# Testosterone, Dihydrotestosterone, and Incident Cardiovascular Disease and Mortality in the Cardiovascular Health Study 

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Context: Low testosterone (T) is associated with prevalent cardiovascular disease (CVD) and mortality. DHT, a more potent androgen, may also be associated with CVD and mortality, but few studies have examined this.

Objective: The study objective was to examine whether T and DHT are risk factors for incident CVD and mortality.

Design: In a longitudinal cohort study, we evaluated whether total T, calculated free T (cFT), DHT, and calculated free DHT were associated with incident CVD and mortality in men in the Cardiovascular Health Study (mean age 76, range 66-97 years) who were free of CVD at the time of blood collection.

Main Outcome: The main outcomes were incident CVD and all-cause mortality.
Results: Among 1032 men followed for a median of 9 years, 436 incident CVD events and 777 deaths occurred. In models adjusted for cardiovascular risk factors, total T and cFT were not associated with incident CVD or all-cause mortality, whereas DHT and calculated free DHT had curvilinear associations with incident CVD ( $P<.002$ and $P=.04$, respectively) and all-cause mortality ( $P<.001$ for both).

Conclusions: In a cohort of elderly men, DHT and calculated free DHT were associated with incident CVD and all-cause mortality. Further studies are needed to confirm these results and to clarify the underlying physiologic mechanisms. (J Clin Endocrinol Metab 99: 2061-2068, 2014)

Cardiovascular disease (CVD) is a major cause of morbidity and mortality in older men. The increased burden of CVD in aging men may be related to concomitant decreases in serum testosterone $(\mathrm{T})$ levels, which have been

[^0]associated with risk factors for CVD, including increased body mass index (BMI), waist circumference, insulin levels, inflammatory markers, dyslipidemia, diabetes, hypertension, peripheral arterial disease, and atherosclerosis
(1-5). Although low T levels are associated with risk factors for CVD, it is unclear whether low T is independently associated with risk for incident CVD. Previous case-control studies reported no association between low T and incident CVD in men, $(6-8)$, whereas more recent studies reported associations with low T and risk for CVD (9-16). Many of the recent studies that reported an association between low T and CVD included men with prevalent CVD who were less than 70 years old, and CV mortality was often the sole CV outcome. As a consequence, uncertainty remains about whether low T is a risk factor for men without preexisting CVD, for men older than 70 years, and for nonfatal CV events.

In addition to the potential association of T with incident CVD, DHT, which is a much more potent androgen than T, may also affect CVD risk. Testosterone is converted to DHT via the enzyme $5 \alpha$-reductase in prostate,
skin, and hair follicles and acts locally in these tissues. Serum DHT derives primarily from these tissues and the liver. However, the physiological significance of serum DHT is poorly studied and unclear, in large part because accurate measurements of serum DHT have not been available. Serum DHT could influence CVD risk by mechanisms involving inflammation, platelets, and vasoreactivity $(17,18)$. Previous studies have found that low serum DHT is associated with CVD and ischemic heart disease mortality (19, 20). However, these studies included men with and without prevalent CVD.

In this study, we evaluated the association of T and DHT with incident CVD and all-cause mortality in elderly men in the Cardiovascular Health Study (CHS). Based on past studies, we hypothesized that low levels of T and DHT would be associated with an increased risk for in-

