Published in final edited form as:

Am J Gastroenterol. 2008 February ; 103(2): 360–367.

XRCC2 and XRCC3 Gene Polymorphism and Risk of Pancreatic Cancer

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Abstract

OBJECTIVES—XRCC2 and XRCC3 are key components of the homologous recombination (HR) machinery that repairs DNA double-strand breaks. We hypothesized that the altered HR repair capacity conferred by single nucleotide polymorphisms (SNPs) would modify individual susceptibility to sporadic pancreatic cancer.

METHODS—In a hospital-based case-control study, genomic DNA and exposure information was obtained from 468 patients with pathologically confirmed pancreatic adenocarcinoma and 498 frequency-matched healthy controls at M.D. Anderson Cancer Center during January 2000 to September 2006. Genotypes of XRCC2 31479 G>A (Arg¹⁸⁸His) and XRCC3 17893 A>G and 18067 C>T (Thr²⁴¹Met) were determined using the Masscode technology. Unconditional logistic regression models were used to estimate the odds ratio (OR) and its 95% confidence interval (CI) in non-Hispanic whites (408 cases and 449 controls).

RESULTS—The distribution of genotype frequencies was not different between cases and controls. We observed a significant effect modification between XRCC2 polymorphism and smoking status and pack-year of smoking in modifying pancreatic cancer risk (*P* value for interaction 0.02 and 0.05, respectively). Compared with never-smokers carrying the XRCC2 Arg¹⁸⁸Arg genotype, the OR (95% CI) for individuals carrying the ¹⁸⁸His allele was 2.32 (1.25-4.31) among ever-smokers, 1.43 (0.59-3.48) among light smokers (<22 pack-years), and 3.42 (1.47-7.96) among heavy smokers (≥22 pack-years). The two XRCC3 SNPs are in strong linkage disequilibrium, but there was no suggestive association between XRCC3 genotype and the risk of pancreatic cancer.

CONCLUSION—XRCC2 Arg¹⁸⁸His polymorphism may be one of the genetic modifiers for smoking-related pancreatic cancer.

INTRODUCTION

Pancreatic cancer is the fourth leading cause of cancer-related death in both men and women in the United States. It is a rapidly fatal disease, with the poorest prognosis among all malignancies (1). Adenocarcinoma, which arises from ductal epithelial cells of the exocrine pancreas, is the most common and the most aggressive histological type of pancreatic cancer (2). It is estimated that 5-6% of pancreatic cancer cases can be accounted for by preexisting chronic pancreatitis (3), and 3-5% of cases by high-penetrance genetic predisposition (*e.g.*, germline mutation of the *BRCA2* gene) (4). The etiology of sporadic pancreatic cancer is still

largely unknown. Ample evidence has pointed to cigarette smoking as a risk factor of pancreatic cancer (5). However, very little is known about the effect of low-to-moderate penetrance genetic variants on the development of pancreatic cancer.

A few previous studies have provided evidence showing that genetic variants in DNA repair genes could be risk modifiers in smoking-related pancreatic cancer (6,7), which prompt us to further investigate the genetic variants of different DNA repair pathways in association with pancreatic cancer. Among the diverse DNA damage induced by complex tobacco carcinogens, the double-strand break (DSB) formed by oxygen free radicals is one of the most detrimental. DSB can also originate from normal metabolic processes, such as DNA replication and meiotic chromosome alignment. The unrepaired or misrepaired DSB can introduce mutations in genes that are critical to the regulation of cell growth and can result in cancer development. In DSB repair-deficient cells, several types of chromosome aberrations have been observed, including chromatid and chromosome breaks, exchanges, translocations, and deletions (8,9). Chromosome instability is well documented in the pathogenesis of cancers, including pancreatic cancer (10). Therefore, it is biologically plausible that DSB repair deficiency has a role in pancreatic tumorigenesis.

