

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

Jacob B. Hjelmborg^{1,2}, Thomas Scheike³, Klaus Holst³, Axel Skytthe^{1,2}, Kathryn L. Penney^{5,6}, Rebecca E. Graff⁵, Eero Pukkala^{7,10}, Kaare Christensen^{1,2}, Hans-Olov Adami^{5,11}, Niels V. Holm^{2,4}, Elizabeth Nuttall⁵, Steinbjorn Hansen⁴, Mikael Hartman^{11,12,13}, Kamila Czene¹¹, Jennifer R. Harris¹³, Jaakko Kaprio^{8,9}, and Lorelei A. Mucci^{5,6,14}

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. *Cancer Epidemiol Biomarkers Prev*; 23(11); 2303–10. ©2014 AACR.

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

¹Department of Epidemiology, Biostatistics, and Biodemography, Institute of Public Health, University of Southern Denmark. ²The Danish Twin Registry, University of Southern Denmark. ³Department of Biostatistics, University of Copenhagen. ⁴Department of Oncology, Odense University Hospital, Denmark. ⁵Department of Epidemiology, Harvard School of Public Health. ⁶Channing Division of Network Medicine, Brigham and Women's Hospital, Boston, Massachusetts. ⁷Finnish Cancer Registry, Institute for Statistical and Epidemiological Cancer Research. ⁸Department of Public Health and Institute for Molecular Medicine, University of Helsinki. ⁹Department of Mental Health and Substance Abuse Services, National Institute for Health and Welfare, Helsinki. ¹⁰School of Health Sciences, University of Tampere, Tampere, Finland. ¹¹Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden; ¹²Department of Surgery, National University Hospital and NUHS, Singapore. ¹³Division of Epidemiology, The Norwegian Institute of Public Health, Oslo, Norway. ¹⁴Centre for Public Health Sciences, University of Iceland, Reykjavik, Iceland.

Corresponding Author: Jacob B. Hjelmborg, The Danish Twin Registry, University of Southern Denmark, J. B. Winsløvsvej 9 B, 5000 Odense, Denmark. Phone: 45-6550-3075; Fax: 45-6550-3075; E-mail: jhjelmborg@health.sdu.dk

doi: 10.1158/1055-9965.EPI-13-0568

©2014 American Association for Cancer Research.

Table 1. Description of the NorTwinCan cohorts

	Denmark	Finland	Norway	Sweden
Birth cohorts	1870–1999	1887–1957	1896–1979	1886–1999
Number of male twins	53,076	12,154	12,318	65,919
Number of MZ/DZ pairs	6,092/11,132	1,792/4,222	2,392/3,026	8,438/11,731
End of follow-up	December 31, 2009	December 31, 2009	December 31, 2008	December 31, 2009
Number of MZ/DZ pairs uncensored at follow-up	1,300/2,456	388/819	231/298	1,632/2,843
Cancer registration since	1943	1953	1953	1958
Number of prostate cancer cases	821	547	356	2,385

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 Lichtenstein 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 an international, transdisciplinary collaboration of researchers studying the genetic and environmental underpinnings of cancer. A key component of

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. For members of the Norwegian cohort to qualify for inclusion, both members had to be alive as of January 1964 (8). Through 2009, 356 men were diagnosed with prostate cancer. For the Swedish cohort, both twin pairs had to be alive as of January 1961 (9, 10). Through 2009, 2,385 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 $\kappa \text{ var}(A) + \gamma \text{ var}(D) + \text{Var}(C)$, 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

Table 2. Number of pairs by vital status with respect to cancer diagnosis at end of follow-up among MZ (lower left triangle) and DZ twin pairs (upper right triangle) in the NorTwinCan database

Pairs (MZ/DZ)	Cancer	Dead without cancer	Alive without cancer
Cancer	194/146	982	699
Dead without cancer	476	3,103/5,606	4,110
Alive without cancer	320	1,756	12,865/18,568

NOTE: Bold indicates number of pairs (DZ/MZ) that are concordant for status.

