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A Polymorphism in the Promoter of *FRAS1* is a Candidate SNP Associated with Metastatic Prostate Cancer

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Data Use and Consent Statement

Data was made available by data use agreements and are stored locally by the relevant researchers. All patients were consented for germline SNP analyses and secondary use of the data was conducted with the approval by DF/HCC IRB.

Abstract

BACKGROUND—Inflammation and one of its mediators, NF-kappa B (NFκB), have been implicated in prostate cancer carcinogenesis. We assessed whether germline polymorphisms associated with NFκB are associated with risk of developing lethal disease (metastases or death from prostate cancer).

METHODS—Using a Bayesian approach leveraging NFκB biology with integration of publicly available datasets we used a previously defined genome-wide functional association network specific to NFκB and lethal prostate cancer. A dense-module-searching method identified modules enriched with significant genes from a genome wide association study (GWAS) study in a discovery dataset, Physicians' Health Study and Health Professionals Follow-up Study (PHS/HPFS). The top 48 candidate single nucleotide polymorphisms (SNPs) from the dense-module-searching method were then assessed in an independent prostate cancer cohort and the one SNP reproducibly associated with lethality was tested in a third cohort. Logistic regression models evaluated the association between each SNP and lethal prostate cancer. The candidate SNP was assessed for association with lethal prostate cancer in six of 28 studies in the PRACTICAL Consortium where there was some medical record review for death ascertainment which also had SNP data from the ONCOARRAY platform. All men self-identified as Caucasian.

RESULTS—The rs1910301 SNP which was reproducibly associated with lethal disease was nominally associated with lethal disease (OR=1.40; P=0.02) in the discovery cohort and the minor allele was also associated with lethal disease in 2 independent cohorts (OR=1.35; P=0.04 and OR=1.35; p=0.07). Fixed effects meta-analysis of all three cohorts found an association: OR = 1.37 (95% CI: 1.15–1.62, P-value=0.0003). This SNP is in the promoter region of *FRAS1,* a gene involved in epidermal-basement membrane adhesion and is present at a higher frequency in men with African ancestry. No association was found in the subset of studies from the PRACTICAL consortium studies which had a total of 106 deaths out total of 3,263 patients and a median follow-up of 4.4 years.

CONCLUSIONS: Through its connection with the NFκB pathway, a candidate SNP with a higher frequency in men of African ancestry without cancer was found to be associated with lethal prostate cancer across 3 well-annotated independent cohorts of Caucasian men.

Keywords

Single nucleotide polymorphisms; SNPs; African ancestry; nuclear factor kappa B

Introduction

Each year there are about 1.4 million newly diagnosed cases of prostate cancer and more than 366,000 deaths worldwide[1]. Some patients have indolent localized disease that does not require treatment, while others present with or develop metastatic disease that responds poorly to therapy[2,3]. Clinicopathologic staging can identify patients at higher risk of relapsing with metastatic disease[4,5] and treatment of intermediate and high-risk localized disease decreases prostate cancer deaths[6–9]. Biomarkers such as PTEN loss and gene expression signatures also provide prognostic information[10,11]. Epidemiological and

biological studies have implicated aberrant metabolism[12], inflammation[13] and inherited genetic exome[14] and SNP variants[15] with more advanced prostate cancer.

Nuclear factor kappa B (NFκB) is a transcription factor that controls inflammation and can either promote cancer progression or cancer cell death[16]. NFκB activation can promote proliferation, development of metastases, and evasion of apoptosis in prostate cancer[17–19]. Biomarkers of NFκB activation in localized disease are emerging as a possible strategy to identify patients with a higher risk of relapse with metastases after localized therapy[20–24].

The identification of biomarkers related to NFκB activation that can predict aggressive prostate cancer may identify patients who are at risk of relapse after localized therapy and may benefit from adjuvant systemic therapy to prevent relapses[16,25]. These biomarkers may help identify men who are otherwise candidates for active surveillance but need immediate intervention. Although many SNPs predisposing to risk of prostate cancer have been identified, only a limited number of SNPs with some evidence of possible association with lethal prostate cancer have been identified to date[26,27]. Moreover, a germline biomarker rather than tissue-based biomarker may help identify men at risk of significant prostate cancer who should be screened for prostate cancer. To that end, we leveraged our previous work which integrated publicly available genomic datasets using a Bayesian approach and defined an NFκB-network that was enriched in patients with lethal prostate cancer after a prostatectomy[28,29]. This network was then used to interrogate a prostate cancer GWAS to identify SNPs associated with NFκB-activation and lethal prostate cancer.

