The Heritability of Prostate Cancer in the Nordic Twin Study of Cancer

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Abstract

**Background:** Prostate cancer is thought to be the most heritable cancer, although little is known about how this genetic contribution varies across age.

**Methods:** To address this question, we undertook the world’s largest prospective study in the Nordic Twin Study of Cancer cohort, including 18,680 monozygotic (MZ) and 30,054 dizygotic (DZ) same-sex male twin pairs. We incorporated time-to-event analyses to estimate the risk concordance and heritability while accounting for censoring and competing risks of death, essential sources of biases that have not been accounted for in previous twin studies modeling cancer risk and liability.

**Results:** The cumulative risk of prostate cancer was similar to that of the background population. The cumulative risk for twins whose co-twin was diagnosed with prostate cancer was greater for MZ than for DZ twins across all ages. Among concordantly affected pairs, the time between diagnoses was significantly shorter for MZ than DZ pairs (median, 3.8 versus 6.5 years, respectively). Genetic differences contributed substantially to variation in both the risk and the liability (heritability = 58% [95% confidence interval, 52%–63%]) of developing prostate cancer. The relative contribution of genetic factors was constant across age through late life with substantial genetic heterogeneity even when diagnosis and screening procedures vary.

**Conclusions:** Results from the population-based twin cohort indicate a greater genetic contribution to the risk of developing prostate cancer when addressing sources of bias. The role of genetic factors is consistently high across age.

**Impact:** Findings affect the search for genetic and epigenetic markers and frame prevention efforts.

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Introduction

The etiology of prostate cancer remains enigmatic and poorly understood. Positive family history is consistently associated with a 2- to 4-fold increased risk of disease (1–3). Family history reflects both genetic and environmental factors shared by family members; however, several lines of research indicate that the familial effect for prostate cancer is largely genetic. Family and twin studies have proved essential for elucidating the relative importance of genetic and environmental factors in explaining differences in the liability to develop prostate cancer (1, 4). A landmark article in 2000 by Lichtenstein and colleagues reported that a substantial amount of the variation (42%) underlying prostate cancer liability in a Nordic twin cohort could be explained by genetic factors; this estimate of heritability (see Materials and Methods for definition) was the highest for any common cancer (4). The twin-based findings collaborate results generated over the past 5 years from genome-wide association studies, which have confirmed 70 susceptibility loci, explaining an estimated 30% of familial risk (5) with each locus accounting for only a minor part of the familial risk. The variation of genetic influence across age has not been studied before using large twin cohorts.

Estimates of the degree to which genetic differences account for familial risk and for the variation in liability to...
prostate cancer are typically based on studies using traditional twin statistical methodologies to study the influence of genetic factors on prostate cancer susceptibility (4, 6). These studies have largely ignored the often considerable censoring that can occur at both the beginning and end of follow-up, as well as competing causes of death, an issue of considerable importance in prostate cancer, given the relatively late in life incidence of the disease. Ignoring censoring can severely bias the incidence and risk concordance estimates, and can affect estimates of heritability. To address these issues, we incorporate novel statistical modeling strategies using data from the Nordic Twin Study of Cancer (NorTwinCan), the largest twin study of cancer in the world. NorTwinCan expands the study base used by Lichtstein and colleagues (4), with the addition of the Norwegian twin cohort, updated information from Danish, Finnish, and Swedish twins, and an additional 10 years of follow-up for incidence. The variation of genetic influence across age has not been studied previously using large twin cohorts, and we also examine age differences in the genetic variation underlying the risk to develop prostate cancer.

The Nordic cohorts are particularly suited for studying cancer because population-based registers with sufficient follow-up are available. Further, the well-known difference in cumulative incidence between the Danish and the remaining Nordic cohorts amounting to a lifetime risk (cumulative incidence at age 100) that is twice as high in Sweden, Norway, and Finland allows us to study the impact of different diagnostic and PSA screening procedures on our estimates of risk and heritability.

We estimate the cumulative incidence of prostate cancer to provide detailed estimates of familial risk among monozygotic (MZ) and dizygotic (DZ) pairs, as well as heritability, which provides a frame of reference for prostate cancer genome-wide association studies, and etiological research on prostate cancer in general.

