Pooled Analysis of Phosphatidylinositol 3-Kinase Pathway Variants and Risk of Prostate Cancer

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Abstract

The phosphatidylinositol 3-kinase (PI3K) pathway regulates various cellular processes, including cellular proliferation and intracellular trafficking, and may affect prostate carcinogenesis. Thus, we explored the association between single-nucleotide polymorphisms (SNP) in PI3K genes and prostate cancer. Pooled data from the National Cancer Institute Breast and Prostate Cancer Cohort Consortium were examined for associations between 89 SNPs in PI3K genes (PIK3C2B, PIK3AP1, PIK3C2A, PIK3CD, and PIK3R3) and prostate cancer risk in 8,309 cases and 9,286 controls. Odds ratios (OR) and 95% confidence intervals (95% CI) were estimated using logistic regression. SNP rs7556371 in PIK3C2B was significantly associated with prostate cancer risk [OR per allele, 1.08 (95% CI, 1.03–1.14); Ptrend = 0.0017] after adjustment for multiple testing (Padj = 0.024). Simultaneous adjustment of rs7556371 for nearby SNPs strengthened the association [OR per allele, 1.21 (95% CI, 1.09–1.34); Ptrend = 0.0003]. The adjusted association was stronger for men who were diagnosed before the age of 65 years [OR per allele, 1.47 (95% CI, 1.20–1.79); Ptrend = 0.0001] or had a family history [OR per allele, 1.57 (95% CI, 1.11–2.23); Ptrend = 0.0114], and was strongest in those with both characteristics [OR per allele = 2.31 (95% CI, 1.07–5.07), P-interaction = 0.005]. Increased risks were observed among men in the top tertile of circulating insulin-like growth factor-I (IGF-I) levels [OR per allele = 1.46 (95% CI, 1.04–2.06); Ptrend = 0.075]. No differences were observed with disease aggressiveness (Gleason grade ≥8 or stage T3/T4 or fatal). In conclusion, we observed a significant association between PIK3C2B and prostate cancer risk, especially for familial, early-onset disease, which may be attributable to IGF-dependent PI3K signaling. Cancer Res; 70(6); 2389–96. ©2010 AACR.

Introduction

The phosphatidylinositol 3-kinase (PI3K) pathway regulates various cellular processes such as cell growth, proliferation, apoptosis, motility, differentiation, survival, and intracellular trafficking (1). PI3Ks are heterodimeric lipid kinases that are composed of regulatory and catalytic subunits that catalyze the production of several phosphoinositides critical for the signal transduction in these multiple cellular processes (2). In addition, numerous growth factors signal through the PI3K pathway (3, 4), including insulin-like growth factors (IGF), which have also been linked with prostate carcinogenesis (5-8). Mutation, amplification, and rearrangement in the PI3K pathway and its downstream targets have been observed in several cancer sites, including prostate cancer (9–11). Because of the role of PI3Ks in cell proliferation, much of the research on this pathway concerns its potential as a target for anticancer therapies.

Despite the well-known role of this pathway in cancer progression, genetic variants in PI3K genes have not been well studied. One study found an association between a PIK3C4 SNP (rs2865084) and endometrioid ovarian cancer but not overall ovarian cancer risk (12). Two studies have evaluated the functional variant Met326Leu (rs3730089) in PIK3R1 and risk of colon (13) and prostate (14) cancers and found no

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Note: Supplementary data for this article are available at Cancer Research Online (http://cancerres.aacrjournals.org/).

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association with either cancer site. Although some studies have looked at polymorphisms in genes of downstream targets of PI3K and prostate cancer risk (15–17), there is minimal information about the effect of variants in the PI3K gene family on prostate cancer risk.