Table 1. Baseline Characteristics ${ }^{\text {a }}$

## Total T

| Characteristic | $\geq 278$ ng/dL | <278 ng/dL | $P$ Value |
| :---: | :---: | :---: | :---: |
| n | 776 | 256 |  |
| Age, y | 76.3 (4.9) | 77.2 (5.8) | . 02 |
| African-American, \% | 13.5 | 18.8 | . 04 |
| High school education or less, \% | 47.7 | 53.9 | . 09 |
| Smoking status, \% |  |  |  |
| Never | 25.4 | 32.0 |  |
| Former | 62.9 | 55.9 |  |
| Current | 11.7 | 12.1 | . 09 |
| Alcoholic drinks per week, \% |  |  |  |
| None | 42.9 | 50.4 |  |
| $<7$ | 36.7 | 29.3 |  |
| 7-13 | 10.2 | 9.8 |  |
| $\geq 14$ | 10.2 | 10.6 | . 14 |
| BMI, $\mathrm{kg} / \mathrm{m}^{2 \mathrm{~b}}$ | 26.2 (3.5) | 28.1 (4.0) | <. 001 |
| Waist circumference, $\mathrm{cm}^{\text {c }}$ | 97.7 (10.0) | 102.4 (10.9) | <. 001 |
| Physical activity, $\mathrm{kcal}^{\text {c }}$ | 1978.5 (2041.6) | 1644.0 (1712.7) | . 02 |
| Systolic BP, mm Hg | 131.6 (19.0) | 133.9 (21.8) | . 11 |
| Diastolic BP, mm Hg | 71.0 (10.8) | 71.3 (11.0) | . 70 |
| Total cholesterol, mg/dL | 189.0 (34.6) | 186.6 (36.0) | . 35 |
| LDL cholesterol, mg/dL ${ }^{\text {c }}$ | 116.3 (31.8) | 111.2 (30.0) | . 03 |
| HDL cholesterol, mg/dL ${ }^{\text {c }}$ | 48.6 (12.0) | 45.9 (10.9) | . 002 |
| Triglycerides, mg/dL ${ }^{\text {c }}$ | 128.1 (67.4) | 161.2 (105.3) | <. 001 |
| Glucose, mg/dL | 111.3 (42.9) | 132.4 (63.2) | <. 001 |
| Insulin, IU/mL ${ }^{\text {c }}$ | 11.7 (16.6) | 14.8 (18.5) | . 01 |
| Hypertension, \% | 49.7 | 62.9 | <. 001 |
| Diabetes, \% | 13.9 | 27.3 | <. 001 |
| Self-reported health good/excellent, \% | 85.2 | 80.4 | . 07 |
| CES-D score | 4.5 (4.1) | 5.1 (4.6) | . 05 |
| Aspirin >2 days in past 2 weeks, \% | 38.4 | 31.1 | . 04 |
| Diuretics, \% | 16.7 | 22.4 | . 04 |
| Antihypertensives, \% | 42.3 | 57.0 | <. 001 |
| Calcium channel blockers, \% | 13.8 | 23.9 | <. 001 |
| Digitalis, \% | 5.3 | 7.5 | . 20 |
| Lipid-lowering medication, \% | 4.4 | 7.1 | . 09 |

[^1]cident CVD and all-cause mortality in older men who had no known history of CVD.

## Subjects and Methods

## Study population

The CHS is a longitudinal cohort study that was initiated in 1989 to identify risk factors for CVD in older adults (21). From 1989 to 1990,5201 participants were enrolled and from 1992 to 1993, an additional 687 African American participants were enrolled. Eligible participants were 65 years or older, noninstitutionalized, expected to stay in the area for 3 years, and able to give informed consent. Those excluded were wheelchair-bound, hospice patients or receiving cancer treatment. Each study center's institutional review board approved the study, and each participant provided written informed consent. Clinic examinations were performed annually from 1989 to 1999 and again in 2005. Our study sample consisted of men participating in the CHS clinical examination in 1994 who had no history of prostate cancer or CVD (defined as myocardial infarction [MI], coronary artery bypass grafting, percutaneous coronary intervention, heart failure, or stroke) at that visit. Frozen sera from the 1994 visit were used to measure total T and DHT. Incident CVD and deaths were classified by the CHS Events Committee, which used standardized algorithms to adjudicate CVD outcomes and cause of death using information from Medicare, hospital records, death certificates, autopsy reports, and interviews with research subjects, relatives, and attending physicians (21). Event adjudication was available through December 2010. The CVD outcome consisted of incident MI, incident stroke, and CV mortality.