Methods:

SNP Selection Using a Bayesian-Based Analysis Leveraging NFκB Biology

Using multiple publicly available data sets and the network approach previously described by our team[28,29], we defined a genome-wide functional interaction network specific to the NFκB-pathway and metastatic disease or prostate cancer death, (“lethal cases”) compared to patients with prostate cancer who had not developed radiographic evidence of metastases on computerized tomography or technetium bone scan imaging (conventional scans) at least ten years after their diagnosis (“non-metastatic cases”). The genome-wide functional interaction network identified 8,154,133 high-confidence protein-protein functional associations. The dense-module-searching method, dmGWAS[30], was then used to define a candidate subnetwork of interacting genes related to both (i) the NFκB pathway and (ii) lethal prostate cancer. We then searched the NFκB interaction network for modules enriched with genes represented by SNPs with the lowest additive model p-value for lethal disease (Supplementary Table 1) with PHS/HPFS GWAS data[31] as the discovery set. After assigning each SNP on the Affymetrix 5.0 chip to a gene if the SNP is located within 20kb of the gene, a single SNP with the lowest additive model p-value for lethal disease was selected to represent each of 16,387 genes. The SNP-gene pairs were weighted by the GWAS p-values of the SNP during dense-module-searching. We arbitrarily limited our analysis to 50 genes included in the top 26 modules out of 10,171 valid modules with the highest normalized scores. Of the 52 SNPs (Supplementary Table 1) used to represent the genes in the selected subnetwork (two genes were represented by two SNPs with the same GWAS p-values each), four failed probe design using the Sequenom platform in the Gelb

Center/ECOG cohort detailed below. This resulted in 48 SNPs to test in the first independent cohort (Gelb Center/ECOG cohort).

The PHS/HPFS GWAS was conducted on self-identified Caucasian men including 196 lethal and 368 indolent cases. Endpoints were confirmed by medical record and death certificate review by a physician.

Patients and Genotyping Sequencing Methods in Independent Cohorts

The candidate SNPs were then tested in an independent cohort Gelb Center/ECOG patients of self-identified Caucasian men who provided consent. Using the Gelb Center prostate cancer hospital registry database at Dana-Farber Cancer Institute[32] we identified an independent cohort of 254 self-identified men with a blood sample available for analysis and a history of localized prostate cancer treated with curative intent with surgery or radiation and had not developed radiographic evidence of metastatic disease with a median follow-up of 8.4 years. The patients from the ECOG cohort of 256 self-identified Caucasian men with a blood sample available all had documented metastatic disease (relapsed post-local therapy or *de novo* metastatic)[3] as determined by eligibility at time of enrolment on the therapeutic clinical trial. Patients included in this analysis provided consent and institutional review board approval were obtained for all studies conducted[3]. All DNA samples were extracted from peripheral whole blood using QIAamp DNABlood mini kit (QIAGEN Inc, Valencia, CA). Genotyping was completed at the same time using Sequenom iPLEX matrix-assisted laser desorption/ionization (MALDI)-time of flight mass spectrometry technology (Carlsbad, CA). SNP assays were combined into four multiplex pools in 384-well format. Approximately 5% of samples were randomly selected and genotyping duplicated for quality control. Concordance rate for duplicate genotyping was 100%. Call rate overall was greater than 99%.

The SNP which was found to be associated with lethal disease in the Gelb Center/ECOG cohort was then also tested in The Fred Hutchinson Cancer Research Center (FHCRC) study. These patients were previously enrolled in population-based prostate cancer case–control studies[33,34]. The subset of self-identified Caucasian patients included (n = 1,548) for these analyses were diagnosed with histologically confirmed adenocarcinoma of the prostate using the Seattle-Puget Sound Surveillance, Epidemiology, and End Results (SEER) Program cancer registry. Prostate cancer recurrence status was determined from prospectively collected information from follow-up surveys that were completed by patients in 2004–2005 and in 2010–2011, review of medical records, and/or physician follow-up as needed. Metastatic progression was confirmed by positive bone scan, MRI, CT, or biopsy. Cause-specific deaths were ascertained from the SEER registry, which links quarterly with the Washington State Vital Statistics Database and annually with the National Death Index. Endpoints were also ascertained by medical record and death certificate review by a study physician. There were 570 cases who had no evidence of recurrence or progression during a follow-up period of at least 10 years after diagnosis indolent and 104 cases of metastatic disease or death from prostate cancer. A custom designed TaqMan SNP genotyping assay (Applied Biosystems, Foster City, CA) was used for genotyping on the ABIPrism 7900HT sequence detection system according to the manufacturer’s instructions. The study was

approved by the institutional review board of the Fred Hutchinson Cancer Research Center, and written informed consent was obtained from all participants. Genotyping was approved by the institutional review board of the Intramural Program for the National Human Genome Research Institute and was done using Affymetrix Human Mapping 500K array (Affymetrix, Inc).

