Survival and cardiovascular events in men treated with testosterone replacement therapy: an intention-to-treat observational cohort study

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Summary

Background Conflicting evidence exists for the association between testosterone replacement therapy and mortality and cardiovascular events. The US Food and Drug Administration recently cautioned that testosterone replacement therapy might increase risk of heart attack and stroke, based on evidence from studies with short treatment duration and follow-up. No previous study has assessed the effect of duration of testosterone treatment on these outcomes. We aimed to assess the association between long-term use of testosterone replacement therapy and mortality, cardiovascular events, and prostate cancer diagnoses, using a time-varying exposure analysis.

Methods We did a population-based matched cohort study of men aged 66 years or older newly treated with testosterone replacement therapy and controls matched for age, region of residence, comorbidity, diabetes status, and index year from 2007–12 in Ontario, Canada, using data from the Ontario Drug Benefit database, the Canadian Institute for Health Information (CIHI) Discharge Abstract Database, the CIHI National Ambulatory Care Reporting System, the Ontario Health Insurance Plan database, the Ontario Myocardial Infarction Database, the Ontario Diabetes Database, the Ontario Cancer Registry, and the Registered Persons database. We assessed the association between cumulative testosterone replacement therapy exposure and mortality, cardiovascular events, and prostate cancer using marginal models with a time-varying testosterone exposure.

Findings We included 10 311 men treated with testosterone replacement therapy and 28 029 controls between Jan 1, 2007, and June 30, 2012. Over a median follow-up of 5·3 years (IQR 3·6–7·5) in the testosterone replacement therapy group and 5·1 years (3·4–7·4) in the control group, patients treated with testosterone replacement therapy had lower mortality than did controls (hazard ratio [HR] 0·88, 95% CI 0·84–0·93). Patients in the lowest tertile of testosterone exposure had increased risk of mortality (HR 1·11, 95% CI 1·03–1·20) and cardiovascular events (HR 1·26, 95% CI 1·09–1·46) compared with controls. By contrast, those in the highest tertile of testosterone exposure had decreased risk of mortality (HR 0·67, 95% CI 0·62–0·73) and cardiovascular events (HR 0·84, 95% CI 0·72–0·98), with a significant trend across tertiles (p<0·0001). Risk of prostate cancer diagnosis was decreased for those with the highest tertile of exposure (HR 0·60, 95% CI 0·45–0·80) compared with controls, but not for those with the shortest exposure.

Interpretation Long-term exposure to testosterone replacement therapy was associated with reduced risks of mortality, cardiovascular events, and prostate cancer. However, testosterone replacement therapy increased the risk of mortality and cardiovascular events with short durations of therapy. In view of the limitations of observational data and the potential for selection bias, these results warrant confirmation in a randomised trial.

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Introduction

Testosterone deficiency syndrome is defined by low serum testosterone concentrations and signs and symptoms attributable to the physiological effects of low testosterone, such as reduced sexual desire, fatigue, depressed mood, and reduced strength.[1,2] Incidence increases with age, affecting 20% of men older than 60 years, 30% of men older than 70 years, and 50% of men older than 80 years.[1] Testosterone deficiency syndrome is also associated with an increased incidence of obesity, diabetes, dyslipidaemia, and hypertension.[1] Furthermore, patients with low testosterone have an increased risk of cardiovascular-related events.[3]

Several studies have been done to investigate the relation between testosterone replacement therapy and cardiovascular events. Of these, results from two large observational studies[4,5] and a meta-analysis of randomised controlled trials[6] showed evidence of harm. Subsequently, findings from several observational studies have shown either no association between testosterone replacement therapy and cardiovascular events[7] or a beneficial effect.[8–11] Because of this uncertainty, the US Food and Drug Administration (FDA) has warned that testosterone replacement therapy might increase risk of heart attack and stroke.[12] Thus, although investigators of a recent meta-analysis[13]

Research in context

Evidence before this study

Previous studies into the effect of testosterone replacement therapy on cardiovascular outcomes have been conflicting, with some showing significant benefit and others showing harm. Investigators of a recent meta-analysis concluded that there was no conclusive evidence of an association in either direction. However, based on a search of MEDLINE up to July 1, 2015 (and updated in March, 2016), of studies published in English, no previous study had assessed the effect of testosterone replacement therapy exposure duration on mortality or cardiovascular disease. Search terms included:"testosterone" and ("dose" or "exposure" or "length" or "time") and "mortality" and "cardiovascular" and "myocardial infarction" and "stroke" and "cerebrovascular accident" and "prostate cancer".