Given the limited evaluation of PI3K gene variants, we sought to further explore the association between germline polymorphisms, which may alter the function of these genes and the proteins they encode, and risk of prostate cancer. Pooled data from seven prospective studies in the National Cancer Institute (NCI) Breast and Prostate Cancer Cohort Consortium (BPC3; ref. 18) were examined for associations between 89 single-nucleotide polymorphisms (SNP) in five PI3K pathway genes, PIK3C2B (chromosome 1), PIK3API (chromosome 10), PIK3C2A (chromosome 11), PIK3CD (chromosome 1), and PIK3R3 (chromosome 1), and prostate cancer risk in 8,751 cases and 9,742 controls. We also evaluated the effects of these PI3K pathway SNPs and circulating IGF-I and IGFBP-3 on prostate cancer risk in a subgroup of 6,076 men using prediagnostic sera.

Materials and Methods

Study population. The BPC3 has been described elsewhere (18). Briefly, the consortium includes large well-established cohorts assembled in the United States and Europe that have DNA for genotyping and extensive questionnaire data from cohort members. The prostate cancer study includes seven case-control studies nested within these cohorts: the American Cancer Society (ACS) Cancer Prevention Study II, the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study (ATBC), the European Prospective Investigation into Cancer and Nutrition (EPIC), the Health Professionals Follow-up Study (HPFS), the Physicians Health Study (PHS), the Hawaii-Los Angeles Multiethnic Cohort (MEC), and the Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial (PLCO). With the exception of MEC and PLCO, most subjects in these cohorts were Caucasian. Cases were identified in each cohort by self-report, with subsequent confirmation of the diagnosis from medical records, and/or linkage with population-based tumor registries. Controls were free of prostate cancer at selection and were matched to cases within each cohort by age, ethnicity, and other select factors, such as country of residence for EPIC. Written informed consent was obtained from all subjects, and each cohort study was approved by the appropriate institutional review boards.

IGF-I and IGFBP-3 blood levels. Data on prediagnostic blood levels of IGF-I and IGFBP-3 were available in six of the seven BPC3 cohorts (ATBC, EPIC, HPFS, MEC, PHS, and PLCO; IGF-I: n = 6,076; IGFBP-3: n = 6,059; refs. 5–8, 19–21). Samples from three studies (ATBC, HPFS, and PHS) were measured in the laboratory of the Cancer Prevention Research Unit, Departments of Medicine and Oncology, Lady Davis Research Institute of the Jewish General Hospital and McGill University, and the remaining three studies (EPIC, MEC, and PLCO) were measured in the laboratory of the Hormones and Cancer Team at IARC; all used ELISA from Diagnostic System Laboratories. Blood levels were categorized into tertiles based on the distribution among the controls. Although the blood level assays were done at different times, batch effects did not seem to confound the results, and the analyses were pooled across cohorts controlling for study.

Genotyping and SNP selection. Tag SNPs for the five PI3K candidate genes potentially related to the IGF pathway were selected using the CEU HapMap data assuming a minor allele frequency >0.02, an Illumina design score >0.4, and an R^2 >0.8 for binning (22). Genotyping in the prostate cancer cases and controls was done in four laboratories (University of Southern California, Los Angeles, CA; Harvard School of Public Health, Boston, MA; Core Genotyping Facility, National Cancer Institute, Bethesda, MD; and Imperial College, London, United Kingdom) using Illumina GoldenGate technology as part of a larger array of 1,536 SNPs in total. Each genotype center genotyped 30 CEU HapMap trios to evaluate interlaboratory reproducibility, and for the 1,536 SNPs that made up the Illumina platform, the interlab concordance was 99.5% (before excluding failed SNPs or samples). Within each study, blinded duplicate samples (~5%) were also included and concordance of these samples ranged from 97.2% to 99.9% across studies.

