Cancer Epidemiology, Biomarkers & Prevention

Inflammation-Related Gene Variants as Risk Factors for Pancreatic Cancer

Kaye M. Reid-Lombardo[1], Brooke L. Fridley[2], William R. Bamlet[2], Julie M. Cunningham[4], Michael G. Sarr[1], and Gloria M. Petersen[3]

Abstract

Background: Recent reports support an association between chronic inflammation and progression to pancreatic cancer (PC).

Methods: This case–control, candidate gene association study evaluated 1,354 Caucasian patients with pancreatic ductal adenocarcinoma and 1,189 healthy Caucasian controls. We genotyped 1,538 single nucleotide polymorphism (SNP) in 102 genes from inflammatory pathways involving *NF-κB*. Primary tests of association assumed a multiplicative (log-additive) genotype effect; secondary analyses examined dominant, additive, and recessive SNP effects.

Results: After adjusting for known risk factors for PC, single SNP analysis revealed an association between four SNPs in *NOS1* and one in the *CD101* gene with PC risk. These results, however, were not replicated in a PC case–control and cohort population.

Conclusion: *NOS1* and *CD101* may be associated with a risk of PC; however, these findings did not replicate in other PC populations. Future research is needed into the possible role of *NOS1* and *CD101* for PC.

Impact: This research shows a lack of association between genetic variation in 102 inflammation-related genes and PC. Future research is needed into the possible role of other inflammation-related genes and PC risk. *Cancer Epidemiol Biomarkers Prev; 20(6); 1251–4. ©2011 AACR.*

Introduction

Chronic inflammation has been recognized as a contributing factor in the development of a subset of highly lethal pancreatic adenocarcinomas (PC; ref. 1). Studies linking chronic inflammation to PC report an upregulation of cyclooxygenase (*COX*; ref. 2) and nitric oxide (*NO*) gene expression (3), important mediators of inflammation in PC tissue specimens. The trigger for upregulation of *COX* and *NO* is the transcription factor, *NF-κB*. NF-κB activates these genes and others that are involved in inflammation and apoptosis and seems to promote pancreatic cell growth by inhibition of apoptosis (4). We hypothesized that genetic polymorphisms in inflammation-related genes involving *NF-κB*–related inflammatory pathways are associated with risk of PC.

Authors' Affiliations: Divisions of [1]Gastroenterologic and General Surgery, [2]Biomedical Statistics and Informatics, and [3]Epidemiology, and [4]Department of Laboratory Medicine and Pathology, Mayo Clinic, Rochester, Minnesota

Corresponding Author: Kaye M. Reid-Lombardo, Division of Gastroenterologic and General Surgery, Mayo Clinic, 200 First Street S.W., Rochester, MN 55905. Phone: 507-284-2717; Fax: 507-284-5196. E-mail: reidlombardo.kaye@mayo.edu

doi: 10.1158/1055-9965.EPI-11-0264

Methods

PC patients were recruited prospectively to a PC research registry using processes approved by the Mayo Clinic Institutional Review Board. Consenting patients completed a comprehensive questionnaire, donated a blood sample, and provided medical release for study of archival tissue.

Subjects

Cases were patients with histologically proven pancreatic ductal adenocarcinoma evaluated at Mayo Clinic from 2000 to 2008. Controls were healthy, clinic-based patients without a personal history of cancer (except nonmelanoma skin cancer) and were frequency matched to cases for age (±5 years), sex, race, and geographic region of residence (5).

Candidate gene and single nucleotide polymorphism selection

Candidate genes were selected using a combination of literature review and the bioinformatics tools Ingenuity Systems and MetaCore from GeneGo, Inc. to identify genes involved in the pathogenesis of PC that are in the *NF-κB*–related inflammatory pathways. Genotyping and Quality Control measures were followed as reported previously (5). Call rates for single nucleotide polymorphisms (SNP) were 99.2% and 97.3%. Eighty-four of 1,538 SNPs failed to amplify.

Table 1. Patient characteristics[a]