Homologous recombination (HR) and nonhomologous end joining are two major mechanisms for the repair of DNA DSB, and genes involved in these repair pathways are predicted to have a genomic caretaker's role (8). DNA DSB is a major initiator of HR that restores the continuity of broken DNA by using homologous DNA as a template. The recruitment of an intact copy of DNA requires strand exchange mediated by *RAD51*, which polymerizes onto DNA to form a nucleofilament that searches for homologous DNA (9). X-ray repair cross-complementing (*XRCC*) group 2 and 3 are members of the family of *RAD51*-related proteins that participate in HR. *XRCC2* is a core component and forms a heterodimer with *RAD51*-like protein. High levels of aneuploidy, chromosome rearrangement, and haploinsufficiency have been observed in *XRCC2* gene knockout cells (11). *XRCC3* has a diverse set of functions and acts both early and late in the HR repair pathway (12).

A number of studies have shown significant associations between single nucleotide polymorphisms (SNPs) of DNA repair genes and levels of DNA damage, individual DNA repair capacity, and risk of different types of human cancer (13,14). A relatively rare polymorphism at nucleotide position 31479 (G-to-A substitution, exon 3 Arg¹⁸⁸His, rs#3218536) in the *XRCC2* gene (located at chromosome 7q36.1) was one of these SNPs (15). Two SNPs of the *XRCC3* gene (located at chromosome 14q32.3), *i.e.*, A17893 G (rs#1799796) and C18067 T (rs#861539, a.k.a. Thr²⁴¹Met) (16), have been investigated in association with the risk of other types of cancer (17,18). In the current case-control study, the main *a priori* hypothesis is that three SNPs of the *XRCC2* and *XRCC3* genes modify the risk of pancreatic cancer. Because smoking is a known risk factor for pancreatic cancer and tobacco carcinogens can induce DNA DSB, we investigated whether the SNPs modify the risk of pancreatic cancer, particularly in smokers.

MATERIALS AND METHODS

Study Design

The study design and data collection have been described previously (6). The present study was a hospital-based, frequency-matched case-control study. The matching factors were age (by ± 5 yr), sex, and race. Both cases and controls were recruited from The University of Texas M. D. Anderson Cancer Center during the period between January 2000 and September 2006. All cases and controls were U.S. residents and were able to communicate in English. Written informed consent was obtained from each participant for personal interview and blood samples.

The research protocol was approved by the Institutional Review Board of M. D. Anderson Cancer Center.

Case and Control Accrual

During the course of the study, a total of 787 pancreatic cancer patients were approached and 613 (78%) were enrolled. The common reasons for refusal to participate included the patients being too sick or too upset and time constraints. The eligibility criteria of cases for the current study were: patients with pathologically confirmed primary pancreatic ductal adenocarcinoma (International Classification of Diseases for Oncology code C25.3, World Health Organization, 2000), without prior cancer history except for nonmelanoma cancer of the skin, with blood samples, and with complete exposure information. Accordingly, 468 patients were eligible for the study. The same eligibility criteria, except the cancer diagnosis, were applied in control recruitment. Controls were ascertained from cancer patient companions who were not genetically related to the patients and were seemingly healthy. Individuals accompanying pancreatic cancer patients were excluded from the study. The eligible controls were identified by a brief self-administered questionnaire acquiring demographic information and cancer history. A total of 647 eligible individuals were approached and 498 (77%) consented to participate in the study. Twenty were excluded due to incomplete exposure information.

Among the study participants, 408 of 468 cases and 449 of 498 controls were non-Hispanic whites. We performed risk estimation analysis among non-Hispanic whites and presented genotype frequency for other ethnic groups. The majority (67%) of our study participants were from the state of Texas and surrounding states and the rest of the participants were from other states across the country.

Data Collection

A structured risk factor questionnaire was administered by direct personal interview. The interview usually took 20 minutes and was conducted by personnel specifically trained to administer this instrument. Information collected during the interview included demographic data, state of residence, smoking history, family history of cancer, and medical history including pancreatitis and diabetes.

An individual who had never smoked or had smoked <100 cigarettes in his or her lifetime was defined as a never-smoker. An individual who had smoked at least 100 cigarettes in his or her lifetime was defined as an ever-smoker. Ever-smokers included former and current smokers. Former smokers were defined as those who had quit smoking for >1 yr before recruitment for controls and before cancer diagnosis for cases. Current smokers included recent quitters (within 1 yr). Other smoking-related information included the duration (years smoked), intensity (average cigarettes per day), and if applicable, time of smoking cessation. Cumulative smoking was calculated as pack-years, *i.e.*, the number of packs smoked per day multiplied by the number of years of smoking.