The frequency of the SNP which was reproducibly associated with lethal prostate cancer from the three first cohorts was then assessed for frequency by ethnicity in men with and without prostate cancer in the PRACTICAL consortium[35] who had SNP data from the ONCOARRAY platform. The individual PRACTICAL study principal investigators (N=63) were queried to identify studies which had some physician review of medical records in addition to use of use national registries for ascertaining cause of death. Six studies were identified out of 28 studies where investigators provided process for death ascertainment with a total of 3,263 patients and a total of 106 deaths and a median follow-up of 4.4 years.

Statistical Methods

Odds ratios (OR) and 95% confidence intervals (CI) were estimated using univariable logistic regression, assuming an additive model for the association between each SNP and lethal disease and modeled as a continuous variable (0, 1 or 2 copies), assuming constant effect going from 0 to 1 and from 1 to 2. Having identified the candidate SNP in the first three well annotated cohorts, a fixed effects meta-analysis was performed to obtain an overall OR and 95% CI for the first three cohorts. The impact of the SNP of interest on overall survival for the ECOG clinical trial data-set, the only data-set where outcome to androgen deprivation therapy data was known, was estimated using the Kaplan-Meier method and differences between survival distributions was tested using the log-rank test.

Results:

The GWAS results from the PHS/HPFS discovery cohort has been previously described[31]. Table 1 details patient characteristics of the two test cohorts used for assessment of reproducibility. Figure 1 shows the subnetwork consisting of the 26 modules and the top 48 SNPs from the discovery cohort are listed in Supplementary Table 1.

One of these 48 SNPs, rs1910301, was replicated to be associated with lethal disease in the independent cohort of GC/ECOG patients, OR: 1.35, (95%CI:1.02–1.80, P-value 0.04). In the original PHS/HPFS GWAS analysis, the rs1910301 SNP had an OR of 1.40 (95%CI: 1.05–1.84, P-value = 0.02) for lethal disease. This SNP was the only SNP taken forward to be tested in the FHCRC cohort and had an OR for lethal disease of 1.35 (95%CI: 0.97–1.85, p=0.07). A fixed effects model of all three cohorts resulted in a meta-analysis OR for lethal prostate cancer of 1.37 (95% CI:1.15–1.62; p=0.0003). We also explored whether this SNP was associated with a poorer response to androgen deprivation therapy using the using the overall survival data from the parent ECOG trial[3]. The presence of at least one copy of allele A was not significantly associated with shorter overall survival OS [HR=0.87 (95% CI 0.56–1.34) p=0.52], (Figure 2).

Using the PRACTICAL consortium data the genotype frequency was then assessed in men with prostate cancer (cases) and no cancer (controls) and reported by ethnicity (N=102,026 total, 91,309 European descent, 2,437 Asian descent and 8,280 African descent). The allele frequency was 23% for Europeans and Asians and 60% for men of African descent (Table 2B). Of the 6 studies with 3,263 patients with a median follow-up of 4.4 years we identified 258 patients alive for more than 10 years and compared them to the 106 total prostate cancer deaths resulting in an OR of 0.95 (95% CI:0.65–0.38, p=0.80). Given there were only 106 (3.2%) prostate cancer deaths recorded in 3,263 patients consistent with a short median follow-up of 4.4 years, this cohort was deemed too immature for assessment of prostate cancer death and inclusion in the meta-analysis.

Biological plausibility of the rs1910301 finding was assessed by querying genomic databases and revealed it is in the promoter of *FRAS1* (Supplemental Figure 1A). We further explored the regional 3D genomic structure surrounding rs1910301 using publicly available Hi-C data from human fibroblast IMR90 cell profiles[36,37] This SNP was also found to reside in a topologically associating domain (TAD) of 8 genes including *MRPL1* (Figure 3), a gene highly represented in the top gene modules as well as CNOT6L. The SNP also has some conservation across species and is in a region with DNAse hypersensitivity (Supplemental Figure 1B). *The Human Protein Atlas* reported FRAS1 immunohistochemistry protein staining from 10 prostate cancer samples: 0-high 3-medium; 4-low, 3-none. The TCGA data noted patients in the lower quartile had a better overall survival than patients with higher expression (Supplemental Figure 2). Expression quantitative trait loci (eQTL) analysis using the GTEx Portal revealed rs1910301 impacts CNOT6L RNA expression including prostate tissue (Supplemental Figure 3).