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

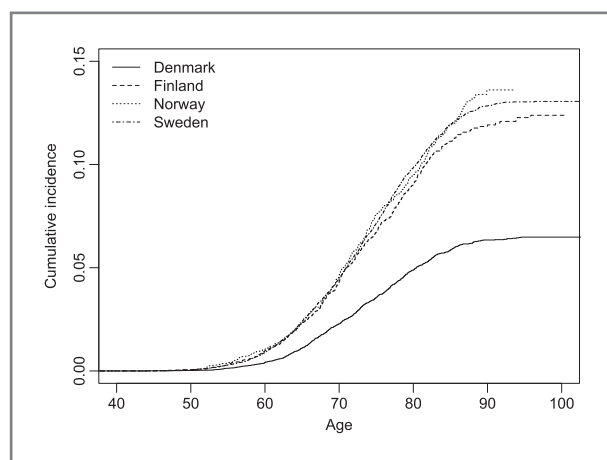


Figure 1. The cumulative incidence of prostate cancer in Nordic twin cohorts adjusted for censoring, delayed prostate due to initiation time of cancer registration in cohorts, and competing risk of death.

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, λ , 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 $\lambda-1$ 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

Table 3. Cumulative risk, casewise concordance risk, and heritability of liability to prostate cancer diagnosis, overall and by cohort in the NorTwinCan database

Cohort	Lifetime risk ^a	Casewise concordance ^a		Estimates from twin modeling ^b	
		MZ	DZ	Shared environment (c^2)	Heritability (h^2)
Denmark	6.3% (5.8%–6.8%)	0.33 (0.25–0.42)	0.16 (0.14–0.19)	0 (–)	0.59 (0.47–0.70)
Nordic ^c	12.7% (12.2%–13.2)	0.40 (0.35–0.45)	0.25 (0.22–0.29)	0.05 (–)	0.52 (0.31–0.72)

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

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

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

^cOverall 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 c^2 . Estimates and 95% CIs.

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.

Figure 2. A, casewise concordance of prostate cancer risk by age at diagnosis. The risk of prostate cancer in a co-twin by age for MZ pairs (black) and DZ pairs (red). Cumulative incidence twins (thin black). Risks are adjusted for cohort effects, censoring, and competing risk of death and are significantly different for MZ and DZ pairs over time (P value < 0.0001). B, heritability of prostate cancer risk by age at diagnosis. Heritability in risk is twice the difference between MZ and DZ concordance to the total variance in risk (adjusted for cohort effects, censoring, and competing risk of death).

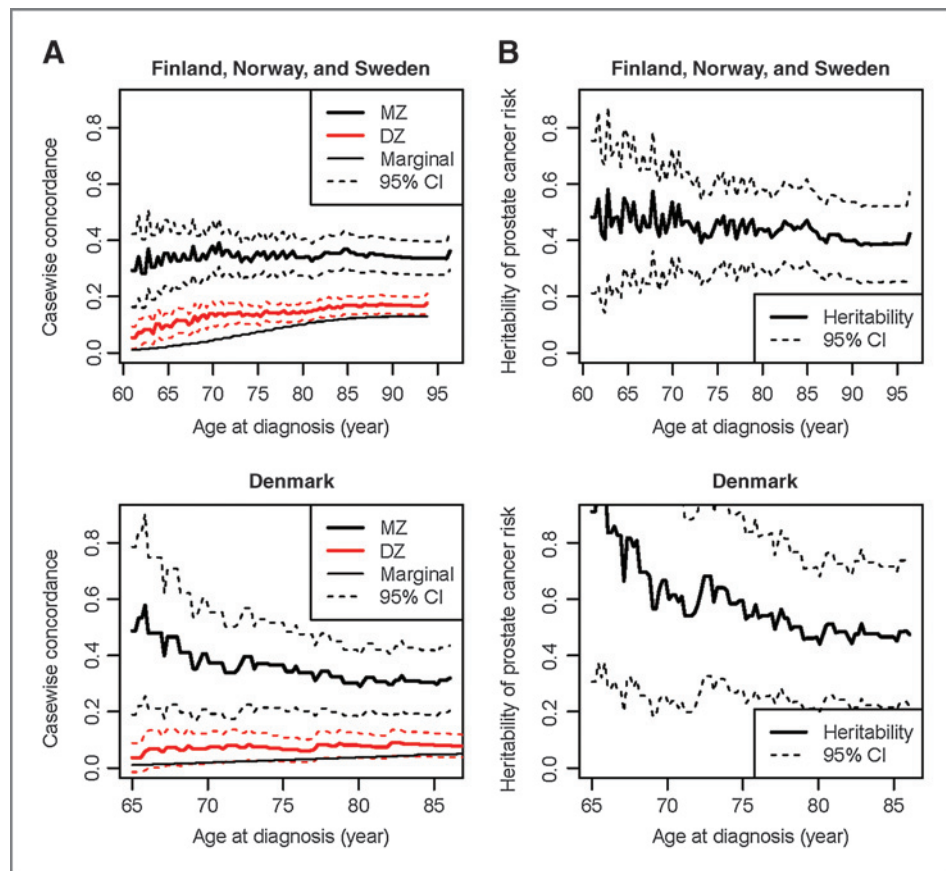


Table 4. Relative recurrence risk, λ , and multilocus index by decades of age in the NorTwinCan database

