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Prostate Cancer

A Genetic Score Can Identify Men at High Risk for Prostate Cancer Among Men With Prostate-Specific Antigen of 1–3 ng/ml

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Abstract

Background: The diagnostic performance of a genetic score based on single nucleotide polymorphisms (SNPs) is unknown in the prostate-specific antigen (PSA) range of 1–3 ng/ml. A substantial proportion of men in this PSA span have prostate cancer (PCa), but biomarkers to determine who should undergo a prostate biopsy are lacking.
Objective: To evaluate whether a genetic risk score identifies men in the PSA range of 1–3 ng/ml who are at higher risk for PCa.
Design, setting, and participants: Men aged 50–69 yr with PSA 1–3 ng/ml and without a previous prostate biopsy were selected from the STHLM2 cohort. Of 2696 men, 49 SNPs were genotyped, and a polygenic risk score was calculated. Of these men, 860 were invited according to risk score, and 172 underwent biopsy.
Outcome measurements and statistical analysis: The risk of PCa was assessed using univariate and multivariate logistic regression analysis.
Results and limitations: PCa was diagnosed in 47 of 172 participants (27%), with Gleason sum 6 in 36 of 47 men (77%) and Gleason sum $\geq$7 in 10 of 47 men (21%); one man had intraductal cancer. The genetic score was a significant predictor of a positive biopsy ($p = 0.028$), even after adjusting for PSA, ratio of free to total PSA, prostate volume, age, and family history. There was an increase in the odds ratio of 1.60 (95% confidence interval, 1.05–2.45) with increasing genetic risk score. The absolute risk difference of positive biopsy was 19 percentage points, comparing the high and low genetic risk group (37% vs 18%).
Conclusions: A risk score based on SNPs predicts biopsy outcome in previously unbiopsied men with PSA 1–3 ng/ml. Introducing a genetic-based risk stratification tool can increase the proportion of men being classified in line with their true risk of PCa.

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1. Introduction

Prostate-specific antigen (PSA) testing has been used since the 1990s to identify men at risk of prostate cancer (PCa) [1]. Numerous studies have addressed the limited diagnostic accuracy of PSA, yet it remains the only widely adopted biomarker for PCa [2,3]. Thompson and coworkers have illustrated the operating characteristics of PSA and the frequent finding of PCa among men with low PSA levels in the Prostate Cancer Prevention Trial, reporting a 17–24% cancer detection rate in the PSA range of 1–3 ng/ml [4,5]. Following a vigorous scientific discussion, many urologists today use a cutoff value for biopsy of 2–3 ng/ml, taking multiple factors—including free and total PSA, patient age, PSA velocity, PSA density, family history, ethnicity, prior biopsy history, and comorbidities—into account [6,7].

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Even then, the sensitivity for both PCa in general and high-grade cancer is inadequate. To further increase the predictive performance of PCa testing, several additional biomarkers have been suggested, none of which have reached widespread clinical use.

Twin studies indicate that genetic factors account for ≥42% of the etiologic factors in PCa, the highest among common cancers [8]. Despite this finding, individual genes with high penetrance have not been identified. There are at least 50 low-penetrant risk alleles (single nucleotide polymorphisms [SNPs]) that have been associated with risk for PCa (Table 1). In the STHLM1 study, it was shown that a combined genetic risk score based on 35 SNPs can be used in a clinical setting to avoid unnecessary biopsies in men with a PSA value between 4 and 10 ng/ml. Recently, the genetic score was shown to improve PCa risk prediction in a rebiopsy setting among men with PSA >2.5 ng/ml in the Reduction by Dutasteride of Prostate Cancer Events (REDUCE) study [9]. Despite these studies, the clinical utility of a genetic risk score combining SNPs identified in genomewide association studies (GWASs) has been questioned, both in general for complex diseases and in PCa [10].

We hypothesized that the genetic score can be used to identify men at a higher risk for PCa among men with low PSA. In this study, we invited men with PSA between 1 and 3 ng/ml at inclusion with no history of prior biopsy of the prostate or known PCa to evaluate how their genetic risk score influences the detection rate of PCa.

2. Material and methods

2.1. Study population

Men who were referred for PSA testing in laboratories connected to the Karolinska University Laboratory and Aleris Medilab in Stockholm County, Sweden, between November 1, 2010, and September 1, 2012, were invited to the population-based cohort STHLM2 at the blood-sampling visit. The men who agreed to participate were asked to donate four test tubes of blood and a urine sample. A total of 24 642 men were included during the 22-mo study period, compared with approximately 53 000 men aged 50–69 yr tested in Stockholm during 2011 [11].