## Hormone assays

Blood samples were obtained from participants at the 1994 CHS examination and were stored at $-70^{\circ} \mathrm{C}$ at the CHS Central


Figure 1. Distributions of total and free T and DHT among 1032 men in the CHS. Means (SDs) are as follows: total T, 389 (176) ng/dL; free T, 5.3 (2.2) ng/dL; DHT, 45 (23) ng/dL; free DHT, $0.26(0.13) \mathrm{ng} / \mathrm{dL}$.

Laboratory in Burlington, Vermont. The time of day for sample collection was not recorded, but the majority were likely collected in the morning, based on the order of data collection in the protocol. In 2010, frozen serum samples were shipped on dry ice to an endocrine research laboratory (conducted in laboratory of author A.M.M.) that has over 20 years of experience in conducting hormone assays. Total T and DHT were measured simultaneously using a liquid chromatography-tandem mass spectrometry assay (22). All assays were conducted in duplicate, and the average value was used in these analyses. The lower limit of detection for total T was $1.0 \mathrm{ng} / \mathrm{dL}$, with an intra-assay coefficient of variation of $4.9 \%$ and an interassay coefficient of variation of $5.1 \%$. The Centers for Disease Control and Prevention Hormone Standardization Program certified the serum T assay over a range of 1.0 to $2000 \mathrm{ng} / \mathrm{dL}$. The lower limit of detection for DHT was $0.02 \mathrm{ng} / \mathrm{mL}$ with an intra-assay coefficient of variation of $5.9 \%$ and an interassay coefficient of variation of $6.2 \%$. SHBG was assayed using a time-resolved fluoroimmunoassay (Delfia; PerkinElmer). The lower limit of detection for SHBG was $0.5 \mathrm{nmol} / \mathrm{L}$, with an intra-assay coefficient of variation of $1.4 \%$ and an interassay coefficient of variation of $6.6 \%$ at 31 $\mathrm{nmol} / \mathrm{L}$. We determined calculated free $\mathrm{T}(\mathrm{cFT})$ by the Vermeulen (23) and Mazer methods (24). The Mazer method uses a mass action model with an iterative approach, and the Vermeulen method uses a mass action model that has been validated via equilibrium dialysis. The cFT values by the Mazer and Vermeulen methods were highly correlated ( $r=0.998$ ), which is not unexpected because they both use mass-action models. In our analyses, we used the Mazer formula, rather than the Vermeulen formula, because it allowed calculation of both free T and free DHT (24).

## Covariates

Descriptions of data collection methods, including instruments and protocols, have been reported previously (21). The covariates were measured at the 1994 visit, unless otherwise noted, and included known risk factors for CVD reported in previous CHS studies (25). Age, race, educational level, smoking status, and usual consumption of alcoholic drinks were based on self-report. Physical activity (kilocalories per week), weight, height, and waist circumference were assessed at the 1992 examination. Blood pressure (BP) was measured using standardized protocols. Laboratory measures included glucose, insulin, and lipids. The insulin and lipid measures were obtained during the 1992 examination. We defined diabetes as fasting glucose $\geq 126 \mathrm{mg} / \mathrm{dL}$, nonfasting glucose $\geq 200 \mathrm{mg} / \mathrm{dL}$, or use of diabetes medication. We defined hypertension as systolic BP $\geq 140 \mathrm{~mm} \mathrm{Hg}$, diastolic $\mathrm{BP} \geq 90 \mathrm{~mm}$ Hg , or physician diagnosis of hypertension combined with use of antihypertensive medication. Medication use was ascertained using a validated medication inventory.