Data filtering and imputation. Any sample where more than 25% of the SNPs attempted on a given platform failed was removed from the data set. Data were filtered by study to remove poorly performing SNPs: All SNPs that failed in 25% or more samples were excluded from the data set, as were all SNPs that showed statistically significant (P < 10^-5) deviations from Hardy-Weinberg equilibrium genotype frequencies among European-ancestry controls and all SNPs with minor allele frequency <1%. Any SNP that was missing or excluded in more than three studies or exhibited large differences in European-ancestry allele frequencies across cohorts (Fst > 0.02) was excluded from further analysis. The current analysis included 8,751 prostate cases and 9,742 controls. Among these, 8,309 cases and 9,286 controls had genotype information.

The MACH software program (13) was used to impute SNPs that were polymorphic in any of the HapMap reference panels using observed genotypes from the BPC3 subjects and phased haplotypes from HapMap samples (release #21; ref. 23). Genotypes for European-ancestry subjects were imputed using the CEPH European (CEU) reference panel; those for Japanese Americans were imputed using the combined Han Chinese and Japanese panels (CHB + JPT); those for remaining subjects (African Americans, Latinos, and Native Hawaiians) were imputed using a “cosmopolitan” panel of all HapMap samples (CEU + CHB + JPT + YRI; ref. 24). Imputation was done stratified by study and ethnicity. Poorly imputed SNPs with an estimated correlation between the imputed and true genotypes <30% were excluded from analysis (23). SNPs that were dropped in more than three European-ancestry cohorts were excluded from analysis.

13 http://www.sph.umich.edu/csg/abecasis/MaCH/index.html
Statistical analysis. Odds ratios (OR) and 95% confidence intervals (95% CI) adjusted for age (5-y intervals), cohort (plus country for EPIC), and ethnicity for each SNP with minor allele frequency >1% were estimated using unconditional logistic regression. Genotypes were coded either as counts of the variant allele (trend test) or as two indicator variables, one for heterozygotes and one for variant homozygotes (2-df test). Analyses were done in all subjects, separately for each ethnicity, and separately for each ethnicity within study. Aggressive disease was defined as Gleason grade ≥8 or stage T3/T4 or fatal prostate cancer. Heterogeneity between studies and interactions between the SNPs and other covariates were assessed by including the cross-product terms as well as the main effect terms in regression models, and the statistical significance of the interaction was evaluated by comparing nested models with and without the cross-product terms using a likelihood ratio test. All analyses were done using SAS statistical software (SAS Institute, Inc.).

Pairwise linkage disequilibrium measures (\(D'\) and \(r^2\)) were estimated and haplotypes were constructed using the expectation-maximization algorithm (25) in HaploStats (26, 27).

Risks for individual haplotypes were calculated among whites only in PIK3C2B, assuming a log-additive model for each haplotype adjusting for age and cohort. Haplotypes with a frequency of <1% were collapsed into a single category, and the most common haplotype was used as the reference.

To take into account the large number of tests done, the number of effective independent variables, \(M_{eff}\), was calculated for each gene by use of the SNP Spectral Decomposition approach (28), and \(P\) values adjusted for multiple testing were calculated using the gene-wide \(M_{eff}\) values.

Results

Demographic and prostate cancer–related characteristics among BPC3 prostate cancer cases and controls are presented in Table 1. The majority of subjects were white (76.5%) and diagnosed over the age of 65 years (70.8%). Aggressive cases of prostate cancer made up about a quarter of the cases (25.3%), and 7.8% of subjects reported a family history of disease in first-degree relatives. Family history was not available for the EPIC and PHS cohorts.