Materials and Methods

The population-based twin cohorts

NorTwinCan is the population-based cohorts from the Danish, Finnish, Norwegian, and Swedish twin registries. Each twin has a unique national registration number, allowing for linkage to the national cancer and mortality registries with essentially complete follow-up.

Characteristics of the four twin cohorts are summarized in Table 1. The Danish cohort had the earliest cancer registration, initiated in January 1943. During follow-up for cancer incidence through 2009, 821 participants were diagnosed with prostate cancer. Although cancer registration in Finland started in 1953, we analyzed incident cases from 1975 onwards, when zygosity was determined as part of a questionnaire and both members of each twin pair were alive as of January 1975 (7). Through 2009, 547 men were diagnosed with prostate cancer. In all cohorts, zygosity was determined by validated questionnaire methodology, which classifies more than 95% of pairs of twins correctly.

The study was approved by the ethical committees for each country.

Statistical analysis

We extended conventional twin methods to address issues of censoring at follow-up and competing risk of death, which is particularly relevant in later life. Results from these analyses would agree with those obtained from the conventional twin approach (11, 12) if no censoring or competing risk of death were present.

We considered the underlying risk and liability to develop prostate cancer taking into account three possible outcomes: (i) prostate cancer diagnosis, (ii) no diagnosis and survival through end of follow-up in 2009, and (iii) no diagnosis or death during follow-up. Heritability is defined as the proportion of variance for a measure that is attributable to genetic differences in the population under study. Here, we estimate the heritability of (i) the risk of prostate cancer and (ii) the liability to develop prostate cancer. Casewise concordance estimates an
individual risk of disease conditional on disease in a close relative; in a twin study, it is defined as the risk of cancer in a twin, conditional on his co-twin having the same cancer. The twin can be as genetically similar as a full sibling, i.e., DZ twin or identical at the sequence level, i.e., MZ twin. Differences in concordance rates by zygosity provide insights into the influence of genetic and shared environmental factors on disease risk. Assuming that the environmental influences on disease risk are equally shared by MZ and DZ twins, greater concordance among MZ pairs compared with DZ pairs signals the importance of genetic effects.

We defined dates of entry and follow-up separately for each cohort, and included participants lost to follow-up to emigration (<2%). We accounted for left-censoring from variable initiation of cancer registration, and right-censoring among those censored at the end of follow-up or at competing risk of death by measuring the three possible outcomes for each individual at each timepoint and modeling the transition from no cancer diagnosis to either diagnosis or death. Before conducting pairwise analyses, we examined the individual risk of prostate cancer diagnosis by age by estimating cumulative prostate cancer incidence using the nonparametric Aalen–Johansen estimator (13). The lifetime risk reported is the cumulative incidence at age 100 years.

We modeled potential competing deaths as described in Scheike and colleagues (14, 15). This allows studying prostate cancer risk in a twin, given the occurrence of disease in the co-twin. To supplement the classical approach of estimating heritability of disease liability (explained below), we also estimated heritability of the risk scale. We obtained the casewise concordances by age and the relative recurrence risks in MZ and DZ pairs providing the multilocus index (16, 17) and the heritability of risk (14; i.e., twice the difference in MZ and DZ covariance in risk to the total variance in risk). Equality of casewise concordance curves for MZ and DZ pairs by age was tested by Pepe and Mori test (15).

**Biometric modeling of prostate cancer risk in twin pairs**

Quantitative models were analyzed to estimate the magnitude of the genetic and environmental variances (11, 12) that explain differences, i.e., variances, in disease liability. The general approach analyzes disease covariance between members of MZ and DZ pairs to decompose the variation into a sum of components: additive genetic effects (A), dominant genetic effects (which model deviations of the heterozygote genotype from the mean of the homozygote genotype; D), common environmental effects (C), and individually unique environmental effects (E). The genetic parameters of the model are specified on the basis of biologic relation between the co-twins. Within-pair covariance of liability is expressed as $\kappa \cdot \text{var}(A) + \gamma \cdot \text{var}(D) + \text{var}(C)$, in which $\kappa = \gamma = 1$ for MZ pairs and $\kappa = 1/2$ and $\gamma = 1/4$ for DZ pairs (11, 12). Because of statistical issues of identifiability, A, D, and C cannot be estimated simultaneously (12). Therefore, a series of models are analyzed that allow for sequential testing of the significance of specific parameters. Measurement error is estimated in E as this is the component of variance that does not contribute to within-pair resemblance. Dominance effects are, typically, biologically implausible in the absence of additive effects. The primary models are thus the ACE and ADE models, as well as their submodels AE, CE, and E.