	Control ($N = 1,171$)	Case ($N = 1,329$)	Total ($N = 2,500$)	*P*
Sex				
Female	574 (49%)	564 (42.4%)	1,138 (45.5%)	0.0010
Male	597 (51%)	765 (57.6%)	1,362 (54.5%)	
Race				
White/Caucasian	1,171 (100%)	1,329 (100%)	2,500 (100%)	
Age at time of PC diagnosis				
N	1,171	1,329	2,500	0.3356
Mean (SD)	66.1 (10.51)	65.7 (10.71)	65.9 (10.62)	
Median	67.0	67.0	67.0	
Q1, Q3	59.0, 74.0	58.0, 74.0	59.0, 74.0	
Range	(30.0–95.0)	(28.0–91.0)	(28.0–95.0)	
Ever smoker				
No	627 (53.5%)	532 (40%)	1,159 (46.4%)	<0.0001
Yes	544 (46.5%)	797 (60%)	1,341 (53.6%)	
Pack years				
N	1,146	1,083	2,229	<0.0001
Mean (SD)	9.6 (17.61)	14.8 (22.16)	12.1 (20.12)	
Median	0.0	0.4	0.0	
Q1, Q3	0.0, 12.8	0.0, 25.0	0.0, 19.0	
Range	(0.0–125.0)	(0.0–140.0)	(0.0–140.0)	
Diabetes > 2 years prior to study entry				
No	1,106 (94.4%)	1,156 (87%)	2,262 (90.5%)	<0.0001
Yes	65 (5.6%)	173 (13%)	238 (9.5%)	
Body mass index				
N	1,171	1,329	2,500	<0.0001
Mean (SD)	27.2 (4.63)	28.4 (5.69)	27.8 (5.26)	
Median	26.6	27.6	27.1	
Q1, Q3	24.0, 29.5	24.4, 31.1	24.3, 30.3	
Range	(17.7–54.2)	(15.3–59.0)	(15.3–59.0)	
First-degree relative with pancreatic cancer				
No	1,127 (96.2%)	1,244 (93.6%)	2,371 (94.8%)	0.0029
Yes	44 (3.8%)	85 (6.4%)	129 (5.2%)	

[a]Only subjects included in final analysis.

Statistical analysis

Unconditional logistic regression analysis was used to estimate ORs and 95% CIs for the risk of PC with each SNP. The regression model included covariates of age, sex, smoking status, body mass index (kg/m^2), family history of PC, and preexisting diabetes (>2 years). Primary tests of association assumed a multiplicative (log-additive) genotype effect, equivalent to the Armitage test for trend. To account for multiple testing, SNPs were deemed significantly associated with PC if $P < 0.001$.

Validation studies

Genotyping data from PanScan, the Pancreatic Cancer Cohort Consortium, and Pancreatic Cancer Case-Control Consortium (6, 7) were obtained from dbGap (8). Imputation in non-Mayo samples was accomplished with MACH software (9) using HapMap. The imputed allele dosage was used to validate SNPs of interest. All analyses were unadjusted.

Results

Demographics for all subjects are reported in Table 1. Single SNP analysis revealed an association between 4 SNPs in *NOS1* and 1 SNP in the *CD101* gene and the risk of PC (Table 2). An additional 14 and 4 imputed SNPs with values of $P = 5E^{-4}$ to $8E^{-4}$ were observed in CD101 and NOS1, respectively (data not shown). The significant SNPs failed confirmation in the validation set using both PanScan data sets.

Discussion

We report the results of a case–control study evaluating the risk of inflammation-related gene variants with PC.

Table 2. Genotyping and validation results

SNP	Primary set					PanScan (cohort)					PanScan (case–control)				
	Location	Number cases	Number controls	OR	*P*	Number cases	Number controls	Frequency of coded allele	OR	*P*	Number cases	Number controls	Frequency of coded allele	OR	*P*
CD101, rs10923193	117338325	1,312	1,169	0.8	0.000903	1,408	1,461	0.27	1.04	0.48	1,090	1,168	0.26	0.96	0.58
G/G		755 (0.58)	613 (0.52)	1	1										
A/G		492 (0.42)	460 (0.35)	0.87	0.00126										
A/A		65 (0.05)	96 (0.08)	0.54	0.00126										
Dominant				0.81	0.013										
Recessive				0.58	0.000996										
NOS1, rs3782203	116204794	1,328	1,171	1.24	0.0016	1,408	1,461	0.2	1	0.95	1,090	1,168	0.2	0.97	0.64
G/G		780 (0.59)	748 (0.64)	1	1										
A/G		464 (0.4)	378 (0.28)	1.17	0.0733										
A/A		84 (0.06)	45 (0.04)	1.8	0.00222										
Dominant				1.23	0.0109										
Recessive				1.7	0.005										
NOS1, rs9658350	116208811	1,327	1,170	1.24	0.00175	1,408	1,461	0.2	0.99	0.87	1,090	1,168	0.2	0.96	0.58
A/A		774 (0.58)	742 (0.63)	1	1										
G/A		469 (0.4)	383 (0.29)	1.16	0.0779										
G/G		84 (0.06)	45 (0.04)	1.79	0.00225										
Dominant				1.23	0.0124										
Recessive				1.7	0.005										
NOS1, rs532967	116216722	1,329	1,170	1.25	0.00159	1,408	1,461	0.18	0.98	0.76	1,090	1,168	0.18	0.92	0.31
G/G		817 (0.61)	780 (0.67)	1	1										
A/G		447 (0.38)	357 (0.27)	1.19	0.0502										
A/A		65 (0.05)	33 (0.03)	1.89	0.00356										
Dominant				1.25	0.00972										
Recessive				1.79	0.00733										
NOS1, rs547954	116238889	1,329	1,171	1.27	0.00085	1,408	1,461	0.17	0.98	0.79	1,090	1,168	0.17	0.90	0.18
G/G		829 (0.62)	796 (0.68)	1	1										
A/G		437 (0.37)	343 (0.26)	1.21	0.0273										
A/A		63 (0.05)	32 (0.03)	1.9	0.00373										
Dominant				1.27	0.00433										
Recessive				1.79	0.00882										