Table 1. Characteristics of study population by cohort

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Pooled</th>
<th>ACS</th>
<th>ATBC</th>
<th>EPIC</th>
<th>HPFS</th>
<th>MEC</th>
<th>PHS</th>
<th>PLCO</th>
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<td>Prostate cancer</td>
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<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Case</td>
<td>8,751</td>
<td>1,296</td>
<td>1,058</td>
<td>953</td>
<td>700</td>
<td>2,320</td>
<td>1,101</td>
<td>1,323</td>
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<tr>
<td>Control</td>
<td>9,742</td>
<td>1,293</td>
<td>1,058</td>
<td>1,320</td>
<td>700</td>
<td>2,290</td>
<td>1,430</td>
<td>1,651</td>
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<td></td>
<td></td>
<td></td>
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<tr>
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<td>1,052</td>
<td>335</td>
<td>373</td>
<td>523</td>
<td>1,516</td>
<td>719</td>
<td>1,054</td>
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<td>204</td>
<td>324</td>
<td>141</td>
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<td>40</td>
<td>399</td>
<td>439</td>
<td>22</td>
<td>25</td>
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<td>Age at case diagnosis (y)</td>
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<tr>
<td>&lt;65</td>
<td>5,397</td>
<td>436</td>
<td>371</td>
<td>1,098</td>
<td>356</td>
<td>1,439</td>
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<tr>
<td>≥65</td>
<td>13,096</td>
<td>2,153</td>
<td>1,745</td>
<td>1,175</td>
<td>465</td>
<td>3,171</td>
<td>1,864</td>
<td>1,944</td>
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<td>Ethnicity</td>
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<td>White</td>
<td>14,138</td>
<td>2,566</td>
<td>2,116</td>
<td>2,273</td>
<td>1,315</td>
<td>909</td>
<td>2,415</td>
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<td>Black</td>
<td>1,762</td>
<td>95</td>
<td>1,175</td>
<td>1,175</td>
<td>1,333</td>
<td>1,296</td>
<td>—</td>
<td>429</td>
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<td>Hispanic</td>
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<td>70</td>
<td>1,296</td>
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<td>933</td>
<td>933</td>
<td>933</td>
<td>933</td>
<td>—</td>
<td>—</td>
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<tr>
<td>Native Hawaiian</td>
<td>139</td>
<td>0.7</td>
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<td>139</td>
<td>139</td>
<td>139</td>
<td>—</td>
<td>—</td>
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<tr>
<td>Other/unknown</td>
<td>225</td>
<td>1.2</td>
<td>23</td>
<td>85</td>
<td>116</td>
<td>116</td>
<td>1</td>
<td>1 (0.03)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;25</td>
<td>6,919</td>
<td>985</td>
<td>803</td>
<td>714</td>
<td>493</td>
<td>1,624</td>
<td>1,500</td>
<td>800</td>
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<tr>
<td>25–30</td>
<td>8,599</td>
<td>1,273</td>
<td>1,149</td>
<td>1,189</td>
<td>465</td>
<td>2,213</td>
<td>948</td>
<td>1,497</td>
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<tr>
<td>≥30</td>
<td>2,501</td>
<td>299</td>
<td>297</td>
<td>364</td>
<td>84</td>
<td>731</td>
<td>83</td>
<td>643</td>
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<td>474</td>
<td>32</td>
<td>2</td>
<td>6</td>
<td>358</td>
<td>42</td>
<td>—</td>
<td>34</td>
</tr>
<tr>
<td>Family history†</td>
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<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>No</td>
<td>11,582</td>
<td>2,163</td>
<td>1,762</td>
<td>1,155</td>
<td>3,783</td>
<td>2,719</td>
<td>—</td>
<td>2,719</td>
</tr>
<tr>
<td>Yes</td>
<td>1,439</td>
<td>426</td>
<td>89</td>
<td>245</td>
<td>424</td>
<td>255</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Missing</td>
<td>5,472</td>
<td>296</td>
<td>265</td>
<td>403</td>
<td>2,531</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

aAggressive disease: Gleason ≥8 or stage T3/T4 or fatal.

†Prostate cancer in first-degree relatives.
Eighty-nine genotyped and 251 imputed SNPs in PIK3CD, PIK3CA, PIK3R3, PIK3AP1, and PIK3CB were evaluated for their association with prostate cancer (Table 2). Among the five PI3K pathway genes, only PIK3CB showed a cluster of SNPs related to prostate cancer risk, where at least one genotyped SNP remained associated with risk after adjustment for the number of effective tests (P = 0.024). Specifically, of the 15 genotyped and 89 imputed SNPs, 11 SNPs (1 genotyped, 10 imputed) in PIK3CB showed an association for trend with prostate cancer at \( P < 0.01 \). Main-effect \( P \) values for trend for all SNPs (genotyped and imputed) in PI3K genes and risk of prostate cancer are presented in Supplementary Table S1.