Added value of this study

Using a large cohort of patients treated with testosterone replacement therapy and matched controls, we assessed the effect of cumulative testosterone dose exposure on mortality, cardiovascular events, and prostate cancer diagnoses. With a follow-up of more than 5 years, short durations of testosterone replacement therapy treatment were associated with an increased risk of death and cardiovascular events, whereas longer durations of therapy were associated with decreased risk of these events.

Implications of all the available evidence

Long-term treatment with testosterone replacement therapy seems to be safe and might be associated with decreased risk of cardiovascular events. However, patients and physicians should be aware of the risk of events in the short term. In view of potential biases in observational study design, the findings of this study and others require validation in a randomised controlled trial.

and a position statement from the American Association of Clinical Endocrinologists and the American College of Endocrinology[14] both concluded that there was no conclusive evidence that testosterone replacement therapy either increased or decreased cardiovascular events.

Most of the studies done to investigate the association had short follow-up; however, evidence suggests that the effects of hormone treatments change over time. Improvements in bone mineral density, increased lean body mass, and reduced body fat in men increase with increasing durations of testosterone treatment.[15] In women, outcomes differ depending on duration of follow-up after hormone-replacement therapy.[16] In an observational study reported in 2015,[17] first-time use of testosterone replacement therapy, but not ongoing treatment, was associated with an increased risk of myocardial infarction. However, no previous study has assessed the effect of duration of testosterone replacement therapy exposure on mortality and cardiovascular events.

In this study, we sought to assess the effect of testosterone replacement therapy on overall mortality and cardiovascular events in men with long-term follow-up using a time-varying testosterone exposure. Prostate cancer was examined since testosterone replacement therapy has historically been considered a risk factor for developing prostate cancer.

Methods

Study design and data sources

We did a population-based, retrospective matched cohort study of men aged 66 years or older newly treated with testosterone replacement therapy between Jan 1, 2007, and June 30, 2012, in Ontario, Canada. In Ontario, medical care is reimbursed by a single, government-operated health insurance system (Ontario Health Insurance Plan). Prescription medications are provided to all citizens aged 65 years or older through the Ontario Drug Benefit. Since March, 2006, the Ontario Drug Benefit has covered all testosterone formulations for the indication of symptomatic testosterone deficiency. We excluded prescriptions before March, 2006, because of potential irregularities during early programme implementation. Patients were followed up from index date (the date of first prescription for patients exposed to testosterone replacement therapy, and corresponding date for controls) until the development of an outcome or March 21, 2013. Sunnybrook Health Sciences Centre Research ethics board approved this study.

We linked the following datasets: the Ontario Drug Benefit database, which provides information for all outpatient drug prescriptions;[18] the Canadian Institute for Health Information (CIHI) Discharge Abstract Database, which contains records for hospital admissions;[19] the CIHI National Ambulatory Care Reporting System, which contains records for ambulatory and emergency room visits; the Ontario Health Insurance Plan database, which tracks claims paid for physician billings, laboratories, and out-of-province providers;[20] the Ontario Myocardial Infarction Database, a validated registry of all patients diagnosed with acute myocardial infarction;[21] the Ontario Diabetes Database, which provides validated data for all people living in Ontario diagnosed with diabetes;[22] the Ontario Cancer Registry, which is estimated to be more than 95% complete;[23] and the Registered Persons database, which contains validated demographic information.[24] All these databases have been validated (appendix p 2).

See Online for appendix

	Testosterone replacement therapy (n=10 311)	Controls (n=28 029)
RUB (standardised difference 0·06*)	3·00 (3·00–4·00)	3·00 (3·00–4·00)
Diabetes diagnosis (standardised difference 0·01*)		
Yes	3589 (35%)	9607 (34%)
No	6722 (65%)	18 422 (66%)
Age group (standardised difference 0·07*)		
65–69 years	3724 (36%)	10 478 (37%)
70–74 years	3186 (31%)	8783 (31%)
75–79 years	1974 (19%)	5304 (19%)
80–84 years	974 (9%)	2510 (9%)
85–89 years	346 (3%)	812 (3%)
>90 years	107 (1%)	142 (1%)
Income quintile (standardised difference 0·10*)		
1	1730 (17%)	5042 (18%)
2	1807 (18%)	5575 (20%)
3	1957 (19%)	5610 (20%)
4	2184 (21%)	5662 (20%)
5	2611 (25%)	6060 (22%)
Missing	22 (<1%)	80 (<1%)
Index calendar year (standardised difference 0·01*)		
2007	4243 (41%)	11 382 (41%)
2008	1153 (11%)	3105 (11%)
2009	1300 (13%)	3557 (13%)
2010	1436 (14%)	3968 (14%)
2011	1366 (13%)	3780 (13%)
2012	813 (8%)	2237 (8%)
LHIN† (standardised difference 0·04*)		
Region 1	433 (4 %)	1175 (4%)
Region 2	556 (5%)	1439 (5%)
Region 3	102 (1%)	216 (1%)
Region 4	918 (9%)	2523 (9%)
Region 5	587 (6%)	1582 (6%)
Region 6	1125 (11%)	3128 (11%)
Region 7	1250 (12%)	3503 (12%)
Region 8	358 (3%)	922 (3%)
Region 9	942 (9%)	2531 (9%)
Region 10	684 (7%)	1839 (7%)
Region 11	743 (7 %)	2050 (7%)
Region 12	1452 (14%)	4065 (15%)
Region 13	416 (4%)	1069 (4%)
Region 14	745 (7%)	1987 (7%)