We tested for equal thresholds (i.e., normal quantiles of prevalence) between MZ and DZ twins, which is equivalent to assuming that the risk of disease does not differ by zygosity. The biometric modeling approach we applied is comparable with that of Lichtenstein and colleagues (4), but adjusted for censoring. To test for differences in heritability with age at diagnosis, we estimated the within-pair correlations for MZ and DZ pairs and the cumulative heritability of prostate cancer liability at each decade of age. The fit of these submodels was assessed for each decade using the Akaike information criterion.

To correct for possible bias due to censoring at follow-up, individuals were assigned weights obtained by calculating the inverse probability of being censored at time of follow-up. Because censoring is dependent within pairs, the same weight was applied to twins within a pair (18). The probabilities of being censored were estimated using the Kaplan–Meier method and the Aalen additive model. We then analyzed the weighted sample of complete observations to obtain within-pair dependence estimates corrected for bias and heritability in liability to prostate cancer.

We examined differences in age at prostate cancer diagnosis within pairs, and estimated mean and median difference in age at diagnosis for pairs in which both members had the event. All analyses were conducted using the statistical program R with the package mets (15).

**Results**

The population-based NorTwinCan cohort comprised 143,467 male twins of which 4,109 men were diagnosed with prostate cancer through 2009 (Table 1). Restricting to same-sexed male pairs, there were 3,181 cases of prostate cancer diagnosed among 37,528 MZ and 60,411 DZ twins. Information about the 45,528 male twins from opposite-sexed DZ pairs was used for estimating incidence, but did not alter the incidence results. Within-pair concordances for vital status at end of follow-up are presented in Table 2. Information on the relatively large number of twins alive without cancer diagnosis at follow-up (censorings) shown in Table 2 has been accounted for in our results provided below.

The cumulative incidence of prostate cancer over time in each country is shown in Fig. 1. The lifetime risk of prostate cancer was 6.3% among men in the Danish cohort, compared with 12.0%, 12.8%, and 12.8% among men in the Finnish, Norwegian, and Swedish cohorts, respectively. This difference was stable across birth cohort (results not shown). Importantly, if we had ignored...
Censoring at follow-up, the cumulative lifetime incidence of prostate cancer would have been estimated to be 3% in the Nordic cohorts. The estimates in Fig. 1 are consistent with incidence rates of prostate cancer in the general male population within each country (19, 20). Cumulative incidence differed only slightly by zygosity with roughly 1% higher incidence in MZ than in DZ twins overall.

Table 3 presents the cohort-specific results for lifetime risk of disease, casewise concordances for disease risk by zygosity, and the genetic and shared environmental variance components underlying variation in disease liability. These results reveal a considerably increased risk of prostate cancer in a co-twin, and this familial effect is significantly greater among the MZ than the DZ pairs. Estimates of shared environmental effects and heritability varied across the four countries when applying the ACE model. Some indication of shared environmental influence on liability was suggested by findings based on the Norwegian and Finnish cohorts, but there was no evidence in the larger Swedish and Danish cohorts. The country-specific heritabilities in Table 3 did not vary significantly ($P$ value = 0.55). Overall, when adjusting for country-specific effects and censoring, the models AE, ACE, and ADE all yielded similar estimates of heritability between 55% and 58% with estimated but nonsignificant 5% shared environmental effects (C) and no indication of dominant genetic effects (D). The model with only environmental effects (the CE model) fitted very poorly to the data, whereas the AE model gave the most parsimonious fit and yielded a heritability estimate of 58% (95% Confidence Interval, CI, 0.52–0.63). These results were robust to stratification by age group and by birth cohort, both before and after the initiation of PSA screening.