SNPs related to prostate cancer were clustered upstream and in the first two introns of PIK3CB (Table 3). The strongest genotyped association was for rs7556371 with an \( OR_{per	ext{ allele}} = 1.24 \) (95% CI, 1.09–1.41); \( P_{\text{trend}} = 0.0008 \). The association was stronger for men with prostate cancer diagnosed before age 65 years [\( OR_{\text{per	ext{ allele}}} = 1.47 \) (95% CI, 1.20–1.79); \( P_{\text{trend}} = 0.0001, p\text{-interaction} = 0.06 \] and for men with a family history of prostate cancer [\( OR_{\text{per	ext{ allele}}} = 1.57 \) (95% CI, 1.11–2.23); \( P_{\text{trend}} = 0.0114, P\text{-interaction} = 0.02 \]. Men diagnosed before age 65 years who also had a family history were found to have a 2-fold increased risk of prostate cancer [\( OR_{\text{per	ext{ allele}}} = 2.31 \) (95% CI, 1.07–5.07); \( P_{\text{trend}} = 0.034, P\text{-interaction} = 0.005 \]. Increased risks were also observed among obese men [body mass index (BMI) ≥30 kg/m\(^2\); \( OR_{\text{per	ext{ allele}}} = 1.30 \) (95% CI, 0.98–1.71); \( P_{\text{trend}} = 0.0003 \] and among men in the top tertile of circulating IGFBP-3 levels [\( OR_{\text{per	ext{ allele}}} = 1.46 \) (95% CI, 1.04–2.06); \( P_{\text{trend}} = 0.075 \], although interactions with BMI and IGFBP-3 levels were not statistically significant. There was no association among tertiles of IGFBP-3 (data not shown). No differences were observed with disease aggressiveness.

**Discussion**

In this large pooled analysis of prostate cancer cases and controls, we explored the associations between common SNPs in five PI3K pathway genes (PIK3CB, PIK3AP1, PIK3CA, PIK3CD, and PIK3R3) and prostate cancer risk. Among the five genes, we observed significant associations between a cluster of variants located upstream and in the promoter

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**Table 2. Overview of PI3K genes and risk of prostate cancer**

<table>
<thead>
<tr>
<th>Gene abbreviation</th>
<th>Genotyped SNPs</th>
<th>Imputed SNPs</th>
<th>Total no. of SNPs</th>
<th>No. of SNPs with ( P &lt; 0.01 )</th>
<th>Minimum ( P_{\text{trend}} ) for imputed SNPs</th>
<th>Minimum ( P_{\text{trend}} ) for genotyped SNPs</th>
<th>Adjusted minimum ( P_{\text{trend}} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>PIK3CD</td>
<td>15</td>
<td>5</td>
<td>20</td>
<td>0</td>
<td>0.42</td>
<td>0.129</td>
<td>—</td>
</tr>
<tr>
<td>PIK3CA</td>
<td>7</td>
<td>37</td>
<td>44</td>
<td>0</td>
<td>0.099</td>
<td>0.28</td>
<td>—</td>
</tr>
<tr>
<td>PIK3R3</td>
<td>10</td>
<td>45</td>
<td>55</td>
<td>0</td>
<td>0.068</td>
<td>0.069</td>
<td>—</td>
</tr>
<tr>
<td>PIK3AP1</td>
<td>42</td>
<td>75</td>
<td>117</td>
<td>1*</td>
<td>0.0033</td>
<td>0.012</td>
<td>0.396(^1)</td>
</tr>
<tr>
<td>PIK3CB</td>
<td>15</td>
<td>89</td>
<td>104</td>
<td>11(^2)</td>
<td>0.0004</td>
<td>0.0017</td>
<td>0.024(^5)</td>
</tr>
</tbody>
</table>

\(^*\)Imputed SNP.  
\(^1\)One genotyped, 10 imputed.  
\(^2\)Adjusted minimum. 