The same interviewers performed interviews for both cases and controls. No proxy interviews were conducted. Blood samples from both cases and controls were collected in heparinized Vacutainers (BD Biosciences, Franklin Lakes, NJ). Consecutively, unique study identification numbers were issued to study participants upon their recruitment. Therefore, the case-control status of the blood samples was unknown to the laboratory personnel who extracted DNA and performed genotyping.

Laboratory Analyses

Peripheral blood mononuclear cells (PBMC) were separated from freshly drawn blood by Ficoll-Hypaque (Amersham Pharmacia Biotech, Piscataway, NJ) density gradient

centrifugation and were stored at -80°C. PBMC DNA was extracted using a FlexiGene DNA kit following the manufacturer's instructions (Qiagen, Valencia, CA), and the aliquot was stored at 4°C for immediate use. DNA was extracted at M.D. Anderson Cancer Center and was successful for all blood specimens. The DNA samples were then shipped to Bioserve Biotechnologies Ltd., Laurel, MD, for genotyping. Genotype of the *XRCC2* 31479 G>A (Arg¹⁸⁸His) and *XRCC3* 17893 A>G and 18067 C>T (Thr²⁴¹Met) polymorphism was determined using the Masscode SNP genotyping technology based on a PCR-based (polymerase chain reaction) allele-specific assay (Bioserve Biotechnologies, Ltd.). For quality control, positive allele controls were run in every experiment and 10% quality control repeats were imbedded. In addition, we included 5% random sample repeats in each batch of samples. The discrepant call rate was <0.1%, mostly caused by allele dropout as a result of poor DNA quality or quantity. The no-call rate was 6.1% for the *XRCC2* SNP (31 controls and 22 cases) and 5.2% for the *XRCC3* 17893 A>G SNP (18 controls and 27 cases). The missing rate of genotype data for minority was high, because we prioritized our analysis in non-Hispanic whites and genotyping for other ethnic groups had not been completed. There were no significant difference between participants who were included and those who were excluded from the final analysis in terms of the distribution of age, sex, and state of residence.

Statistical Analysis

All data analyses were performed using STATA 9.0 software (Stata Corporation, College Station, TX). All tests were 2-tailed, and $P \leq 0.05$ was considered statistically significant. The Pearson's χ^2 test was used to examine the differences in the risk factors and genotype distributions between the cases and controls. Hardy-Weinberg equilibrium was tested by the goodness-of-fit χ^2 test with 1 degree of freedom among cases and controls separately. Risk association analysis was performed in non-Hispanic whites only because the small number of minorities did not allow meaningful analysis in these groups. Genotype-specific risks were estimated as crude odds ratios (ORs) and their associated 95% confidence intervals (CIs) by univariate unconditional logistic regression models. In all estimations, the reference group consisted of individuals who were homozygous for the common alleles. The median values of controls were used as the cutoff to categorize "younger" (<61 yr) and "elderly" (≥ 61 yr) individuals, and "light smokers" (<22 pack-years) and "heavy smokers" (≥ 22 pack-years). To explore the potential gene-environment interaction by smoking status (never- vs ever-smoker) and smoking pack-years (0, <22, and ≥ 22), the cross product term between genotype and the variable of interest was generated in unconditional logistic regression models. A two-by-four table was constructed to evaluate the scale of interaction (*i.e.*, the departure from an additive or multiplicative model) (19). The significance of the interaction term was tested using a likelihood ratio test, with the full model containing the interaction term, the main effect of genotype, and the exposure variable, and the reduced model lacking the interaction term. Trends in the ORs were examined using the score test by treating the interaction term as a continuous variable. Potential confounding factors (pack-years of smoking, diabetes, and family history of cancer) were adjusted in the multivariate model when their removal from the multivariate model caused the β parameter estimates to change by >5%. As none of the variables was identified as a confounding factor, the crude OR is presented in the tables. Pairwise linkage disequilibrium of two alleles of the *XRCC3* gene was tested in the controls using SNPalyze software (Dynacom Co., Ltd., Mobara, Japan) and measured by D' and r^2 . Haplotypes were reconstructed from the genotype data using the PHASE (version 2) program (20). Analyses were stratified by age and smoking status.