Age	Danish cohort			Nordic ^a cohort		
	λ (MZ)	λ (DZ)	Multilocus ^b	λ (MZ)	λ (DZ)	Multilocus
All	5.69 (1.01)	1.70 (0.42)	6.66 (4.24)	2.94 (0.24)	1.68 (0.15)	2.86 (0.73)
–70	24.04 (6.05)	4.09 (1.73)	7.44 (4.60)	10.41 (1.29)	4.25 (0.67)	2.90 (0.72)
70–80	9.03 (1.76)	2.22 (0.64)	6.59 (3.77)	4.38 (0.39)	2.13 (0.23)	2.99 (0.69)
80–90	5.86 (1.05)	1.65 (0.43)	7.43 (5.18)	3.01 (0.25)	1.69 (0.15)	2.91 (0.74)
90+	5.23 (0.96)	1.68 (0.43)	6.23 (4.16)	2.75 (0.23)	1.62 (0.15)	2.80 (0.78)

NOTE: Bias correction due to censoring and competing risk of death.

^aOverall for Finland, Norway, and Sweden combined. Estimates and SE in parentheses.

^bThe multilocus index measuring genetic heterogeneity is defined as the ratio of $\lambda(\text{MZ}) - 1$ to $\lambda(\text{DZ}) - 1$.

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 MZ than DZ correlation across age is congruent with the observed within-pair difference in occurrence between MZ and DZ case pairs of roughly 3 years on average.

There are several important ways in which the current study extends the work by Lichtenstein and colleagues (4) that had been conducted on the largest previous twin

sample. We accounted for differential follow-up time, problems of censoring, and competing risk of death (4). Furthermore, our study includes more than twice the number of pairs, an increased follow-up of 10 years, and an increased number of prostate cancer events. Furthermore, we estimate the importance of genetic variance on both the risk and the liability to develop prostate cancer. Our estimate of the genetic contribution is considerably higher than that described by Lichtenstein and colleagues, which reported heritability to be 42% (95% CI, 29%–50%; ref. 4). Without accounting for censoring, our overall heritability estimate was 57% with shared environment accounting for 9% of the variance and the lifetime risk was underestimated (at 3%). Among U.S. World War II veteran twins, 57% of variation in liability for prostate cancer was attributed to genetic influences in an AE model (21), although with wide CIs. Unlike our study (in which an ACE structure was indicated), the veteran cohort did not indicate a shared environmental influence for its younger age group, 65 to 75 years old, although the sample size was much smaller and did not account for censoring. Further, our study provides novel insight into the variation across age for measures of risk and liability of prostate cancer diagnosis. The remarkably stable pattern of substantial genetic heterogeneity with age is consistent with synergistic genetic effects.

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 onset and risk of death. This approach resulted in higher than previously reported genetic influence on prostate cancer across age into late life.

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

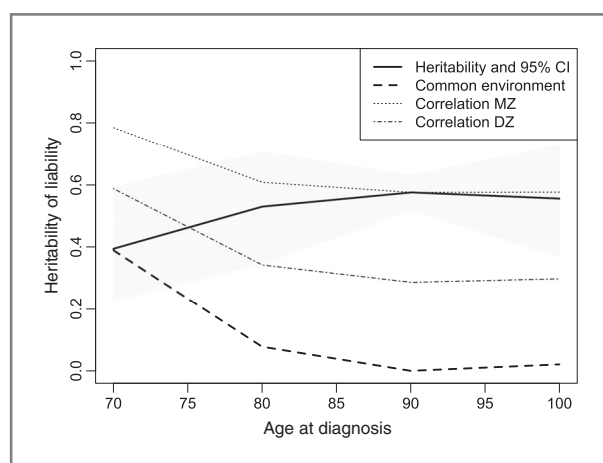


Figure 3. The cumulative heritability in liability to prostate cancer by age at diagnosis (time) modeling additive genetic, common, and unique environmental components of liability to disease. Correlation in MZ and DZ pairs. Bias correction due to censorings by the inverse probability weighting technique. Transparent areas, 95% confidence bands.