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PI3Ks play a pivotal role in signal transduction pathways linking insulin with many of its cellular responses (31). Furthermore, class II PI3Ks, including PIK3C2β, have been shown to be activated by insulin (32). The IGF axis has been related to prostate cancer, with elevated blood levels of IGF-I associated with increased risks (33, 34). Obesity, a chronic hyperinsulinemic state resulting in altered IGF levels, has also been associated with prostate cancer risk (35–38). In this analysis, we observed increased risks associated with PIK3C2B variants among obese men and among men in the top tertile of circulating IGF-I levels. Although these interactions were not statistically significant, men <65 years of age who also had a family history of prostate cancer had the highest mean levels of serum IGF-I and had a higher BMI than other age-family history subgroups. The association between PIK3C2B variants and prostate cancer among men <65 years of age who also had a family history of prostate cancer was not appreciably attenuated after adjustment for IGF-I levels or BMI, suggesting an alternative mechanism; however, a mechanism related to IGF-dependent PI3K signaling cannot be ruled out.

### Table 3. Risk of prostate cancer for genotyped or imputed SNPs located between 120.18 and 120.20 Mb in PIK3C2B

<table>
<thead>
<tr>
<th>SNP chromosomal order</th>
<th>Position</th>
<th>MAF*</th>
<th>$r^2$</th>
<th>Gene neighborhood</th>
<th>Status</th>
<th>Imputation quality</th>
<th>MACH $r^2$ range (mean)</th>
<th>OR† (95% CI)</th>
<th>$P_{\text{trend}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs4951389</td>
<td>−36904 G&gt;A</td>
<td>0.301</td>
<td>0.89</td>
<td>PIK3C2B, MDM4</td>
<td>Imputed</td>
<td>0.72–0.90 (0.84)</td>
<td>1.09 (1.03–1.15)</td>
<td>0.0005</td>
<td></td>
</tr>
<tr>
<td>rs10900594</td>
<td>−31199 C&gt;G</td>
<td>0.301</td>
<td>0.89</td>
<td>PIK3C2B, MDM4</td>
<td>Imputed</td>
<td>0.67–0.92 (0.84)</td>
<td>1.09 (1.03–1.15)</td>
<td>0.0006</td>
<td></td>
</tr>
<tr>
<td>rs11240751</td>
<td>−23119 G&gt;A</td>
<td>0.301</td>
<td>0.89</td>
<td>PIK3C2B</td>
<td>Imputed</td>
<td>0.78–0.95 (0.91)</td>
<td>1.09 (1.03–1.15)</td>
<td>0.0004</td>
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<td>0.279</td>
<td>0.99</td>
<td>PIK3C2B</td>
<td>Imputed</td>
<td>0.95–0.98 (0.97)</td>
<td>1.08 (1.03–1.14)</td>
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<td>—</td>
<td>PIK3C2B</td>
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<td>1.08 (1.03–1.14)</td>
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<td>1.07 (1.02–1.11)</td>
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<td>Imputed</td>
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<td>0.78–0.98 (0.93)</td>
<td>0.94 (0.90–0.98)</td>
<td>0.0083</td>
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</table>