Data are n (%) or median (IQR). LHIN=Local Health Integration Network. RUB=Resource Utilization Band. †In keeping with the privacy regulations of the Institute for Clinical Evaluative Sciences, the identities of each region cannot be disclosed.

Table 1: **Baseline characteristics of men treated with testosterone-replacement therapy and age-matched and comorbidity-matched controls**

For the **local health integration networks** see http://www.lhins.on.ca

Study population

We included patients aged 66 years or older who received one or more new prescriptions for any testosterone formulation identified from computerised prescription records from the Ontario Drug Benefit database. We selected age 66 years as the cutoff to allow for a 1-year look-back to ensure that patients were not exposed to testosterone therapy before study entry, which would preclude accurate assessment of testosterone exposure. Based on product monographs, serum testosterone concentrations return to baseline within 2 weeks of cessation for all testosterone formulations included in this analysis (appendix p 3), thus residual effects of previous treatment are unlikely to be present after 1 year.

We matched each patient exposed to testosterone replacement therapy with up to three men from the general population who had no use of testosterone replacement therapy (so-called hard match). Controls were matched to patients for age at within 1 year of the index date, region of residence, comorbidity (Resource Utilization Band), diabetes status, and index year. We identified region of residence using local health integration networks, which are regions that plan and deliver health-care resources and thus affect a patient's access to care. The Resource Utilization Band is a well established measure of comorbidity designed to measure overall morbidity such that individuals with the same Resource Utilization Band score (ranging from 0 [non-user] to 5 [very high morbidity]) have the same intensity of use of health-care resources,[25] and is based on the Johns Hopkins Aggregate Disease Groups.[26] With both ambulatory and hospital data, all diagnoses for a particular patient are coded by cause, duration, severity, diagnostic certainty, and involvement of specialist care. When we could not identify a suitable control, we used fewer than the planned three controls per patient.

Procedures

Using the Ontario Drug Benefit database, we identified patients treated with any formulation of testosterone replacement therapy (appendix p 3). These data have been shown to accurately reflect testosterone prescriptions in Ontario.[27] We assessed the effect of testosterone therapy in two ways. First, we treated testosterone as a dichotomous exposure (any or never). Second, we assessed cumulative dose exposure, stratified into tertiles. We measured cumulative dose exposure using the total number of days of testosterone replacement therapy dispensed according to prescription records. We analysed by tertiles to minimise loss of study power for this covariate and to allow us to assess cumulative dose exposure effects in a clinical meaningful way. We censored exposure duration at the date of development of an outcome or end of follow-up when prescriptions continued beyond these dates (<1% of cases)

Outcomes

Our primary outcome was overall mortality. Secondary outcomes were a composite cardiovascular outcome (consisting of myocardial infarction, cerebrovascular accident, or venous thromboembolic event) and prostate cancer diagnosis (appendix p 4). Because we used a

survival analysis, we considered only the first event for each patient for each outcome.

Statistical analysis

We compared baseline characteristics of the testosterone replacement therapy group and controls using standardised differences.[28] Standardised differences capture clinically meaningful, rather than statistically significant, differences between groups. Groups are considered similar if the standardised difference is less than or equal to 10%.

We used the Kaplan-Meier method to assess each of our endpoints, stratifying by testosterone exposure status. Patients were considered at risk from the index date until the event or date of last follow-up. We used the stratified log-rank test to compare survival between groups. In view of the clustering inherent in matched data, we used generalised estimating equation survival models with a sandwich variance estimator (marginal models) to assess the effect of testosterone therapy on each outcome. We used the matching identifier as a clustering variable. For our primary outcome, we used a time-varying testosterone replacement therapy exposure to correct for a potential immortal-time bias.[29,30] We partitioned exposure and outcome time for each man exposed to testosterone replacement therapy into tertiles: each man contributed time and outcome status to the first tertile group until he reached 120 days of cumulative exposure, to the second tertile group from 120 to 510 days of exposure, and to the third tertile group thereafter.[31] We assessed the global null hypothesis using the Wald test. We verified the assumptions underlying the model and identified no evidence of violations.