Power calculation was based on the main hypothesis and was performed using the logistic regression module of Sample Power software (SPSS, Inc., Chicago, IL). It showed that with a sample size of 857 (48% cases and 52% controls), the study had >80% power to detect an OR of 1.66 for the *XRCC2* and *XRCC3* gene polymorphisms at significance level $\alpha = 0.05$ (2-

sided), assuming a dominant mode of gene inheritance and that the frequency of the variant allele in the control group was 5% for *XRCC2* Arg¹⁸⁸His, 30% for *XRCC3* A17893 G, and 38% for Thr²⁴¹Met of *XRCC3* SNP.

RESULTS

The demographic and risk factors for the 468 cases and 498 controls are described in Table 1. There were no significant differences between cases and controls in terms of the distribution of age, sex, and race as a result of frequency matching. There were significantly more ever-smokers and heavy smokers (≥ 22 pack-years) among cases than among controls (P values ≤ 0.002).

The distribution of genotypes of three polymorphisms was comparable between cases and controls in the three ethnic groups (Table 2). Among non-Hispanic whites, the minor allele frequency was 0.058 for the *XRCC2* ¹⁸⁸His and was 0.30 and 0.39 for the *XRCC3* 17893 G and ²⁴¹Met polymorphism, respectively. All genotype distributions were in Hardy-Weinberg equilibrium among controls (all P values ≥ 0.20). As a small number of individuals carried the *XRCC2* His¹⁸⁸His genotype, the heterozygous and homozygous variant groups were pooled in further analyses. The two *XRCC3* SNPs (A17893 G and C18067 T) were in strong linkage disequilibrium in controls, with $D' = -0.9859$ and $r^2 = 0.279$ ($\chi^2 = 160.3$, $P < 0.001$). Three common haplotypes (AT, AC, and GC) were inferred, with a frequency of 0.39, 0.30, and 0.30, respectively. Haplotype GT was found in two study participants. There was no significant difference in the frequency of haplotype distribution between cases and controls overall ($P = 0.46$).

Table 3 shows a significant effect modification between *XRCC2* Arg¹⁸⁸His polymorphism and smoking exposure in modifying pancreatic cancer risk (P value for interaction = 0.02). Compared with never-smokers carrying the *XRCC2* Arg¹⁸⁸Arg, ever-smokers carrying the same genotype had an OR of 1.39 (95% CI 1.03-1.88, $P = 0.03$), and ever-smokers carrying at least one copy of the His variant allele had an OR of 2.32 (95% CI 1.25-4.31, $P = 0.008$). There was no significant effect modification between *XRCC3* genotype or haplotype and smoking exposure (data not shown).

Furthermore, Table 4 shows that the effect of *XRCC2* polymorphism was more prominent in heavy smokers. Compared with never-smokers carrying the Arg¹⁸⁸Arg, the ¹⁸⁸His specific OR (95% CI, P value) was 0.57 (0.29-1.11, 0.09) for never-smokers, 1.43 (0.59-3.48, 0.42) for light smokers, and 3.42 (1.47-7.96, 0.004) for heavy smokers (P value for trend test was 0.0002 and P value for interaction was 0.05). The effect modification between *XRCC2* polymorphism and the number of pack-years of smoking in modifying risk of pancreatic cancer was observed in both former smokers and current smokers (data not shown).

DISCUSSION

To our knowledge, there has been no report on the association between HR repair gene polymorphisms and the risk of pancreatic cancer. The current study has tested the hypothesis that deficient HR repair capacity conferred by the SNPs of the relevant genes predisposes individuals, with excess carcinogen exposure, to an increased risk of pancreatic cancer. Results from this study showed a significant interactive effect between the *XRCC2* Arg¹⁸⁸His polymorphism and smoking in modulating the risk of pancreatic cancer. This study, therefore, provides further evidence supporting the low-penetrance genetic polymorphisms of DNA repair as a susceptibility factor of smoking-related pancreatic cancer.

