27.9)	57 (35.6)	0.94	0.53 to 1.67
	GC+CC	8 (5.7)	6 (3.8)	1.83	0.55 to 6.11	CC+CA	16 (11.4)	7 (4.4)	3.02	1.04 to 8.79
	IOR ^d : 1.02 (0.21 to 4.78); RERI ^e : 0.15 (–2.63 to 2.66); API ^f : 0.08 (–1.22 to 1.38)			IOR ^d : 4.8	IOR ^d : 4.89 (1.09 to 21.90); RERI ^e : 2.42 (-0.76 to 5.61); API ^f : 0.80 (0.46 to 1.67)					
Daily EASI (mg) ^c										
Low (<7.84)	GG ^b	60 (42.9)	64 (40.0)	Ref	erence	AA ^b	48 (34.3)	44 (27.5)	Ref	erence
	GC+CC	20 (14.3)	16 (10.0)	1.22	0.53 to 2.81	CC+CA	32 (22.9)	36 (22.5)	0.98	0.48 to 1.98
High (≥7.84)	GG	42 (30.0)	60 (37.5)	0.83	0.46 to 1.51	AA	37 (26.4)	52 (32.5)	0.74	0.38 to 1.44
	GC+CC	18 (12.9)	20 (12.5)	1.24	0.55 to 2.80	CC+CA	23 (16.4)	28 (17.5)	1.18	0.54 to 2.59
	IOR ^d : 1.22 (0.38 to 3.98); RERI ^e : 0.19 (–1.16 to 1.54); API ^f : 0.15 (–0.88 to 1.19)				IOR ^d : 1.63 (0.58 to 4.61); RERI ^e : 0.46 (-0.51 to 1.44); API ^f : 0.39 (-0.36 to 1.14)					

TABLE III Combined effects of the study variables

а Adjusted for age, body mass index, age at menarche, age at first delivery, number of live births, and breastfeeding.

b Wild-type homozygous genotype.

С Categorized as higher or lower than the mean of the intake by control subjects (7.84 mg/day).

d

е

 $\begin{aligned} & \mathsf{RR}_{\mathsf{GE}} / (\mathsf{RR}_{\mathsf{G}} \times \mathsf{RR}_{\mathsf{E}}). \\ & \mathsf{RR}_{\mathsf{GE}} - \mathsf{RR}_{\mathsf{G}} - \mathsf{RR}_{\mathsf{E}} + 1. \\ & (\mathsf{RR}_{\mathsf{GE}} - \mathsf{RR}_{\mathsf{G}} - \mathsf{RR}_{\mathsf{E}} + 1) / \mathsf{RR}_{\mathsf{GE}}. \end{aligned}$ f

IGF-1 = insulin-like growth factor 1; OR = odds ratio; CI = confidence interval; IGFBP-3 = insulin-like growth factor binding protein 3; EASI = energyadjusted intake of soy isoflavone; IOR = interaction odds ratio; RR_{GE} = relative risk in the presence of genetic and environmental factors; RR_{G} =relative risk in the presence of genetic factors; RR_E =relative risk in the presence of environmental factors; RERI = relative excess risk of interaction; API =attributable proportion of interaction.

involving 9299 cases and 11,413 controls showed that, in Chinese women, dietary soy intake was associated with a BCa risk that was lower by $35\%^{45}$. The stimulation and inhibition of the IGF-1 signalling pathway by genistein, a major component of isoflavone, have been observed in various studies^{26,27}. An earlier study by our group found that, in carriers of the CC genotype of *IGF1* rs1520220, high soy intake might act to lower serum IGF-1 in women less than 50 years of age⁴⁶. However, in the present analysis, we found no effect of EASI alone or of *IGF1* or *IGFBP3* polymorphism combined with EASI. Further studies with larger sample sizes are needed to further delve into possible combined effects.

Given the limited sample size in the present study, the estimated 95% cis of the interaction items were wide, indicating that the results are unstable. Also, we did not differentiate oc types. Further studies with larger sample sizes and oc type differentiation are needed. However, to our knowledge, ours is the first study to estimate the combined effects of *IGF1* and *IGFBP3* polymorphisms, xenoestrogen, and phytoestrogen on premenopausal BCa risk, and we believe that it provides some clues for research into the associated mechanisms.

CONCLUSIONS

Our results suggest that oc use in the presence of the *IG*-*FBP3* rs2854744 C allele synergistically increases the risk of premenopausal BCa for women in southwestern China.

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CONFLICT OF INTEREST DISCLOSURES

We have read and understood *Current Oncology*'s policy on disclosing conflicts of interest, and we declare that we have none.

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