## Statistical analysis

The group of men with low hormone levels was defined a priori as men who had hormone values in the lowest quar-
tile of each hormone's respective distribution. Men with low hormone levels were compared with men who had hormone levels above the lowest quartile. To maximize statistical power, we also modeled each hormone continuously. We used generalized additive models and penalized regression splines to explore the functional form of the relationship between the hormones and the outcomes. Nonlinearity of associations was tested with the gain statistic (26). Based on the spline plots, the simplest functional form that adequately characterized each association was selected; linear associations were modeled with a linear term, and curvilinear associations were modeled using linear and quadratic terms. A quadratic term was only included if the $P$ value for the quadratic term was $<.05$ by the Wald test. In the case of a quadratic model, the overall $P$ value for the hormone was determined by a likelihood ratio test, comparing a model without the linear and quadratic terms to one with both terms. To aid in the interpretation of the nonlinear models, we categorized the hormone into groups of equal length (approximately equal to 1 SD ) to identify relative risk estimates for each category compared with the referent group (the interval with the lowest risk). See "http://press.endocrine.org/doi/suppl/10.1210/ jc.2013-3576/suppl_file/jc-13=3576.pdf" Supplemental Table 1 for additional information on parameterization of the nonlinear model.

We used Cox proportional hazards regression models to estimate the relative risk of incident CVD and all-cause mortality
associated with total T, cFT, DHT, and calculated free DHT. Time at risk was calculated as the interval between the 1994 study examination when sera were obtained (baseline) and date of incident CVD, death, or end of follow-up (December 2010). We evaluated the validity of the proportional hazards assumption using Schoenfeld residuals and found no meaningful violations. A series of sequential models were fit for each hormoneoutcome pair, starting with known confounders, and then covariates were added for which there is more uncertainty regarding whether the variable is a confounder or an intermediate. Model 1 was adjusted for age, race, clinic site, smoking status (never, former, or current), and alcohol consumption. Model 2 was additionally adjusted for hypertensive use, high-density lipoprotein (HDL) cholesterol, BMI, and waist circumference. Model 3 was additionally adjusted for diabetes status, a potential mediator of the association. We conducted a sensitivity analysis in which men who were treated with finasteride, which lowers DHT levels, were excluded from the analysis. Finally, to evaluate the reliability of a single hormone measure to capture variability within an individual over several years, we used a random-effects ANOVA to estimate the intra-class correlations coefficient (ICC) and $95 \%$ confidence intervals for T and DHT across 3 time points from a subset of 74 participants who had repeated hormone measures available from 1989, 1994, and 1996. Results are reported as mean (SD).

Table 2. Association Between Androgens and Risk of Incident CVD (MI, Stroke, or CVD Death)

| Concentration | No. of Events | Incidence per 1000 | Hazard Ratio (95\% Confidence Intervals) |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  | Model $1^{\text {a }}$ | Model $\mathbf{2}^{\text {a }}$ | Model $3^{\text {a }}$ | Model $4^{\text {a }}$ |
| Total T, ng/dL |  |  |  |  |  |  |
| <278 | 123 | 57.0 | 1.28 (1.03-1.58) | 1.16 (0.92-1.45) | 1.12 (0.89-1.41) | 1.11 (0.87-1.43) |
| $\geq 278$ | 313 | 43.4 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |
| Per SD decrease ${ }^{\text {b }}$ | 436 | 46.6 | 1.05 (0.95-1.16) | 1.00 (0.90-1.11) | 0.98 (0.89-1.09) | 0.96 (0.85-1.08) |
| cFT, ng/dL |  |  |  |  |  |  |
| <4.1 | 109 | 49.6 | 0.96 (0.76-1.20) | 0.89 (0.70-1.12) | 0.87 (0.69-1.10) |  |
| $\geq 4.1$ | 327 | 45.6 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |  |
| Per SD decrease ${ }^{\text {b }}$ | 436 | 46.6 | 1.03 (0.93-1.13) | 1.00 (0.90-1.10) | 0.99 (0.90-1.09) |  |
| DHT, ng/dL |  |  |  |  |  |  |
| <25 | 84 | 63.2 | 1.70 (1.26-2.28) | 1.52 (1.11-2.08) | 1.44 (1.05-1.98) | 1.48 (1.06-2.08) |
| 25-49 | 209 | 46.7 | 1.35 (1.06-1.73) | 1.32 (1.03-1.70) | 1.31 (1.02-1.69) | 1.30 (1.00-1.70) |
| 50-74 | 95 | 36.3 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |
| $\geq 75$ | 48 | 51.0 | 1.34 (0.94-1.89) | 1.36 (0.95-1.94) | 1.30 (0.91-1.86) | 1.42 (0.99-2.04) |
| $P$ value ${ }^{\text {c }}$ |  |  | <. 001 | <. 001 | . 002 | . 002 |
| <30 | 124 | 57.8 | 1.36 (1.10-1.67) | 1.21 (0.97-1.51) | 1.17 (0.93-1.46) | 1.21 (0.97-1.51) |
| $\geq 30$ | 312 | 43.2 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |
| Calculated free |  |  |  |  |  |  |
| DHT, ng/dL |  |  |  |  |  |  |
| $<0.13$ | 51 | 66.0 | 1.67 (1.07-2.62) | 1.66 (1.06-2.60) | 1.62 (1.03-2.54) |  |
| 0.13-0.25 | 199 | 47.7 | 1.32 (0.91-1.92) | 1.33 (0.91-1.95) | 1.34 (0.92-1.97) |  |
| 0.26-0.38 | 139 | 43.6 | 1.28 (0.87-1.88) | 1.40 (0.95-2.07) | 1.41 (0.95-2.07) |  |
| 0.39-0.51 | 32 | 36.3 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |  |
| $\geq 0.52$ | 15 | 43.4 | 1.24 (0.67-2.30) | 1.47 (0.79-2.73) | 1.43 (0.77-2.66) |  |
| $P$ value ${ }^{\text {c }}$ |  |  | . 008 | . 04 | . 04 |  |
| $<0.18$ | 105 | 57.8 | 1.22 (0.97-1.52) | 1.13 (0.90-1.42) | 1.11 (0.88-1.40) |  |
| $\geq 0.18$ | 331 | 43.9 | 1.00 (Ref.) | 1.00 (Ref.) | 1.00 (Ref.) |  |