To date, this is the largest evaluation of risk for PC that focuses primarily on genes in the inflammation pathways involving *NF-κB*. The 102 genes code for proinflammatory mediators, inhibitors, or activators of *NF-κB*. Polymorphisms of *NOS1* and *CD101* showed increased (*NOS1*) and decreased (*CD101*) risk association for PC. The *NOS1* SNPs were in high linkage disequilibrium (LD) and located across a region of 2 LD blocks on chromosome 12. Imputed and genotyped SNPs from *CD101* were located in a single LD block. Attempts to validate these data utilizing the PanScan cohort and PanScan case–control studies of PC were unsuccessful. Potential reasons for the lack of validation include the differences in study designs and accrual methods of the 3 data sets and the inability to adjust the PanScan data sets.

Conclusion

Of the 102 genes evaluated, *NOS1* and *CD101* may be associated with an increased and decreased risk of PC, respectively. However, these findings did not replicate in a follow-up study of 2 PC populations. Future research is needed to determine the role, if any, of *NOS1* and *CD101* for risk of PC.

Disclosure of Potential Conflicts of Interest

The contents of this article are solely the responsibility of the authors and do not necessarily represent the official view of NCRR or NIH. No potential conflicts of interests to disclose.

Grant Support

This publication was made possible by the Mayo Clinic SPORE in Pancreatic Cancer grant (P50 CA 102701) and grant number 1 KL2 RR024151 from the National Center for Research Resources (NCRR), a component of the NIH, and the NIH Roadmap for Medical Research. Information on NCRR is available at http://www.ncrr.nih.gov/. Information on Reengineering the Clinical Research Enterprise can be obtained from http://nihroadmap.nih.gov.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked *advertisement* in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

Received March 16, 2011; accepted March 17, 2011; published OnlineFirst April 5, 2011.

References

1. Farrow B, Sugiyama Y, Chen A, Uffort E, Nealon W, Evers M. Inflammatory mechanisms contributing to pancreatic cancer development. Ann Surg 2004;239:763–9; discussion 769–71.
2. Tucker ON, Dannenberg AJ, Yang EK, Zhang F, Teng L, Daly JM, et al. Cyclooxygenase-2 expression is up-regulated in human pancreatic cancer. Cancer Res 1999;59:987–90.
3. Jenkins DC, Charles IG, Thomsen LL, Moss DW, Holmes LS, Baylis SA, et al. Roles of nitric oxide in tumor growth. Proc Natl Acad Sci U S A 1995;92:4392–6.
4. Liptay S, Weber CK, Ludwig L, Wagner M, Adler G, Schmid RM. Mitogenic and antiapoptotic role of constitutive NF-kappaB/Rel activity in pancreatic cancer. Int J Cancer 2003;105:735–46.
5. McWilliams RR, Bamlet WR, de Andrade M, Rider DN, Cunningham JM, Petersen GM. Nucleotide excision repair pathway polymorphisms and pancreatic cancer risk: evidence for role of MMS19L. Cancer Epidemiol Biomarkers Prev 2009;18:1295–302.
6. Petersen GM, Amundadottir L, Fuchs CS, Kraft P, Stolzenberg-Solomon RZ, Jacobs KB, et al. A genome-wide association study identifies pancreatic cancer susceptibility loci on chromosomes 13q22.1, 1q32.1 and 5p15.33. Nat Genet 2010;42:224–8.
7. Amundadottir L, Kraft P, Stolzenberg-Solomon RZ, Fuchs CS, Petersen GM, Arslan AA, et al. Genome-wide association study identifies variants in the ABO locus associated with susceptibility to pancreatic cancer. Nat Genet 2009;41:986–90.
8. Mailman MD, Feolo M, Jin Y, Kimura M, Tryka K, Bagoutdinov R, et al. The NCBI dbGaP database of genotypes and phenotypes. Nat Genet 2007;39:1181–6.
9. Li Y, Willer CJ, Ding J, Scheet P, Abecasis GR. MaCH: using sequence and genotype data to estimate haplotypes and unobserved genotypes. Genet Epidemiol 2006;34:816–834. Available from: http://www.sph.umich.edu/csg/yli/mach/download/.