In a post-hoc analysis, we investigated whether the effect of testosterone replacement therapy on mortality changes over time. We divided the follow-up period into an early period and a late period using threshold dates (we first used 180 days as the threshold between early and late; we then did the analysis with other thresholds: 270 days, 365 days, and 455 days). We then repeated our analyses in each of these periods using testosterone as both a dichotomous and time-varying exposure (appendix p 1). p values less than 0·05 were regarded as significant, based on a two-tailed comparison. We did all statistical analyses with SAS version 9.3.

Role of the funding source

The funders of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. CJDW and RKN had full access to all the data in the study and all authors had final responsibility for the decision to submit for publication.

Results

Between Jan 1, 2007, and June 30, 2012, we identified 10 749 men aged 66 years or older, who newly received testosterone therapy. We excluded 38 (3%) men with a history of cardiovascular events (myocardial infarction, cerebrovascular accident, or venous thromboembolism), 97 (1%) with prostate cancer, and 3 (<1%) with illogical follow-up dates (administrative errors in which patients have negative follow-up). No patients were excluded for missing data. 10 311 men were eligible for inclusion. Of 29 978 identified matched controls, we excluded 264 (1%) who were matched to a case with a previous diagnosis of prostate cancer, 1383 (5%) for a previous diagnosis of prostate cancer, 120 (<1%) because of a prior history of cardiovascular events, and 182 (1%) for illogical follow-up dates. No controls were excluded for missing data.

Based on our hard match of age, comorbidity, diabetes, region of residence, and index year, we matched 28 029 controls who were not exposed to testosterone replacement therapy. All exposed men were matched to at least one control (558 [5%] were matched to one control; 1569 [15%] were matched to two controls; and 8157 [79%] were matched to three controls). Median follow-up was 5·3 years (IQR 3·6–7·5; mean 5·3 years, SD 2·1) in the testosterone replacement therapy group and 5·1 years (3·4–7·4; mean 5·1 years, SD 2·2; p<0·0001) in the control group. The testosterone replacement therapy and control groups had similar baseline characteristics (table 1). 5782 (21%) of people in the control group died during follow-up, 1465 (5%) had a cardiovascular event, and 799 (3%) were diagnosed with prostate cancer (table 2). 1939 (19%) patients in the testosterone replacement therapy group died, 586 (6%) had a cardiovascular event, and 259 (3%) were diagnosed with prostate cancer.

The median time to cardiovascular event was 30·4 months (IQR 13·7–54·5) for patients receiving testosterone replacement therapy and 30·9 months (13·5–54·7; p=0·95).

When we examined the effect of testosterone replacement therapy as a dichotomous variable, 5-year cumulative mortality was significantly lower in patients treated with testosterone replacement therapy (15·4% [95% CI 14·6–16·1]) than in controls (17·7% [17·3–18·2]; p<0·0001; figure 1). Because fewer than 50% of patients in each group died, we were unable to calculate median

	Dichotomous analysis		Cumulative exposure analysis		
	No testosterone replacement therapy (n=28 029)	Testosterone replacement therapy (n=10 311)	Tertile 1 (n=3595)	Tertile 2 (n=3332)	Tertile 3 (n=3384)
Mortality	5782 (21%)	1939 (19%)	739 (20·56%)	669 (20·08%)	531 (15·69%)
Cardiovascular events	1465 (5%)	586 (6%)	200 (5·56%)	195 (5·85%)	191 (5·64%)
Prostate cancer	799 (3%)	259 (3%)	94 (2·61%)	85 (2·55%)	80 (2·36%)

Data are n (%).