The frequencies of variant alleles in our control population are comparable to those reported by other studies. The frequency of the *XRCC2* ¹⁸⁸His allele was 0.058, as compared to 0.06

and 0.07 as reported in two European studies (15,21). The frequency of the *XRCC3* ²⁴¹Met allele was 0.39, which is similar to that reported in U.S. whites (13). It indicates that the use of convenient samples in this hospital-based study is unlikely to introduce substantial bias in estimating the genotype-specific OR.

Previous studies have shown that *XRCC2* ¹⁸⁸His is associated with a significantly increased risk of pharyngeal cancer (22) and breast cancer (15), but not with bladder cancer (23), colorectal adenoma (24), and skin cancer (25). The *XRCC2* Arg¹⁸⁸His polymorphism results in a conservative amino acid change that does not lie in a known functional domain (15). However, a subtle effect of reduced cell survival after mitomycin-C exposure has been observed in cells with the variant allele of this SNP (21). Our study findings support the hypothesis that subtle variation in *XRCC2*-involved HR repair may influence susceptibility to pancreatic cancer in people with significant exposure to cigarette carcinogens. DNA DSB induced by reactive oxygen species can be one of the detrimental effects that cigarette smoking imposes on the genome. Heavy smokers carrying the *XRCC2* ¹⁸⁸His variant allele were at a more than twofold higher risk than never-smokers who carried the homozygous wild-type allele.

Although intronic SNPs are unlikely to have a direct functional role, they may affect RNA splicing, and thus the transcription of the gene (26). The *XRCC3* A17893 G polymorphism resides in the intron region, and the functional significance of this SNP has not been investigated. Two previous studies have reported the main effect of the *XRCC3* 17893 G allele in modestly reducing the risk of lung cancer and breast cancer (15,27). Our study provided no evidence of any association between this SNP and the risk of pancreatic cancer.

The *XRCC3* 17893 A allele is in strong linkage disequilibrium with the 18067 T allele (²⁴¹Met allele). Results from studies of the *XRCC3* Thr²⁴¹Met SNP on the risk of various types of cancer have been inconsistent (17,28-30). Most studies have suggested that the Met allele is related to an increased risk of cancer (28-30). The Thr-to-Met substitution results in an amino acid change from a neutral hydrophilic residue with a hydroxyl group to a hydrophobic one with a methyl sulfur group, which may result in a substantial change in the protein structure and function. A higher level of DNA adducts detected by ³²P-postlabeling was associated with the Met allele in a study of healthy Italians (31). In addition, a possible damaging functional consequence of the *XRCC3* Met allele has been suggested by the sorting intolerant from tolerant (SIFT) algorithm (32). However, phenotypic data showed that this SNP was not associated with radiation-induced G₂-phase delay (33), and had a weak effect on cell survival after cells were treated with mitomycin-C (34). The current study failed to show an association between this polymorphism and the risk of pancreatic cancer.

It has been argued that the variation in relevant genes that actually influence phenotypic expression may be so large so as to preclude detection of simple associations between particular variants and the disease (35). We have attempted to evaluate two polymorphic loci haplotypes of the *XRCC3* gene. As the two polymorphic sites examined are physically close and are in considerable linkage disequilibrium, the haplotype analyses had little advantage over SNP-based analyses in the current study.

The current study had some limitations. Although the study had adequate power in examining the main effect of *XRCC2/XRCC3* polymorphisms on pancreatic cancer risk if the expected OR was 1.66, it is underpowered (<80%) in detecting a gene-smoking interaction for the *XRCC2* polymorphism if only a moderately increased risk was expected (OR <3.0). Therefore, the exploratory analysis of a *XRCC2* genotype-smoking interaction in modifying the risk of pancreatic cancer should be interpreted with caution. Testing multiple possible interaction terms is prone to false-positive findings. Although the interaction is biologically plausible, it

can only be expected to be hypothesis generating and would need confirmation in future study. There are some inherent limitations in a hospital-based case-control study. Our study population (both cases and controls) was from all over the country rather than a random sample from a defined population, which could potentially introduce selection bias. However, the frequencies of the genotypes and the rate of ever-smokers among controls were comparable to those in other U.S. population-based studies (13,36). In addition, similar proportions of cases and controls (52% and 57%) were found from the state of Texas. Therefore, we believe that our controls were the best representatives of the base population from which our cases were derived. As the observations were made in non-Hispanic whites, the results cannot be generalized to other ethnic groups. Because of the limited sample size in African-American and Hispanic participants to our study, data analyses in these ethnic groups will have to depend on meta-analyses or pooled data from future cohort or case-control consortiums.