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 can not 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

Conception and design: J.B. Hjelmborg, E. Pukkala, K. Christensen, H.-O. Adami, N.V. Holm, K. Czene, J.R. Harris, J. Kaprio, L.A. Mucci
Development of methodology: J.B. Hjelmborg, T. Scheike, K. Holst, E. Pukkala, K. Christensen, L.A. Mucci, J. Kaprio
Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): J.B. Hjelmborg, A. Skytthe, E. Pukkala, H.-O. Adami, N.V. Holm, K. Czene, J.R. Harris, J. Kaprio
Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): J.B. Hjelmborg, T. Scheike, K. Holst, A. Skytthe, E. Pukkala, H.-O. Adami, N.V. Holm, S. Hansen, J. Kaprio, L.A. Mucci
Writing, review, and/or revision of the manuscript: J.B. Hjelmborg, T. Scheike, K. Holst, A. Skytthe, K.L. Penney, R.E. Graff, E. Pukkala, K. Christensen, H.-O. Adami, N.V. Holm, E. Nuttall, S. Hansen, M. Hartman, K. Czene, J.R. Harris, J. Kaprio, L.A. Mucci
Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): J.B. Hjelmborg, A. Skytthe, N.V. Holm, E. Nuttall, J. Kaprio
Study supervision: J.B. Hjelmborg, H.-O. Adami, L.A. Mucci, J. Kaprio

Acknowledgments

The authors thank The Danish Twin Registry for hosting and managing the joint Nordic twin data. The NorTwinCan group would also like to thank the Ellison Foundation for the critical funding that made this research possible.

Grant Support

This work was supported by funding from the Ellison Foundation (L.A. Mucci and H.-O. Adami, principal investigators) and the Nordic Union of Cancer (J. Kaprio, principal investigator). R.E. Graff is supported by a training grant from the NIH (R25 CA098566). L.A. Mucci and K.L. Penney are supported by the Prostate Cancer Foundation. The Finnish Twin Cohort was supported by the Academy of Finland Center of Excellence in Complex Disease Genetics (grant numbers 213506 and 129680). The Danish Twin Cohort was supported by the Odense University Hospital AgeCare program (Academy of Geriatric Cancer Research).

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

Received May 31, 2013; revised March 25, 2014; accepted April 26, 2014; published OnlineFirst May 8, 2014.

References

- Adami HO, Hunter DJ, Trichopoulos D. Textbook for cancer epidemiology. 2nd ed. New York: Oxford University Press; 2008.
- Wilson KM, Giovannucci EL, Mucci LA. Lifestyle and dietary factors in the prevention of lethal prostate cancer. *Asian J Androl* 2012;14:365–74.
- Hemminki K. Familial risk and familial survival in prostate cancer. *World J Urol* 2012;30:143–8.
- Lichtenstein P, Holm NV, Verkasalo PK, Iliadou A, Kaprio J, Koskenvuo M, et al. Environmental and heritable factors in the causation of