Figure 2A and B shows the casewise concordance of prostate cancer and the corresponding heritability in risk of cancer by age for the Danish and the Nordic cohorts. At every age, prostate cancer risk for DZ twins whose co-twin had prostate cancer was higher compared with the overall cumulative incidence, indicating familial risk of prostate cancer. Moreover, the prostate cancer risk for an MZ twin, given that the co-twin was already diagnosed was 3-fold higher than the corresponding risk for DZ pairs; this zygosity-difference was remarkably stable across increasing age of diagnosis and similar by country (results not shown). For the Danish cohort, the incidence is half that of the other Nordic cohorts and stronger genetic component is indicated but, as illustrated by the curves of heritability of prostate cancer risk (Fig. 2), the magnitude of the genetic variation does not differ across age in any of the cohorts. The relative recurrence risk, $\lambda$, was higher in MZ than in DZ, and more pronounced in the Danish cohort as shown in Table 4; however, the DZ relative recurrence risks were very similar in the two cohorts. The ratio of $\lambda$ for MZ to DZ pairs, i.e., the multilocus index (15, 16), indicated substantial genetic heterogeneity toward prostate cancer with estimates nonsignificantly higher than two suggesting multiplicative (interaction) genetic effects of, e.g., multiple loci or epigenetic control. The multilocus index was remarkably stable across increasing age of diagnosis in both cohorts.

Among concordant twin pairs, there was a significantly shorter time between the diagnosis of prostate cancer in the first and second twin among the MZ than the DZ pairs. The mean difference was 4.6 years (SE, 0.43) for MZ concordant pairs and 7.8 years (SE, 0.45) for DZ concordant pairs ($P$ value < 0.0001). The corresponding median difference in time of diagnosis was 3.8 years for MZ pairs and 6.5 years for DZ pairs.

Figure 3 shows the cumulative heritability for prostate cancer liability by age at diagnosis derived from the genetic and environmental variance component.
modeling. We first fitted the saturated model providing empirical estimates of within-pair correlations in MZ and DZ pairs; the MZ correlations were significantly greater than the DZ correlations at each year of age. Correlations were highest for early-onset cancers for both MZ and DZ pairs. The model that most parsimoniously described the data included additive genetic, shared, and unique environmental components, i.e., the ACE model, after we corrected for bias due to censoring. That was the case at each timepoint, although the ACE, ADE, and AE models gave nearly identical estimates from age 80 years onwards. Hence, it seems that the type and magnitude of the genetic contribution do not differ with increasing age. Variation attributable to common environment may play a greater role for cancer onset up to age 80 as the ACE model provides the best fit to data; this is consistent with the elevated pattern of correlations for both MZ and DZ pairs for early cancer onset (Fig. 3). The lifetime shared environmental effect was estimated at 5% of the variance in liability.

<table>
<thead>
<tr>
<th>Cohort</th>
<th>Lifetime risk</th>
<th>Casewise concordance</th>
<th>Shared environment (c²)</th>
<th>Heritability (h²)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>MZ</td>
<td>DZ</td>
<td></td>
</tr>
<tr>
<td>Denmark</td>
<td>6.3% (5.8%–6.8%)</td>
<td>0.33 (0.25–0.42)</td>
<td>0.16 (0.14–0.19)</td>
<td>0 (—)</td>
</tr>
<tr>
<td>Nordic⁶</td>
<td>12.7% (12.2%–13.2)</td>
<td>0.40 (0.35–0.45)</td>
<td>0.25 (0.22–0.29)</td>
<td>0.05 (—)</td>
</tr>
</tbody>
</table>

NOTE: Bias correction due to censorings by the inverse probability weighting technique.

⁵Lifetime risk is the cumulative incidence at age 100 years. Casewise concordance and 95% CIs.

⁶Heritability in liability to prostate cancer from additive, shared, and unique environmental components (ACE model). No difference between country-specific heritabilities (P value = 0.55).

Overall ACE model for Finland, Norway, and Sweden combined with adjustment for censoring. Overall, the AE model is most parsimonious with heritability of 0.58 (0.52–0.63). Confidence limits not achievable for c². Estimates and 95% CIs.
Discussion

Results from this large, prospective cohort of Nordic twins provide evidence of genetic differences between people and explain a substantial portion of the variation (58%) in liability to develop prostate cancer. Moreover, among DZ twins who are as genetically similar as siblings, the lifetime probability of developing prostate cancer if the co-twin had cancer is around one of five, which is almost twice the lifetime risk in the general population. Our study also provides new insights about the importance of genetic effects across age of diagnosis. Specifically, the relative contribution of genetic factors was similar with increasing age into later life. The stability of the genetic contribution over age and the consistently greater genetic contribution over age and the remarkably stable pattern of substantial heritability across age into late life.