In conclusion, to our knowledge, this is the first study investigating the selected genetic variants in HR repair genes and the risk of developing pancreatic cancer. We found a risk-modifying effect of *XRCC2* Arg¹⁸⁸His polymorphism on pancreatic cancer among smokers. This observation deserves further replication. Genotype and phenotype association study will help to clarify the role of these genetic variants in cancer predisposition. Knowing the specific expressions of the DNA repair gene in different organs may help distinguish which repair pathways dominate under certain types of exposure attack and may aid in the interpretation of epidemiologic findings.

STUDY HIGHLIGHTS

What Is Current Knowledge

- Cigarette smoking is a known risk factor of pancreatic cancer.
- The genetic determinants for individual susceptibility to smoking-related pancreatic cancer are unknown.

What Is New Here

- A polymorphic variant of the DNA repair gene *XRCC2* was associated with a significantly higher risk of smoking-related pancreatic cancer.

ACKNOWLEDGMENTS

People who agreed to participate in this research made this study possible. We thank Rabia Khan, Kaustubh Mestry, Ajay Nooka, and Hui Liu for their contributions to patient recruitment and data collection and management. We also thank all physicians and the clinical staff at the Gastrointestinal Cancer Clinic who helped with the patient recruitment. We appreciate the constant laboratory support provided by Ping Chang, Yanan Li, Jijiang Zhu, and Yingqiu Du. We thank Kathryn Carnes for editing this manuscript.

CONFLICT OF INTEREST

Guarantor of the article: Donghui Li, Ph.D.

Specific author contributions: Li Jiao: performed the genotyping assay, analyzed the data, wrote the manuscript, and approved the final draft of the manuscript; Manal M. Hassan and Melissa L. Bondy: contributed to the study design and execution of the field work and approved the final draft of the manuscript; Robert A. Wolff, Douglas B. Evans, and James L. Abbruzzese: contributed to patient recruitment and secured The Lockton Research Funds and approved the final draft of the manuscript; Donghui Li: was the principal investigator of the NCI grant and was responsible for the study design and execution, data management and analysis, as well as manuscript preparation.

Financial support: This work was supported by National Institutes of Health grant CA98380, National Institute of Environmental Health Sciences Center grant P30 ES07784, National Institutes of Health Cancer Center Support (Core) grant CA16672, and a research grant from The Lockton Research Funds.

Potential competing interests: None.

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Table 1

Distribution of Selected Variables Among Cases and Controls

	Cases (N = 408 [%])	Controls (N = 449 [%])	Univariate OR (95% CI)	P Value (χ^2 Test)
Age (yr)				
<50	46 (11.3)	59 (13.1)		
50-59	118 (28.9)	130 (28.9)		
60-69	132 (32.3)	157 (35.0)		
≥70	112 (27.4)	103 (22.9)	Matching factor	Matching factor
Sex				
Men	228 (55.9)	242 (53.9)		
Women	180 (44.1)	207 (46.1)	Matching factor	Matching factor
Race [†]				
Non-Hispanic white	408 (87.2)	449 (90.2)		
African American	23 (4.9)	20 (4.0)		
Hispanic origin	32 (6.8)	24 (4.8)		
Other	5 (1.1)	5 (1.0)	Matching factor	Matching factor
Smoking status				
Never	157 (38.5)	221 (49.2)	1.00	
Ever	251 (61.5)	228 (50.8)	1.55 (1.17-2.05)	0.002
Smoking pack-years [*]				
Never	157 (38.5)	221 (49.2)	1.00	
<22	100 (24.5)	114 (25.4)	1.23 (0.88-1.73)	
≥22	151 (37.0)	114 (25.4)	1.86 (1.36-2.56)	0.001

* Categorized by 50th percentile value of the controls who ever smoked.

[†] Race: based on 468 cases and 449 controls.