Table 2: **Crude event rates, stratified by treatment group**

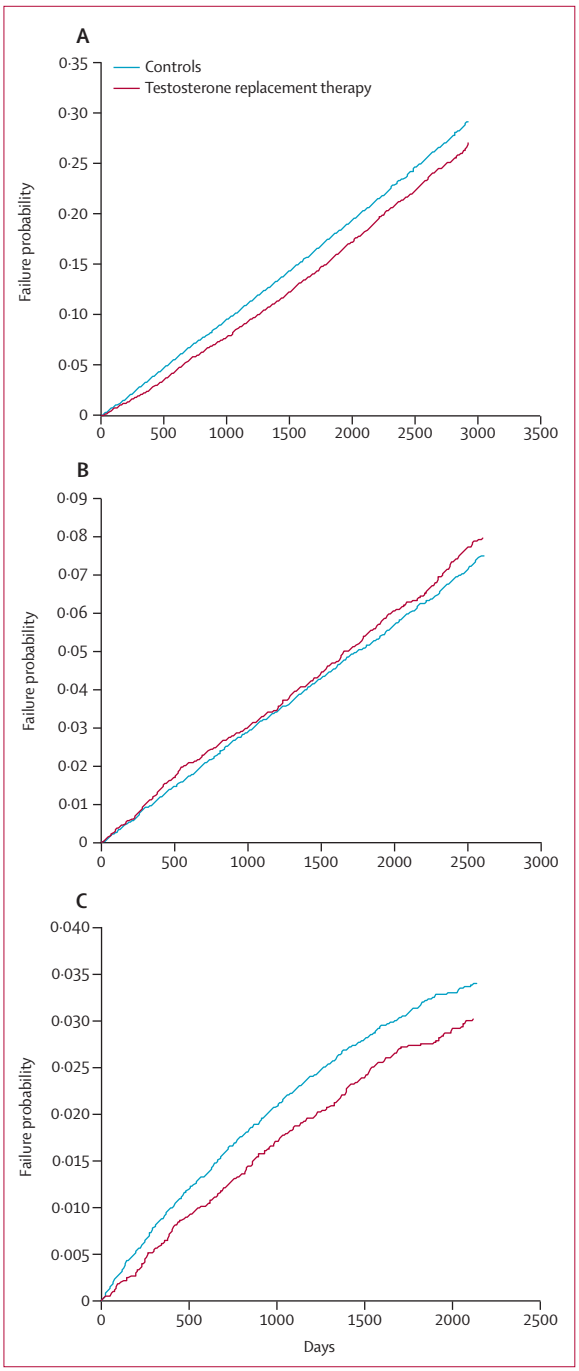

Figure 1: **Kaplan-Meier cumulative incidence of overall mortality, cardiovascular events, and prostate cancer diagnoses, with testosterone exposure as a binary variable**

(A) Overall mortality: 5-year cumulative incidence was 17·7% (95% CI 17·3–18·2) in the control group and 15·4% (95% CI 14·6–16·1; p<0·0001) in those exposed to testosterone replacement therapy. (B) Cardiovascular events: 5-year cumulative incidence was 5·2% (95% CI 4·9–5·5) in the control group and 5·5% (95% CI 4·9–6·0; p=0·25) in those exposed to testosterone replacement therapy. (C) Prostate cancer diagnoses: 5-year cumulative incidence was 3·2% (95% CI 2·9–3·4) in the control group and 2·8% (95% CI 2·4–3·1%; p=0·04) in those exposed to testosterone replacement therapy.

overall survival. In a matched-analysis model, testosterone exposure was associated with a decreased risk of overall mortality (hazard ratio [HR] 0·88, 95% CI 0·84–0·93; table 3).

To better examine the exposure effect of testosterone treatment, we assessed testosterone replacement therapy based on the tertile distribution of cumulative dose exposure. Patients in the lowest tertile had higher mortality than controls, but those in the higher tertiles had lower mortality than controls (figure 2), although we were unable to account for immortal-time bias in the Kaplan-Meier analysis. In a matched-analysis model that used the tertile distribution of testosterone replacement therapy exposure and a time-varying exposure to account for potential immortal-time bias, patients in the lowest tertile of exposure had higher mortality than controls. The risk of mortality was progressively lower with increasing exposure to testosterone, with a significantly decreased risk for patients with the highest tertile of exposure (table 3).

5-year cumulative incidence of cardiovascular events did not differ significantly in the testosterone replacement therapy group compared with controls (5·2% [95% CI 4·9–5·5] in the control group *vs* 5·5% [4·9–6·0] in those exposed to testosterone replacement therapy; p=0·25; figure 1), although the 5-year cumulative incidence of prostate cancer diagnoses was lower in the testosterone replacement therapy group (3·2% [2·9–3·4] in the control group *vs* 2·8% [2·4–3·1%] in those exposed to testosterone replacement therapy; p=0·04; figure 1). In a matched analysis in which testosterone replacement therapy was assessed as a dichotomous exposure, testosterone exposure was not associated with risk of cardiovascular events, but was associated with a decreased risk of prostate cancer (table 3).

When we assessed testosterone replacement therapy based on tertiles of cumulative dose exposure, patients in the lowest tertile had higher cumulative incidence rates of cardiovascular events than did controls, but those in the highest tertile had lower rates than controls (figure 2). Similarly, patients in the lowest tertile of exposure had equivalent rates of prostate cancer diagnosis to controls, whereas those in the highest tertile had lower rates than controls. In a matched analysis, we found an increased risk of cardiovascular events (HR 1·26, 95% CI 1·09–1·46) for patients in the lowest tertile of exposure to testosterone replacement therapy, compared with controls not exposed to testosterone replacement therapy (table 3). We identified no association with prostate cancer diagnoses among patients in the lowest tertile of exposure (table 3). However, the risk of each outcome was progressively lower for patients in the middle and highest tertile of testosterone exposure (p for trend <0·0001 [middle tertile] and p for trend<0·03 [highest tertile]). Patients in the highest tertile had a lower risk for cardiovascular events (HR 0·84, 0·72–0·98) and prostate cancer diagnoses (HR 0·60, 0·45–0·80) than controls (table 3).