*Minor allele frequency (MAF) and $r^2$ among white controls.
†OR per wild-type allele assuming a log-additive model. Adjusted by age, cohort (including country for EPIC), and ethnicity.
promote prostate carcinogenesis. The most significantly as-
could lead to a disruption in apoptotic activity that may
librium with the genotyped region. MDM4 plays a critical
ffects on another nearby gene. The p53 regulator
Koutros et al.
‡
Overall risk 7,900 8,476 1.08 (1.03–1.14) 0.0017 1.21 (1.09–1.34) 0.0003
Cohort
ACS 1,184 1,182 0.98 (0.86–1.11) 0.7240 1.31 (0.98–1.75) 0.0698
ATBC 959 828 1.18 (1.00–1.38) 0.0456 1.18 (0.80–1.74) 0.2740
EPIC 667 1,049 0.99 (0.84–1.16) 0.8680 1.15 (0.80–1.66) 0.4537
HPFS 650 650 1.11 (0.93–1.34) 0.2325 1.31 (0.92–1.89) 0.1393
MEC 2,296 2,254 1.10 (1.01–1.21) 0.0346 1.08 (0.90–1.30) 0.3962
PHS 993 1,098 1.21 (1.06–1.38) 0.0058 1.38 (1.05–1.82) 0.0206
PLCO 1,151 1,415 1.05 (0.93–1.19) 0.4193 1.33 (1.01–1.75) 0.0398 1.4
Ethnicity†
White 5,974 6,383 1.08 (1.02–1.15) 0.0059 1.24 (1.09–1.41) 0.0008
Hispanic 641 646 0.99 (0.86–1.15) 0.8914 1.13 (0.87–1.47) 0.3512
Japanese 461 470 1.11 (0.88–1.38) 0.3798 0.77 (0.34–1.73) 0.5272
Native Hawaiian 70 68 1.46 (0.85–2.51) 0.1698 0.45 (0.07–2.62) 0.3648 0.37
Age at case diagnosis (y)
<65 2,087 2,625 1.12 (1.02–1.23) 0.0158 1.47 (1.20–1.79) 0.0001
≥65 5,813 5,851 1.07 (1.01–1.13) 0.0220 1.13 (0.99–1.27) 0.0561 0.06
Aggressive disease
No 5,155 8,476 1.07 (1.01–1.13) 0.0163 1.20 (1.07–1.35) 0.0018
Yes 2,031 8,476 1.10 (1.01–1.19) 0.0168 1.16 (0.99–1.37) 0.0760
Family history of prostate cancer
No 5,097 5,515 1.04 (0.98–1.11) 0.1932 1.12 (0.99–1.28) 0.0755
Yes 817 525 1.19 (1.01–1.41) 0.0341 1.57 (1.11–2.23) 0.0114 0.02
<65 212 142 1.34 (0.94–1.92) 0.108 2.31 (1.07–5.02) 0.034
≥65 605 383 1.14 (0.93–1.40) 0.207 1.39 (0.93–2.07) 0.106 0.005
BMI (kg/m²)
<25 3,303 3,616 1.11 (1.02–1.20) 0.0128 1.17 (0.99–1.38) 0.0624
25–<30 4,159 4,440 1.07 (0.99–1.15) 0.0677 1.22 (1.05–1.43) 0.0121
≥30 1,447 1,054 1.03 (0.90–1.18) 0.6358 1.30 (0.98–1.71) 0.0690 0.46
P\text{trend} 0.003 0.0003
IGF-I (ng/mL)§
T1 758 974 1.04 (0.89–1.21) 0.6694 0.99 (0.73–1.35) 0.9567
T2 878 969 1.08 (0.92–1.26) 0.3508 1.19 (0.85–1.67) 0.3116
T3 911 970 1.11 (0.96–1.29) 0.1658 1.46 (1.04–2.06) 0.0287 0.80
P\text{trend} 0.101 0.075

*OR per wild-type allele assuming a log-additive model. Adjusted by age, cohort (including country for EPIC), and ethnicity where appropriate.
†Additionally adjusted for rs6594014 and rs11240748.
‡Minor allele frequency: Hispanic, 0.36; Black, 0.41; Japanese, 0.22; Native Hawaiian, 0.32.
§Tertile 1: < 147.3 ng/mL; Tertile 2: 147.3–204.0 ng/mL; Tertile 3: ≥ 204.1 ng/mL.