Table 2
XRCC2 and XRCC3 Genotype Frequency in Cases and Controls and OR Estimates for Pancreatic Cancer*

Genotype	Non-Hispanic Whites		African American†		Hispanic origin†	
	Cases (N [%])	Controls (N [%])	Cases (N [%])	Controls (N [%])	Cases (N [%])	Controls (N [%])
XRCC2 Arg ¹⁸⁸ /His	(N = 408)	(N = 449)	(N = 17)	(N = 14)	(N = 24)	(N = 19)
Arg/Arg	340 (88.1)	368 (88.0)	17 (100.0)	17 (100.0)	21 (87.5)	16 (84.2)
Arg/His	44 (11.4)	49 (11.7)	0	0	3 (12.5)	3 (15.8)
His/His	2 (0.5)	1 (0.2)	0	0	0	0
His allele frequency	0.057	0.058	0	0	0.062	0.079
XRCC3 A>G 17893						
AA	178 (46.7)	207 (48.0)	11 (64.7)	9 (64.3)	12 (50.0)	10 (52.6)
AG	162 (42.5)	185 (42.9)	6 (35.3)	4 (28.6)	9 (37.5)	9 (47.4)
GG	41 (10.8)	39 (9.0)	0	1 (7.1)	3 (12.5)	0
G allele frequency	0.32	0.30	0.17	0.21	0.31	0.24
XRCC3 Thr ²⁴ /Met						
Thr/Thr	137 (36.4)	168 (38.5)	10 (58.8)	7 (46.7)	13 (50.0)	11 (55.0)
Thr/Met	182 (48.4)	194 (44.5)	4 (23.5)	8 (53.3)	12 (46.2)	8 (40.0)
Met/Met	57 (15.2)	74 (17.0)	3 (17.6)	0	1 (3.8)	1 (5.0)
Met allele frequency	0.39	0.39	0.29	0.26	0.27	0.25

* Numbers do not added up to the total because of missing values.

† The participants with missing genotype data were not included for calculating genotype frequency. OR estimate is not presented for African American and Hispanic origin participants.

Table 3
Joint Effect of *XRCC2* and *XRCC3* Polymorphism and Smoking in Modifying Risk of Pancreatic Cancer

Genotype	Ever-Smoker	Cases	Controls	OR*	95% CI	$P_{\text{trend}}^{\dagger}$	$P_{\text{interaction}}^{\#}$
<i>XRCC2</i> Arg ¹⁸⁸ /His							
Arg/Arg	No	135	176	1.00			
Any His	No	14	32	0.57	0.29-1.11		
Arg/Arg	Yes	205	192	1.39	1.03-1.88		
Any His	Yes	32	18	2.32	1.25-4.31	0.002	0.02
<i>XRCC3</i> A17893 G							
AA	No	68	97	1.00			
AG + GG	No	79	117	0.96	0.63-1.47		
AA	Yes	110	110	1.43	0.95-2.14		
AG + GG	Yes	124	107	1.65	1.10-2.48	0.002	0.52
<i>XRCC3</i> Thr ²⁴¹ /Met							
Thr/Thr	No	51	89	1.00			
Any Met	No	91	125	1.27	0.82-1.97		
Thr/Thr	Yes	86	79	1.90	1.20-3.01		
Any Met	Yes	148	143	1.81	1.19-2.73	0.002	0.33

* Crude OR is presented because no confounding factor was identified.

[†] P value for score test of trend of odds.

[#] P value for likelihood ratio test.

Table 4
Joint Effect of XRCC2 Polymorphism and Pack-Years of Smoking in Modifying Risk of Pancreatic Cancer

XRCC2 Arg ¹⁸⁸ His	Pack-Years Smoked	Cases	Controls	OR [*]	95% CI	P _{trend} [†]	P _{interaction} [‡]
Arg/Arg	0	135	176	1.00			
Any His	0	14	32	0.57	0.29-1.11		
Arg/Arg	<22	83	94	1.15	0.79-1.67		
Any His	<22	11	10	1.43	0.59-3.48		
Arg/Arg	>22	122	98	1.62	1.15-2.30		
Any His	≥22	21	8	3.42	1.47-7.96	0.0002	0.05

* Crude OR is presented because no confounding factor was identified.

† P value for score test of trend of odds.

‡ P value for likelihood ratio test.