[^2]
## Results

The mean age of men in the study was 76.5 (5.2) years (range 66-97), and most men ( $84 \%$ ) rated their health as good to excellent (Table 1). Men with low total T levels were older and had higher BMI and waist circumference and a greater prevalence of dyslipidemia, diabetes, and hypertension than men with higher total T levels (Table 1). Distributions of the hormones were skewed with mean (SD) levels of 389 (176) ng/dL total T, 5.3 (2.2) ng/dL cFT, $45(23) \mathrm{ng} / \mathrm{dL}$ DHT, and $0.26(0.13) \mathrm{ng} / \mathrm{dL}$ calculated free DHT (Figure 1). Over a median follow-up of 8.9 years, 436 men had an incident CVD event ( $\mathrm{n}=174$ MIs, 105 strokes, and 157 CV deaths). Over a median follow-up of 10.8 years (maximal follow-up of 16.6 years), 777 men died.

The best model for total T and cFT was a linear model, whereas the best model for DHT and calculated free DHT was a nonlinear one. In fully adjusted analyses, total T and cFT had no significant linear association with incident CVD or mortality (Table 2), whereas DHT ( $P=.002$ ) and calculated free DHT ( $P=.04$ ) had significant curvilinear associations with incident CVD (Table 2 and Figure 2) and all-cause mortality ( $P<.001$ for both) (Table 3 and Figure 2). Results were not substantively changed when SHBG was added to the models for total T and DHT (Tables 2
and 3). SHBG was not associated with incident CVD or mortality in fully adjusted analyses. (The association between hormones and CV mortality is provided in Supplemental Table 2). In a separate sensitivity analysis that excluded men who were taking finasteride ( $\mathrm{n}=31$ ), the results for total T, cFT, DHT, and calculated free DHT were not appreciably different (results not shown). The ICCs for 3 hormone measures over a 7 -year period were 0.82 for total T and 0.79 for DHT.