Discussion:

This unique approach to identifying biomarkers of NFκB activation identified SNP rs1910301 in promoter region of *FRAS1* on chromosome 4q21, as a candidate SNP possibly associated with a 37% increase in risk of development of metastatic disease or death from prostate cancer in three well annotated and mature cohorts. Having identified this candidate SNP in the meta-analysis of the first three cohorts, we were unable to confirm the association in the PRACTICAL cohort. This was despite attempts to define a subset with some degree of medical review of causes of death. After the efforts to define a cohort for analysis, we were only able to define a cohort with a median follow-up 4.4 years and 3.2% prostate cancer deaths. The PRACTICAL consortium is a very robust dataset for SNPs for risk of prostate cancer[15] but longer follow-up is needed for prostate outcome data.

To date, while numerous SNPs from GWAS have been associated with risk of developing prostate cancer, there has been limited success in reproducibly finding SNPs associated with lethal prostate cancer. This may be partly due to relatively few events of metastatic disease in these studies resulting in limited statistical power. Previously rs5993891, in *ARVCF* on chromosome 22q11 has been found to be associated with a 48% reduction in risk of prostate cancer specific mortality in a meta-analysis of four cohorts[38]. In another meta-analysis of seven cohorts with 12,082 patients with 1,544 prostate cancer deaths after adjustment for clinicopathological factors, rs2308327 in the *MGMT*, rs2070874 in *IL4* and in rs2494750

in *AKT1* were associated with risk of prostate cancer specific mortality; and in a cohort of men with an inherited susceptibility to the disease, prostate cancer specific mortality was associated with rs635261 at *RNASEL*; rs915927 in *XRCC1*; rs2494750 in *AKT1*[27,39]. In a case-only GWAS of 12,518 prostate cancer cases, two loci were associated with higher Gleason score (rs35148638 in *RASA1*; rs78943174 in *NAALADL2*)[40].

To address the potential for false negative findings from GWAS level statistics, we used a Bayesian-based analysis leveraging the biology connecting NFκB to lethal prostate cancer and the dmGWAS analytical approach were used. This was a parsimonious biology based-approach to select candidate SNPs. To further minimize false positive findings, we assessed for reproducibility across cohorts and saw the rs1910301 SNP is consistently associated with approximately a 37% greater risk of having lethal prostate cancer disease across three cohorts of Caucasian men with prostate cancer. In short, by using a Bayesian approach and showing reproducibility across three cohorts we have been able to identify one of the few candidate SNPs that are possibly associated with lethal disease (metastatic disease or prostate cancer death).

Notably, the frequency of rs1910301 differs across ethnic groups in the 1000 Genomes Project and was more frequent in patients with African ancestry (allele A frequency, 64%) in comparison to a population of European ancestry without prostate cancer (23%). The new data presented in this paper from the three cohorts for assessment of association with lethal prostate cancer is restricted to men of European ancestry (N=1,748) and the allele frequency of the risk carrying variant, the A-variant, was identical (23%) to that reported by 1,000 Genomes Project. Furthermore, this was the same frequency in the PRACTICAL consortium as it was 23% in patients with prostate case and 23% in the controls with no prostate cancer of European descent (N=91,309). Patients of Asian descent had the same A allele frequency in men with prostate cancer (24%) and no prostate cancer (23%). The A allele frequency in the 8,280 men of African descent confirmed the frequency as 60% in men with prostate cancer and 59% in men without prostate cancer. The clinical implications of racial/ethnic allele frequency heterogeneity are not well defined. Emerging evidence points to an association between racial/ethnic differences in SNP frequency with differential treatment response/relapse risk in localized prostate cancer[41,42], though data remains limited. It is interesting to note that rs1910301 is found at a higher frequency in African-Americans – a group of men who after even accounting for socio-economic factors, still have a higher rate of metastatic disease[43]. Moreover, African-Americans treated with androgen deprivation therapy alone have the same benefit with androgen deprivation therapy as Caucasians[43] and it is notable we did not find an association of this SNP with poorer overall survival in the E3805 analysis in Caucasian men treated with androgen deprivation alone.