Alternatively, the observed association may be due to ef-
effects on another nearby gene. The p53 regulator MDM4 is
located within 30 kb of the observed PIK3C2B variants, and
SNPs located within this gene may be in linkage disequi-
librium with the genotyped region. MDM4 plays a critical
role in p53-dependent apoptosis and thus tumor suppres-
sion (39). Altered expression of MDM4 and, in turn, p53
could lead to a disruption in apoptotic activity that may
promote prostate carcinogenesis. The most significantly as-
sociated genotyped SNP in our study, rs7556371, is in
strong linkage disequilibrium with rs4245735, which has
been associated with MDM4 mRNA expression in lympho-
cytes (40). Thus, the observed association with prostate
cancer risk in this study may be due to altered mRNA ex-
pression of MDM4. Whereas this apoptosis regulatory
region may explain the observed effect, the well-described
interplay of the IGF axis with PI3K signaling and the ob-
served stratified associations with BMI and IGF-I levels
suggest that insulin signaling remains a possible mechanistic pathway.

We observed that the association between PIK2C2B variants was modified by age and family history of prostate cancer, but not by aggressive disease. Risk seemed to be equally related to aggressive and nonaggressive disease despite the tendency for familial, early-onset disease to present more aggressively in other studies (41–43). In our study, the percentage of men with aggressive disease among the familial, early-onset cases was comparable to that of the pooled population, and among those with a family history of disease, the association for rs7556371 persisted for nonaggressive prostate cancer. This suggests that the association we observed with PIK2C2B variants among men with a family history was not due to the fact that they had aggressive disease.

Despite numerous linkage studies, few genes have been identified as being associated with familial prostate cancer risk. The exploration of prostate cancer susceptibility loci identified from population-based genome-wide scans and family history of prostate cancer has not conclusively identified any loci that explain a substantial portion of inherited risk (44). In our study, younger men with a family history of disease who carried the variant allele at rs7556371 were 2.3 times more likely to develop prostate cancer. Genetic linkage analyses have observed significant linkage to chromosomes 1q23–25 and 1q42–43; however, 1q32, where PIK3C2B is located, has not been identified as a region of higher predisposition (45). Given that familial prostate cancer tends to be diagnosed at a younger age than sporadic prostate cancer and that the risk associated with rs7556371 was greatest among early-onset familial cases in our study, this region may be of interest for future exploration in familial studies. It is also possible that this observed subgroup association could be a false-positive finding given the small numbers of subjects diagnosed before age 65 years with a positive family history.

Strengths of our study include a large sample of cases and controls drawn from well-defined cohorts and a comprehensive SNP tagging approach. Further, mathematical imputation of all variants known to HapMap that were not directly genotyped provided a more comprehensive characterization of the genetic variation of the candidate genes in the PI3K pathway. Despite this, the precise genetic variant driving the association in PIK3C2B may not have been completely captured in our genotyping effort or by our imputation analysis. In addition, prostate-specific antigen values were not available for use in adjusting the analysis but likely would not have affected the ORs significantly. SNPs in PIK3C2B have not been associated with risk in individual genome-wide association studies to date; however, this is possibly due to the small effect size, which most genome-wide association studies are underpowered to detect. Although it is possible that our findings are false-positive results, the large sample size, the clustering of the significant variants, and the sustained significance after adjusting for multiple testing make this possibility less likely. Future genotyping for the statistically significant imputed SNPs in this analysis, including the PIK3C2B promoter region SNP rs11240751, is warranted and may yield additional insight.

In conclusion, this large pooled study has identified a cluster of variants in the class II PI3K gene PIK3C2B and the association with prostate cancer risk, especially among men with familial, early-onset disease. The precise genetic variant driving the association, however, is not clear and further studies are needed to replicate and refine this region of interest.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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References


Pooled Analysis of Phosphatidylinositol 3-Kinase Pathway Variants and Risk of Prostate Cancer


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