Whole blood for plasma and DNA was collected in separate ethylenediaminetetraacetic acid tubes without gel. Study samples were transported to KI Biobank, Karolinska Institutet, within 24 h. After centrifugation, plasma was aliqouted and stored at −80 °C. DNA was extracted from whole blood using the magnetic bead separation method (Chemagen Inc.) and was stored at −80 °C. PSA test data, biopsy records, and PCa records were retrieved from the database STHLM0. This database consists of all men in Stockholm County who have had at least one PSA analysis since 2003. Pathology reports on all prostate biopsy results were retrieved through pathology laboratories in Stockholm. Data have been matched against the National Cancer Registry and the National Prostate Cancer Registry to obtain cancer status and clinical information. STHLM0 is updated regularly and has been described in detail by Nordström et al. [11].

In total, there were 2696 men aged 50–69 yr without records of previous prostate biopsy or PCa with PSA levels of 1–3 ng/ml at inclusion in STHLM2. These men defined the study population (Fig. 1).

2.2. Single nucleotide polymorphism selection and genotyping

We selected all SNPs reported to be associated with PCa risk from published GWAS studies (until October 2012) and replicated in at least one large independent population. We identified 50 relevant SNPs, further described in Table 1. Markers were genotyped using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry based on allele-specific primer extension with iPLEX chemistry (SEQUENOM 96 Inc., San Diego, CA, USA). Forty-nine SNPs were genotyped in the entire sample, with a 98.7% average success rate; rs13385191 failed.

2.3. Genetic risk score

We applied a genetic risk model, as previously described [12]. Briefly, for each man we created a genetic risk score by summing the number of risk alleles (zero, one, or two) at each of the 49 SNPs multiplied by the logarithm of that SNP's odds ratio (OR) and divided by the total number of called SNPs for each individual man. Men were assigned low, intermediate, or high risk when they had a genetic risk score in the <10th, 10th–89th, or ≥90th percentile, respectively. Figure 2 describes the distribution of risk scores among STHLM2 participants.

The risk scores and PSA levels were blinded to the urologist and pathologist performing biopsies and prostate biopsy evaluation.

2.4. Sampling and biopsy procedure

Considering the medical adverse effects of prostate biopsy, we restricted the study size to meet requirements suggested by power calculations regarding the main end point. Hence, 192 biopsy occasions were arranged for invitees to book independently.

Guided by previous response rates, 860 genotyped men were randomly drawn from the study population by genetic risk score, with oversampling of the extreme deciles. These men were invited, with 322 (37.4%) from the high-risk group, 323 (37.6%) from the low-risk group, and 215 (25%) from the intermediate-risk group.

There were 192 scheduled appointments for biopsy; 50, 79, and 43 participants from the low-, intermediate-, and high-risk groups, respectively, underwent prostate biopsy. Six men did not show up for biopsy, 14 men were not suitable for prostate biopsy, all in all 172 men underwent a prostate biopsy. All prostate biopsies were performed between May 1 and December 1, 2012, by two urologists (T.N., M.A.) following the current clinical guidelines, including prophylactic antibiotics and local anesthesia [6,7]. Ten to 12 ultrasound-guided core biopsies were taken from the prostatic peripheral zone. Immediately before the biopsy session, a serum PSA test was drawn, and throughout final analysis the level of this test was used. The men signed an informed consent prior to the investigation.

2.5. Biopsy specimen evaluation

Each needle biopsy core was formalin fixed in a separate container. At the pathology laboratory, the biopsy specimens were measured with a ruler. The specimens were dehydrated, paraffin-embedded in separate blocks, cut at 4 μm, and stained with hematoxylin and eosin. The biopsies were reviewed by a single observer who specialized in urologic pathology (L.E.), who was blinded to the PSA and genetic risk score of the individual patient. The biopsies were diagnosed as PCa, high-grade prostatic intraepithelial neoplasia (HGPIN), glandular atypia/suspicious for cancer, intraductal cancer, or benign. When cancer was diagnosed, the cancer length was measured in millimeters in each biopsy core, and the total cancer length was reported together with the number of biopsy cores positive for cancer. Cancer was Gleason graded according to the International Society of Urological Pathology 2005 modification [13].

2.6. Statistical analysis

Associations between PCa diagnosis and evaluated risk factors were explored in univariate and multivariate logistic regression analysis. The