The biological plausibility of this SNP impacting the biology of prostate cancer development can be seen by the genes it is associated with topographically. Reassuringly, rs1910301 is located in the promoter region of a gene that may be relevant to metastatic disease biology, *FRAS1.* Together with *FREM2*, *FRAS1* forms a gene unit that regulates epidermal-basement membrane adhesion and cell migration[44]. *FREM2* is an NFκB regulated gene and mutations in *FREM2* and *FRAS1* are associated with the Fraser syndrome – a congenital syndrome with craniofacial, urogenital and respiratory system abnormalities[44]. In cancer

cells *in vitro*, silencing of *FRAS1* leads to decreased ability of non-small lung cancer, A549 cells to migrate and invade[45]. In addition, *FRAS1* was found to be more frequently mutated in metastatic breast cancer than primary breast cancer[46]. It is therefore possible that a SNP that alters *FRAS1* activity causes dysfunction of *FRAS1-FREM2* gene unit and increases metastatic potential. We also found rs1910301 to reside in a topologically associating domain (TAD) with *MRPL1* (Figure 3), a gene highly represented in the top gene modules, potentially helping to explain the identification of rs1910301 through the dmGWAS approach. MRPL1 encodes the 39S subunit protein that belongs to the L1 ribosomal protein family and altered function may play a role in prostate cancer progression through dysregulation of translation of proteins – for example – of cellular adhesion or cell cycle[47]. The finding that the rs1910301 SNP has some conservation across species and is in a region with some DNAse hypersensitivity also adds biological plausibility. The latter suggests the SNP is in an area where the chromatin is accessible and functionally related to transcriptional activity as it is amenable for the protein binding including transcription factors. The observation using the expression quantitative trait locus showed an association between rs1910301 and CNOT6L is also notable as this is one of the 8 genes found to reside in the topologically associated domain of rs1910301. CNOT6L is cytoplasmic deadenylase with 3-prime-to-5-prime exoribonuclease activity[48].

However, it is recognized that this work is hypothesis generating and fine-mapping and mechanistic studies to define the exact gene and the biological basis for the possible association of rs1910301 with lethal prostate cancer are needed. It is unknown if rs1910301 is pathogenic or in linkage disequilibrium with the actual gene driving the metastatic biology. Other limitations of this study include not inferring ancestry from GWAS data. We also only chose a gene if the SNP was located within 20kb of the gene and thus excluding long regulatory-range elements.

In summary, the rs1910301 SNP was identified by a Bayesian interrogation of GWAS data focused on the cancer-promoting NFκB activation network and reproducibly had an OR for metastatic prostate cancer of about 1.37 across three independent cohorts. The association was not confirmed in the PRACTICAL consortium with limited follow-up for cancer outcomes. The higher frequency of the SNP in the African American population may be a clue to the biology underlying the greater propensity for metastatic disease in this patient population. Based on this data, biological mechanistic work to define the exact biological underpinnings for the association and assessment in prospective trials of localized disease with detailed endpoint ascertainment and accounting for clinico-pathological and treatment variables is needed to determine whether it is a biomarker that can be used in the clinical setting such as identifying men who should be (i) targeted for screening or (ii) with low risk disease who should not be managed with active surveillance or (iii) who need adjuvant systemic therapy.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Conflict of Interest Statement:

Drs Sweeney, Mucci, Lee, Börnigen, and Huttenhower hold a patent for rs1910301 as a biomarker in prostate cancer. No other potential conflicts were disclosed by the other authors.

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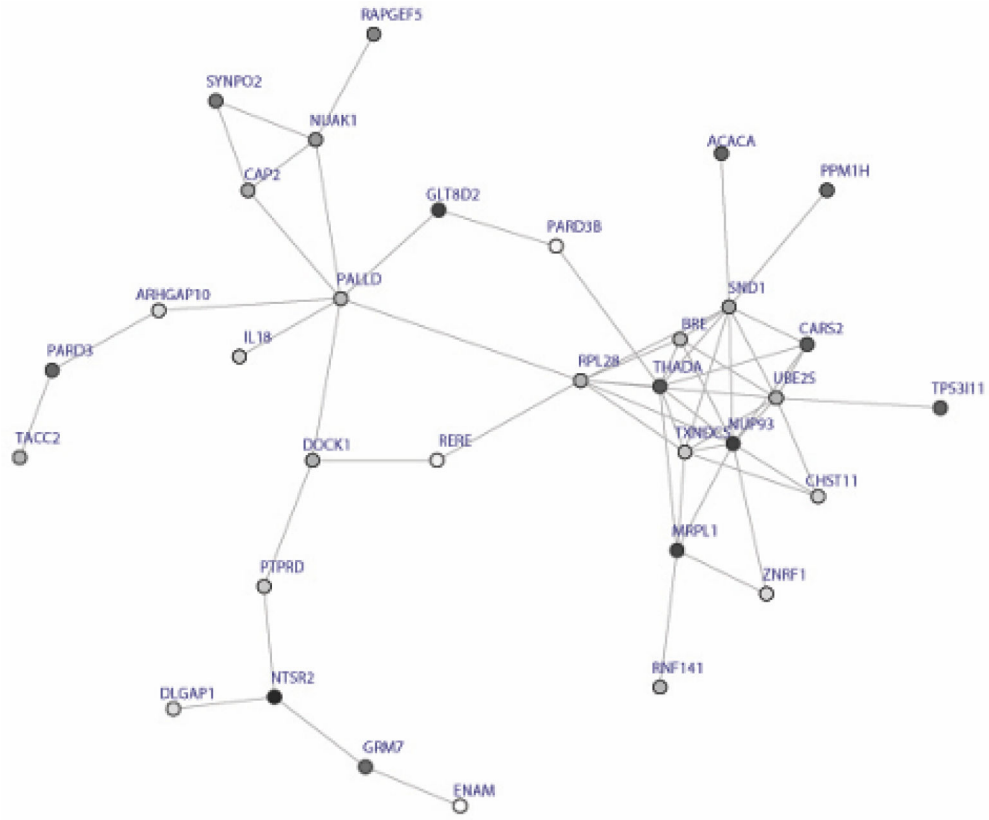