- cancer—analyses of cohorts of twins from Sweden, Denmark, and Finland. *N Engl J Med* 2000;343:78–85.
5. Eeles RA, Olama AA, Benlloch S, Saunders EJ, Leongamornlert DA, Tymrakiewicz M, et al. Identification of 23 new prostate cancer susceptibility loci using the iCOGS custom genotyping array. *Nat Genet* 2013;45:385–91, 91e1–2.
 6. Baker SG, Lichtenstein P, Kaprio J, Holm N. Genetic susceptibility to prostate, breast, and colorectal cancer among Nordic twins. *Biometrics* 2005;61:55–63.
 7. Kaprio J, Koskenvuo M. Genetic and environmental factors in complex diseases: the older Finnish Twin Cohort. *Twin Res* 2002;5:358–65.
 8. Nilsen TS, Brandt I, Magnus P, Harris JR. The Norwegian Twin Registry. *Twin Res Hum Genet* 2012;15:775–80.
 9. Lichtenstein P, De Faire U, Floderus B, Svartengren M, Svedberg P, Pedersen NL. The Swedish Twin Registry: a unique resource for clinical, epidemiological and genetic studies. *J Intern Med* 2002;252:184–205.
 10. Pedersen NL, Lichtenstein P, Svedberg P. The Swedish Twin Registry in the third millennium. *Twin Res* 2002;5:427–32.
 11. Neale MC, Cardon LR. *Methodology for genetic studies of twins and families*. Dordrecht: Kluwer Academic Publishers; 1992.
 12. Sham P. *Statistics in human genetics*. Chichester: John Wiley & Sons Ltd.; 1998.
 13. Allignol A, Schumacher M, Beyersmann J. Empirical Transition Matrix of Multi-State Models: the etm Package. *J Stat Softw* 2011;38.
 14. Scheike TH, Holst KK, Hjelmberg JB. Estimating heritability for cause specific mortality based on twin studies. *Lifetime Data Anal* 2014;20:201–33.
 15. Scheike TH, Holst KK, Hjelmberg JB. Estimating twin concordance for bivariate competing risks twin data. *Stat Med* 2014;33:1193–204.
 16. Risch N. The genetic epidemiology of cancer: interpreting family and twin studies and their implications for molecular genetic approaches. *Cancer Epidemiol Biomarkers Prev* 2001;10:733–41.
 17. Risch N. Linkage strategies for genetically complex traits. I. Multilocus models. *Am J Hum Genet* 1990;46:222–8.
 18. Van der Laan MJ, Robins JM. *Unified methods for censored longitudinal data and causality*. Springer Series in Statistics. New York: Springer; 2003.
 19. Bray F, Lortet-Tieulent J, Ferlay J, Forman D, Auvinen A. Prostate cancer incidence and mortality trends in 37 European countries: an overview. *Eur J Cancer* 2010;46:3040–52.
 20. Meyer MS, Mucci LA, Andersson SO, Andren O, Johansson JE, Tretli S, et al. Homogeneous prostate cancer mortality in the Nordic countries over four decades. *Eur Urol* 2010;58:427–32.
 21. Page WF, Braun MM, Partin AW, Caporaso N, Walsh P. Heredity and prostate cancer: a study of World War II veteran twins. *Prostate* 1997;33:240–5.
 22. Cote ML, Liu M, Bonassi S, Neri M, Schwartz AG, Christiani DC, et al. Increased risk of lung cancer in individuals with a family history of the disease: a pooled analysis from the International Lung Cancer Consortium. *Eur J Cancer* 2012;48:1957–68.
 23. Scheurer ME, Etzel CJ, Liu M, Barnholtz-Sloan J, Wiklund F, Tavelin B, et al. Familial aggregation of glioma: a pooled analysis. *Am J Epidemiol* 2010;172:1099–107.
 24. Adami HO. The prostate cancer pseudo-epidemic. *Acta Oncol* 2010;49:298–304.
 25. Kvale R, Auvinen A, Adami HO, Klint A, Hernes E, Moller B, et al. Interpreting trends in prostate cancer incidence and mortality in the five Nordic countries. *J Natl Cancer Inst* 2007;99:1881–7.
 26. Peto J, Mack TM. High constant incidence in twins and other relatives of women with breast cancer. *Nat Genet* 2000;26:411–4.
 27. Ghousaini M, Fletcher O, Michailidou K, Turnbull C, Schmidt MK, Dicks E, et al. Genome-wide association analysis identifies three new breast cancer susceptibility loci. *Nat Genet* 2012;44:312–8.
 28. Haiman CA, Chen GK, Vachon CM, Canzian F, Dunning A, Millikan RC, et al. A common variant at the TERT-CLPTM1L locus is associated with estrogen receptor-negative breast cancer. *Nat Genet* 2011;43:1210–4.
 29. Kote-Jarai Z, Olama AA, Giles GG, Severi G, Schleutker J, Weischer M, et al. Seven prostate cancer susceptibility loci identified by a multi-stage genome-wide association study. *Nat Genet* 2011;43:785–91.
 30. Peters U, Hutter CM, Hsu L, Schumacher FR, Conti DV, Carlson CS, et al. Meta-analysis of new genome-wide association studies of colorectal cancer risk. *Hum Genet* 2012;131:217–34.
 31. Manolio TA, Collins FS, Cox NJ, Goldstein DB, Hindorf LA, Hunter DJ, et al. Finding the missing heritability of complex diseases. *Nature* 2009;461:747–53.
 32. Visscher PM, Brown MA, McCarthy MI, Yang J. Five years of GWAS discovery. *Am J Hum Genet* 2012;90:7–24.
 33. Borno ST, Fischer A, Kerick M, Falth M, Laible M, Brase JC, et al. Genome-wide DNA methylation events in TMPRSS2-ERG fusion-negative prostate cancers implicate an EZH2-dependent mechanism with miR-26a hypermethylation. *Cancer Discov* 2012;2:1024–35.
 34. Zuk O, Hechter E, Sunyaev SR, Lander ES. The mystery of missing heritability: Genetic interactions create phantom heritability. *Proc Natl Acad Sci U S A* 2012;109:1193–8.
 35. Roberts NJ, Vogelstein JT, Parmigiani G, Kinzler KW, Vogelstein B, Velculescu VE. The predictive capacity of personal genome sequencing. *Sci Transl Med* 2012;4:133ra58.