There are several important ways in which the current study extends the work by Lichtenstein and colleagues (4)

Our study builds upon much prior work describing familial aggregation of prostate cancer (22, 23). It has a number of strengths, including the long-term and complete follow-up of the cohorts, the population-based design that limits potential for selection bias, as well as the large number of cancer events that provides stable estimates of heritability and casewise concordance. Another strength is that the integration of novel statistical approaches in the context of the twin design increases the validity of the results by accounting for time to cancer diagnosis. The remarkably stable pattern of substantial genetic heterogeneity with age is consistent with synergistic genetic effects.

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One possible limitation is the lack of information on PSA screening. The incidence of prostate cancer is largely dependent on diagnostic intensity (24, 25). This is
evidenced by lower incidence of prostate cancer in Denmark relative to the other Nordic countries. PSA screening is less common in Denmark than in Finland, Norway, and Sweden (25). Prostate cancer mortality, however, is roughly equivalent for all Nordic countries (19, 20, 25). Differences in cumulative incidence by cohort affect within-pair dependence, yet heritability and other pairwise characteristics were fairly similar, also before and after initiation of PSA screening, which is also a strength to our findings. Further, there is only a minor and nonsignificant difference in the heritability of liability in Denmark, 59% versus 52% in the other countries combined.

Our analyses assume that the probability of screening among co-twins is independent of zygosity. However, if an MZ co-twin is more likely to be screened than a DZ co-twin of a diagnosed twin, the genetic component might be inflated. In general, as twins age, their behavioral traits become less associated, rendering the effect of differential screening minor. It is noteworthy that MZ twins had only a 1% higher incidence than DZ twins in all countries, and therefore differential screening seems unlikely. The lack of difference in incidence between MZ and DZ twins is consistent with the assumption that the causes (genetic and environmental) of prostate cancer do not differ by zygosity.

In comparison with breast cancer, it is reported that casewise concordance in DZ twins may increase with age (26). We cannot detect such a pattern for prostate cancer in our data. When we take prevalence by age into account, the relative recurrence risk decreases with age in MZ and DZ pairs; however, the corresponding multilocus index is stable across age.

Twin studies can provide context for genome-wide association studies that have identified multiple risk loci for prostate cancer incidence (27–30). Precise estimates of heritability allow for the calculation of the extent to which cancer variability is explained by established risk loci. Indeed, the concept of missing heritability has been proposed to describe the discrepancy between the variance in cancer associated with identified genetic loci and total heritability (31, 32). On the basis of the results of Lichtenstein and colleagues, researchers have estimated that 30% of the variance in prostate cancer risk is accounted for by the 70 established risk loci. Given our even higher estimate of heritability and high genetic influence on the directly comparable risk scale, the estimated missing heritability in prostate cancer is likely even larger; rare variants and gene–gene interactions are possible contributors to that missing heritability as well as epigenetic effects (33).

Heritability is a sample statistic and does not provide an estimate directly translatable to public health prevention (34), but it does provide insight into the reasons why individuals differ in their susceptibility to develop cancer. Further, the results help to frame the findings from genome-wide association studies and missing heritability. Moreover, precise risk estimates of prostate cancer among MZ pairs, who share 100% of their genomes, provide an upper limit of the potential for genotyping and whole-genome sequencing to risk classify individuals (32, 34, 35). Finally, proband concordance among DZ pairs can delineate the probability of cancer risk in families affected by prostate cancer.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

Authors’ Contributions
Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): J.B. Hjelmborg, A. Skytte, E. Pukkala, H.-O. Adami, N.V. Holm, K. Czene, J.R. Harris, J. Kaprio
Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): J.B. Hjelmborg, A. Skytte, N.V. Holm, E. Nuttall, J. Kaprio
Study supervision: J.B. Hjelmborg, H.-O. Adami, L.A. Mucci, J. Kaprio

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