Figure 1:
Modules identified by dmGWAS, a dense module searching method. The subnetwork includes 50 genes and 109 interactions.

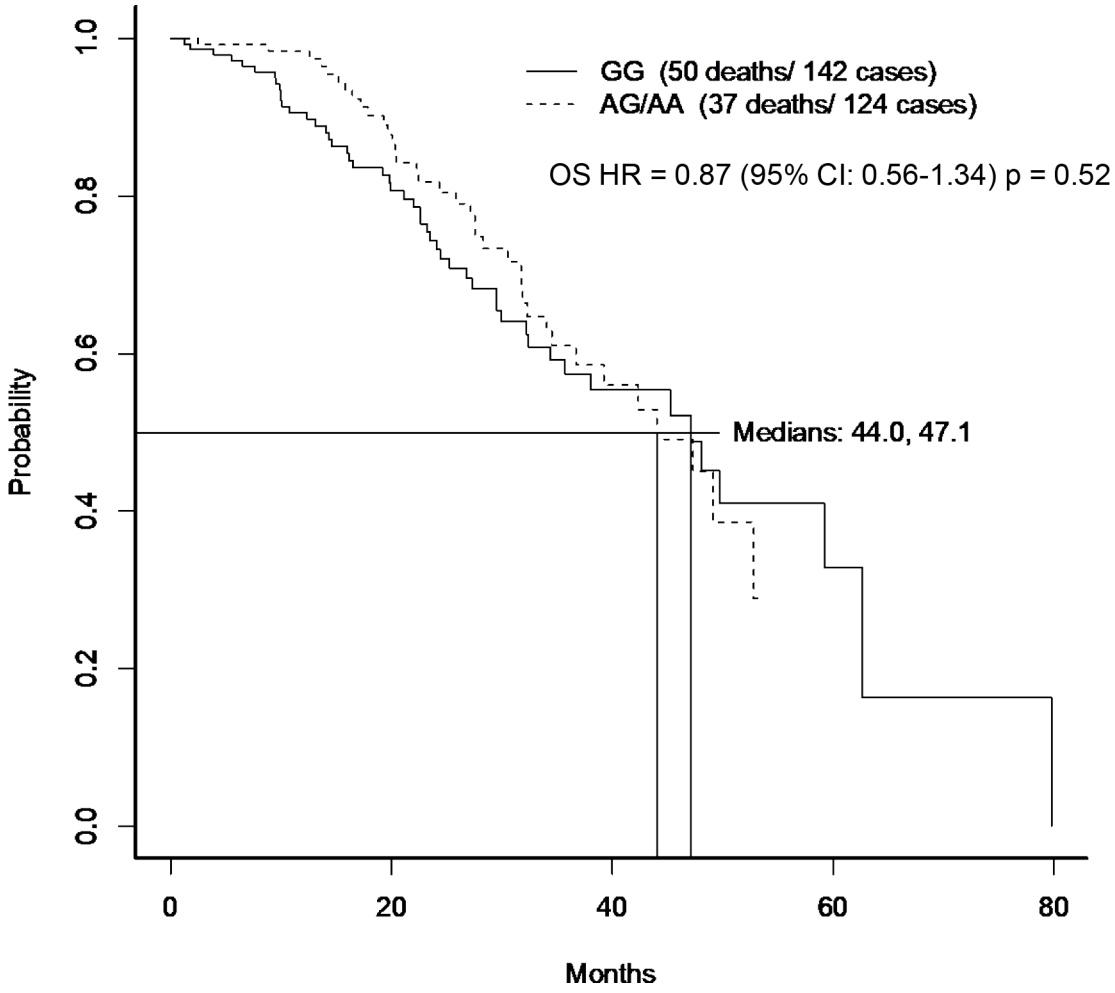

Figure 2:
Kaplan-Meier curves for overall survival by genotype for E3805 patients on androgen deprivation therapy alone with genotype data.