The median duration of testosterone replacement therapy was 2 months (IQR 1–3) or 60 days (IQR 30–90; range 1–120) for patients in the lowest tertile, 9 months (IQR 6–12) or 270 days (IQR 180–360; range 121–510) for those in the middle tertile, and 35 months (25–51) or 1050 days (IQR 740–1530; range 511–4145) for those in the highest tertile.

The results of our post-hoc analyses suggest that the apparent protective effect against mortality of testosterone when assessed as a binary exposure is greatest in the early period (appendix p 5). When testosterone exposure was assessed as a time-varying exposure, the results supported those of our primary analysis (appendix). Patients in the lowest tertile of exposure had a numerically higher risk of death (no statistical comparison was done) in the early period than the late period, whereas those in the middle tertile had a lower risk in the early period than in the late period. The highest tertile could not be assessed in the early period due to the length of exposure required to meet the highest tertile definition (appendix).

	Dichotomous analysis		Testosterone replacement therapy dose exposure		
	No testosterone replacement therapy	Testosterone replacement therapy *vs* controls	Tertile 1 (short exposure)	Tertile 2 (intermediate exposure)	Tertile 3 (long exposure)
Mortality	1·0	0·88 (0·84–0·93, p<0·0001)	1·11 (1·03–1·20, p=0·005)	0·90 (0·83–0·97, p=0·007)	0·67 (0·62–0·73, p<0·0001)
Cardiovascular events	1·0	1·10 (1·00–1·20, p=0·05)	1·26 (1·09–1·46, p=0·002)	1·16 (1·00–1·35, p=0·05)	0·84 (0·72–0·98, p=0·02)
Prostate cancer	1·0	0·86 (0·75–0·99, p=0·04)	0·92 (0·70–1·21, p=0·6)	1·12 (0·88–1·44, p=0·4)	0·60 (0·45–0·80, p=0·0005)

Results are hazard ratios (HRs [95% CI]). All p values derived from comparisons with the no testosterone replacement therapy group as reference. Results adjusted for the effect of age, comorbidity, index year, and geographical region by matching.

Table 3: **Results of survival model assessing the effect of testosterone replacement therapy on the primary and secondary outcomes**

Discussion

In this large, population-based cohort study, we identified an inverse association between cumulative testosterone replacement therapy exposure and mortality and cardiovascular events. Short durations of therapy (median 2 months) were associated with increased risks of mortality and cardiovascular events, whereas longer durations of therapy (median 35 months) were associated with reduced mortality and cardiovascular events, compared with matched controls. Risk of prostate cancer was not increased for patients in the lowest tertile of testosterone exposure, but showed reduced risk with increasing testosterone exposure.

To our knowledge, this is the first study to assess the effect of cumulative dose exposure of testosterone therapy. As a result of this approach, we were able to account for the previous conflicting findings of the effect of testosterone replacement therapy on cardiovascular events and mortality. With a large sample size and long duration of follow-up, we identified a beneficial association of long-term testosterone replacement therapy, but also showed that short-term durations of therapy might be associated with harm. Furthermore, we explored the effect of testosterone on early versus late events. Using a time-varying exposure, we found that patients during their first 120 days of therapy had an increased risk of early death, a risk that subsequently dissipated with time or ongoing treatment.

In a retrospective cohort study, Vigen and colleagues[4] showed that testosterone replacement therapy increased the risk of cardiovascular events (myocardial infarction, stroke, and death) among hypogondal men undergoing angiography. However, concerns with that study included selection bias from the exclusion of events before testosterone replacement therapy, misclassification of the exposure,[12] inclusion of women in the analysis, and inadequate treatment.[32] Analysis of the Testosterone in Older Men with Mobility Limitations randomised controlled trial showed an excess of cardiovascular events in men receiving testosterone,[33] but it was neither designed nor powered for this outcome. Patients in this study received a maximum of 6 months of testosterone therapy and were given doses beyond recommended levels. Finkle and colleagues[5] also identified an increased risk of myocardial infarction after initiation of testosterone replacement therapy, but treatment duration and follow-up were limited to a maximum of 90 days. The treatment given in these studies might be most directly compared with that received by the subset of patients in the lowest tertile of testosterone exposure in our analysis. By contrast, studies assessing patients with long-term use of testosterone replacement therapy have shown a reduced risk of myocardial infarction[7] and overall mortality.[9,10]

In our cohort, the median duration of testosterone replacement therapy in the lowest tertile was 2 months (IQR 1–3). Since the median time to cardiovascular event was 30·4 months (13·7–54·5) for patients on testosterone replacement therapy, the increased risk of cardiovascular events for men in the lowest tertile of testosterone exposure could be driven by underlying testosterone deficiency and inadequate treatment, in view of the known association between testosterone deficiency and cardiovascular disease,[1,3] rather than their testosterone treatment.