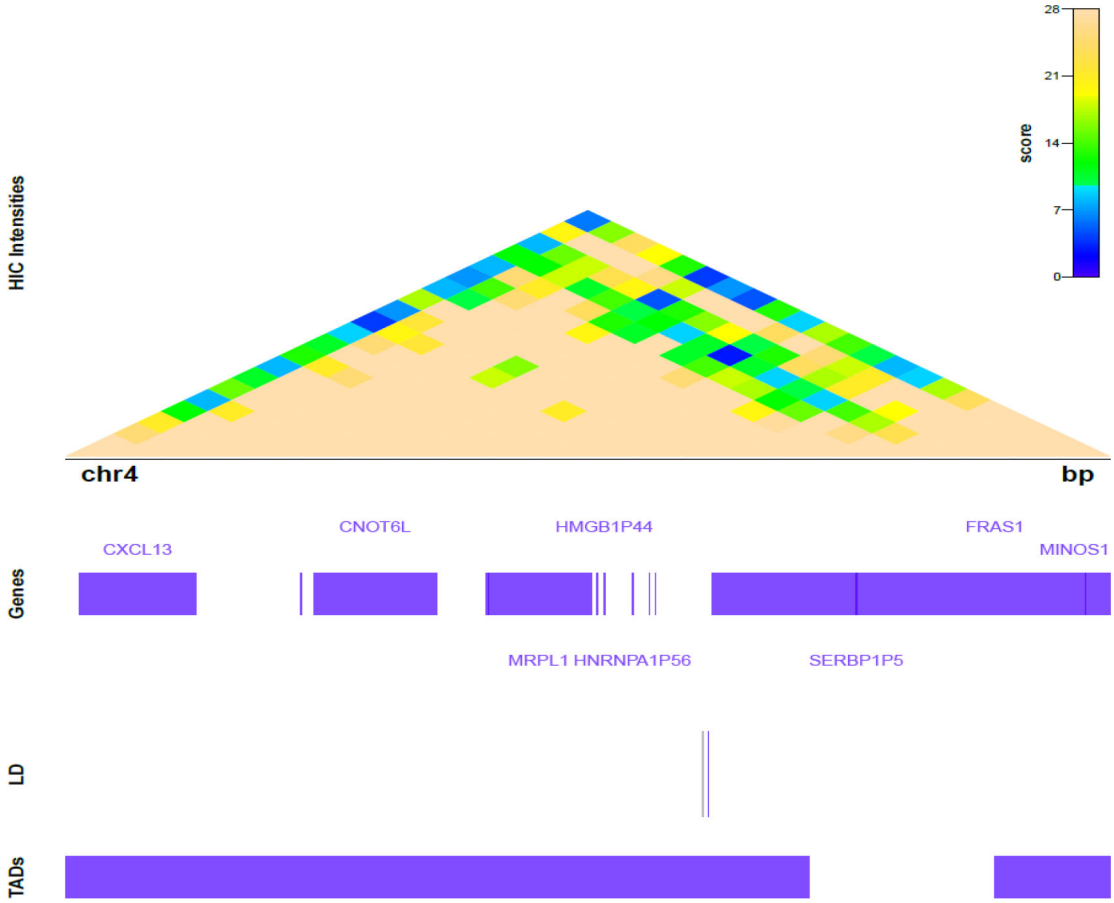

Figure 3:

Topographical map. The SNP rs1910301 was found to reside in a topologically associating domain (TAD) with 8 genes.

Table 1:

Patient Characteristics of the Test Cohorts.