Before initiation of testosterone replacement therapy, men are likely to undergo prostate cancer screening and are therefore at low baseline risk. As a result, the decrease in risk of prostate cancer diagnosis identified in our study might be a result of bias. Because of the limitations of our data, we were unable to obtain prostate-specific antigen concentrations, or histological data for the patients diagnosed with prostate cancer. Our findings for this outcome support those of previous meta-analyses,

which showed no increased risk of prostate cancer for patients receiving testosterone replacement therapy.[34]

Although we matched patients treated with testosterone replacement therapy to controls based on cardiovascular risk factors, we could not match based on the presence of hypogonadism because this is not reliably documented in Ontario[28] and testosterone measurements are not available at the administrative level. As a result, we were unable to compare patients with hypogonadism with and without testosterone replacement therapy, which would have been preferable apporach. The absence of testosterone measurements also precluded assessment of the adequacy of therapy. Based on data suggesting that adequate testosterone replacement (defined as serum testosterone in the mid-normal range) is necessary for survival and cardiovascular benefit, a calculated duration of normalised testosterone would provide an alternative measure of exposure to the cumulative dose that we used.[8]

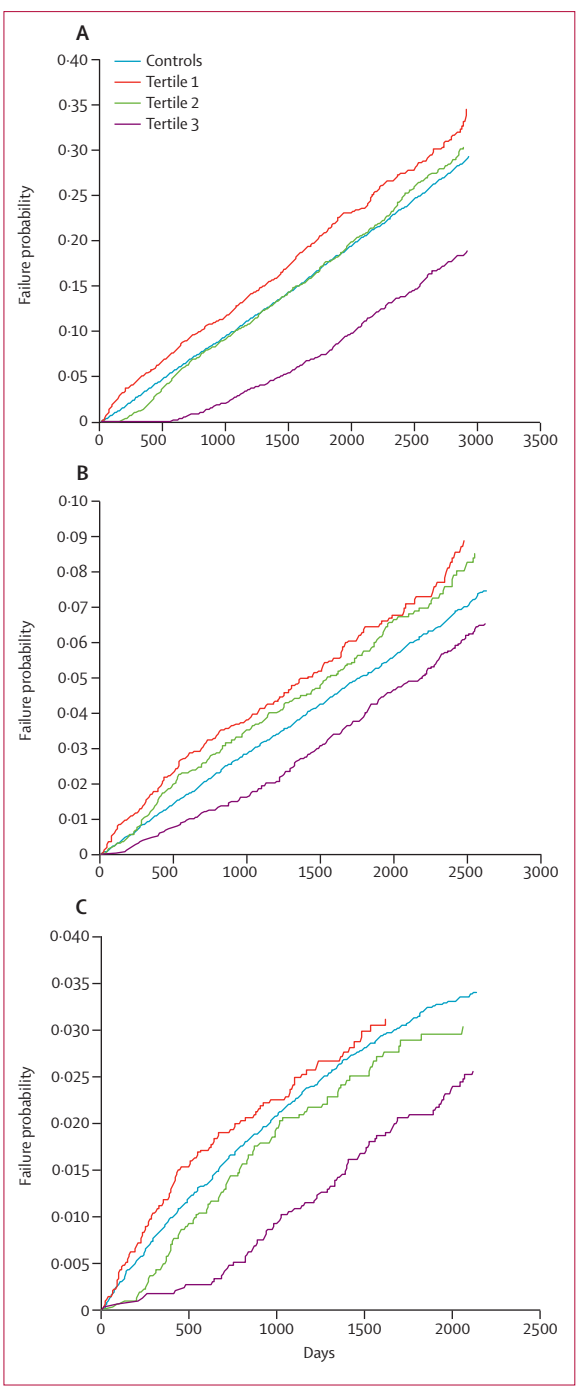

Figure 2: **Kaplan-Meier cumulative incidence of overall mortality, cardiovascular events, and prostate cancer diagnoses, with tertile-based testosterone exposure**
(A) Overall mortality: 5-year cumulative incidence was 17·7% (95% CI 17·3–18·2%) in the control group, 21·4% (95% CI 19·8–22·9%) in tertile 1 (the tertile with the lowest exposure), 17·7% (95% CI 16·3–19·1%) in tertile 2 (the middle tertile), and 7·9% (95% CI 7·0–8·8%; p<0·0001) in tertile 3 (the tertile with the highest exposure) of testosterone replacement therapy cumulative exposure. (B) Cardiovascular events: 5-year cumulative incidence was 5·2% (95% CI 4·9–5·5%) in the unexposed group, 6·5% (95% CI 5·5–7·4%) in tertile 1, 5·8% (95% CI 4·9–6·7%) in tertile 2, and 4·2% (95% CI 3·5–4·9%; p=0·0002) in tertile 3 of cumulative exposure to testosterone replacement therapy. (C) Prostate cancer diagnoses: 5-year cumulative incidence was 3·2% (95% CI 2·9–3·4%) in the unexposed group, 3·1% (95% CI 2·5–3·7%) in tertile 1, 3·0% (95% CI 2·3–3·6%) in tertile 2, and 2·1% (95% CI 1·6–2·6%; p=0·03) in tertile 3 of cumulative exposure to testosterone replacement therapy. Because of limitations of the Kaplan-Meier method, the tertile-based exposure presented here is unable to account for time-varying exposure.

To reduce heterogeneity between men receiving testosterone replacement therapy and those never exposed, we excluded patients with a history of cardiovascular events in the 5 years before the index date. Previous work has suggested that such men might be at increased risk for cardiovascular events after initiation of testosterone replacement therapy.[5] Thus, the effects of testosterone replacement therapy on cardiovascular events among men with a history of cardiovascular events requires further investigation, and these findings cannot be generalised to such men.

Our study and others have assessed cardiovascular events and mortality; in cardiology research, revascularisation is often also included among major cardiac events analysed in trials. Future research into the effects of testosterone replacement therapy with revascularisation as an independent endpoint or included within a composite endpoint would be worthwhile.