Variable	Category	ECOG Metastatic	DFCI GC No recurrence[1]	FHCRC Metastasis or death	FHCRC No adverse outcomes[2]
Total		256	254	104	570
Age	Mean (SD)	61.0 (8.5)	60.4 (7.8)	58.8 (8.2)	59.6 (7.2)
	Median (Q1,Q3)	61 (55,67)	61 (55,66)	59.0 (53.0, 64.0)	59.0 (54.0, 64.0)
	[Min, Max]	[38,90]	[40,81]	[42.0, 74.0]	[35.0, 74.0]
	Freq. of Missing	4	1	0	0
Local therapy	None	181 (71)	31 (12)	32 (30.8)	48 (8.4)
	Prostatectomy	50 (20)	177 (70)	41 (39.4)	367 (64.4)
	Definitive RT	25 (10)	46 (18)	31 (29.8)	151 (26.5)
	Other	0	0	0	4 (0.7)
	Unknown/Missing	0	0	0	0
Adjuvant hormonal therapy	No	242 (94.5)	247 (97)	89 (85.6)	544 (95.4)
	Yes	14 (5.5)	7 (3)	15 (14.4)	26 (4.6)
	Unknown/Missing	0	0	0	0
Clinical stage at diagnosis	Local (T1,T2,T3)	62 (25.3)	225 (100)	73 (70.2)	565 (99.3)
	Regional (T4,N1)	14 (5.7)	0 (0)	9 (8.7)	5 (0.7)
	Metastatic (M1)	169 (69.0)	0 (0)	22 (21.2)	0
	Unknown/Missing	11	29	0	0
Gleason score	<=6	18 (7.6)	148 (59)	29 (28.4)	356 (62.5)
	7	61 (25.7)	81 (32)	37 (36.3)	185 (32.5)
	>=8	158 (66.7)	21 (8)	36 (35.3)	29 (5.1)
	Unknown/Missing	19	4	2	0
Length of follow-up (years from diagnosis)	Mean (SD)	4.3 (2.9)	9.0 (3.5)	9.7 (5.7)	14.7 (4.0)
	Median (Q1,Q3)	3.8 (2.3,5.3)	8.4 (6.3,11.3)	8.9 (5.1, 13.2)	13.1 (11.6, 19.3)
	[Min, Max]	[0.3,15.2]	[0.4,22.2]	[1.1, 22.4]	[10.1, 22.9]
	Freq. of Missing	3	1	0	0

[1] No evidence of recurrence on conventional scans after curative therapy for localized disease.

[2] No evidence of recurrence/progression and survival time of ≥10 years.

Table 2A:

Odds ratio (OR)[1] and p-value for association between SNP rs1910301 and risk of metastatic disease or prostate cancer death in three independent cohorts

Cohort	OR for metastatic disease	P value
PHS/HPFS	1.40 (1.05 – 1.84)	0.02
DFCI GC-EA	1.35 (1.02 – 1.80)	0.04
FHCRC	1.35 (0.97 – 1.85)	0.07
Meta-analysis of 3 cohorts [fixed effects model]	**1.37 (95% CI: 1.15–1.62)**	**0.0003**

PHS/HPFS: Physicians' Health Study and Health Professionals Follow-up Study; DFCI GC: Dana-Farber Cancer Institute Gelb Center; EA: The ECOG-ACRIN Cancer Research Group; FHCRC: Fred Hutchinson Cancer Research Center.

[1]OR modeled as ordinal for the minor allele

Table 2B:

Frequency of SNP rs1910301 across the study populations

		GG	AG	AA	A Allele Frequency
PHS/PFS [1]	Mets or PrCa[2] death (event)	106 (32.4)	69 (34.3)	21 (58.3)	28.3%
	No Mets or prostate cancer death	221 (67.6)	132 (65.7)	15 (41.7)	22.0%
ECOG [1]	Mets or PrCa death (event)	140 (46.4)	96 (55.2)	20 (58.8)	26.5%
Gelb Center [1]	No Mets or PrCa death	162 (53.6)	78 (44.8)	14 (41.2)	20.8%
Fred Hutch [1]	Mets or PrCa death	57 (13.7)	37 (17.1)	10 (23.3)	27.4%
	No Mets or PrCa death	358 (86.3)	179 (82.9)	33 (76.7)	21.4%
PRACTICAL		GG	AG	AA	
European descent	Cases	32451 (58.8)	19585 (35.5)	3126 (5.7)	23.4%
	Controls	21085 (58.3)	12994 (35.9)	2068 (5.7)	23.7%
Asian Descent	Cases	707 (57.6)	447 (36.4)	74 (6.0)	24.2%
	Control	702 (58.0)	437 (36.1)	70 (5.8)	23.9%
African Descent	Cases	721 (16.9)	1943 (45.5)	1603 (37.6)	60.3%
	Controls	685 (17.1)	1879 (46.8)	1449 (36.1)	59.5%

[1] All self-identified Caucasian

[2] PrCa: prostate cancer