The association between testosterone therapy and decreased mortality identified in our study and others[10] in part reflects a healthy user bias, resulting in overestimation of the effect of testosterone replacement therapy. Patients who seek assessment for testosterone deficiency and subsequently receive a prescription for testosterone replacement therapy are likely to engage with the health-care system and participate in healthy behaviours to a greater extent than the general population. Residual confounding remains a potential source of bias because of the observational study design. Therefore, these data require validation in a randomised trial. We would hope that such a trial would be pragmatic, so that not too many patients in routine clinical practice would be excluded from the study. With the results of our binary analysis assessing overall mortality and standard assumptions to prevent false-positive and false-negative findings, such a study would require roughly 2000 events (or deaths) to occur.

Our study has many strengths, including a large, population-based design and long duration of follow-up. Data from one observational study provided longer follow-up (5·8 years), but only included 64 men treated with testosterone replacement therapy.[10] Additionally, although many researchers have investigated testosterone exposure in a binary way, we studied it as a cumulative dose exposure. Assessing cumulative dose exposure avoids two potential pitfalls of a binary approach: bias by indication due to the known association between testosterone deficiency and cardiovascular disease and non-differential misclassification due to overly simplistic exposure

definitions.[31] Because patients must survive at least 1 year in order to receive their 13th month of testosterone replacement therapy prescription, risk for immortal time bias exists when assessing cumulative dose exposure, which would bias towards lower risk in patients with longer exposure. To avoid this bias, we used a time-varying exposure in which we partitioned both exposure and outcome times into tertiles such that each man exposed to testosterone replacement therapy can contribute to all three tertiles.[29,30] Furthermore, we did our analysis in the style of an intention-to-treat observational cohort study, as proposed by Hernan and colleagues[35] to most closely emulate a randomised controlled trial.

In conclusion, although we showed a decreased risk of mortality and cardiovascular events in men on long-term testosterone replacement therapy, these results should be considered hypothesis-generating rather than definitive. We do not believe that these data provide sufficient evidence for a new indication for use of testosterone in prevention of cardiovascular disease. Rather, these findings might inform physicians counselling older men about the potential risks and benefits of testosterone replacement therapy before initiation of therapy. Because of potential biases in observational studies, these findings require validation in a randomised controlled trial.

Contributors

CJDW, KL, and RKN formulated the clinical question and designed the study. CJDW acquired and analysed the data. CJDW, KL., YL, YK, AG, SH, RT.K, PC, SAN, and RKN were involved in data interpretation. CJDW drafted the report. KL, YL, YK, AG, RS, SH, RTK, PC, SAN, and RKN. critically revised the report. CJDW and RKN obtained funding for the study. RKN provided administrative and technical support.

Declaration of interests

We declare no competing interests.

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