## Discussion

In this longitudinal study of community-dwelling elderly men free of baseline CVD, total T and cFT were not significantly associated with incident CVD or all-cause mortality in fully adjusted analyses. DHT had curvilinear associations with incident CVD and all-cause mortality in analyses adjusted for CV risk factors with the lowest risk at DHT levels of 50 to $74 \mathrm{ng} / \mathrm{dL}$. DHT concentrations lower than $50 \mathrm{ng} / \mathrm{dL}$ were inversely associated with risk for CVD and mortality, whereas DHT concentrations greater than $74 \mathrm{ng} / \mathrm{dL}$ appeared to be directly associated with risk. However, although the spline plot suggested a U- or J-shaped association between DHT and incident CVD and mortality, the wide confidence intervals at higher hormone levels reflect sub-


Figure 2. Spline regression graphs depicting the associations between continuous levels of total and free DHT and incident CVD (top 2 panels) and all-cause mortality (bottom 2 panels). The solid line represents the estimated hazard ratio, and the shaded area depicts the $95 \%$ confidence intervals. All models are adjusted for age.
stantial uncertainty regarding the form of the association at higher levels. Due to the wide confidence intervals at higher DHT levels, it is also possible that risk plateaus, rather than increases, at high DHT levels. Similar curvilinear associations were found for calculated free DHT and risk of incident CVD and mortality. The association of DHT with incident CVD is consistent with studies that reported associations between serum DHT and CVD and ischemic heart disease mortality in men (20, 27). The curvilinear associations are consistent with recent studies that reported nonlinear associations of androgen levels and adverse outcomes (20, 28).

Although the basis of the association between DHT and incident CVD and mortality will require further study, a causal association could have important clinical implications. One potential implication is that

Table 3. Association Between Androgens and All-Cause Mortality

| Concentration | No. of Deaths | Mortality per 1000 | Hazard Ratio (95\% Confidence Intervals) |  |
| :---: | :---: | :---: | :---: | :---: |
|  |  |  | Model $1^{\text {a }}$ | Model $2^{\text {a }}$ |
| Total T, ng/dL |  |  |  |  |
| <278 | 196 | 77.1 | 1.05 (0.88-1.25) | 1.06 (0.88-1.29) |
| $\geq 278$ | 581 | 71.2 | 1.00 (Ref.) | 1.00 (Ref.) |
| Per SD decrease ${ }^{\text {b }}$ | 777 | 72.6 | 1.03 (0.95-1.11) | 1.05 (0.96-1.14) |
| Free T, ng/dL |  |  |  |  |
| <4.1 | 209 | 83.6 | 1.04 (0.88-1.23) |  |
| $\geq 4.1$ | 568 | 69.3 | 1.00 (Ref.) |  |
| Per SD decrease ${ }^{\text {b }}$ | 777 | 72.6 | 1.07 (0.99-1.16) |  |
| DHT, ng/dL |  |  |  |  |
| <25 | 137 | 87.4 | 1.31 (1.04-1.65) | 1.39 (1.09-1.79) |
| 25-49 | 360 | 69.5 | 1.09 (0.91-1.31) | 1.14 (0.94-1.38) |
| 50-74 | 197 | 68.1 | 1.00 (Ref.) | 1.00 (Ref.) |
| $\geq 75$ | 83 | 78.4 | 0.99 (0.76-1.28) | 0.99 (0.75-1.29) |
| $P$ value ${ }^{\text {c }}$ |  |  | $<.001$ | $<.001$ |
| <30 | 201 | 80.5 | 1.23 (1.04-1.46) | 1.28 (1.06-1.53) |
| $\geq 30$ | 576 | 70.2 | 1.00 (Ref.) | 1.00 (Ref.) |
| Calculated free DHT, ng/dL |  |  |  |  |
| $<0.13$ | 101 | 115.1 | 1.72 (1.25-2.37) |  |
| 0.13-0.25 | 347 | 72.0 | 1.14 (0.87-1.50) |  |
| 0.26-0.38 | 241 | 66.3 | 1.18 (0.89-1.56) |  |
| 0.39-0.51 | 63 | 65.3 | 1.00 (Ref.) |  |
| $\geq 0.52$ | 25 | 61.9 | 1.02 (0.64-1.62) |  |
| $P$ value ${ }^{\text {c }}$ |  |  | <. 001 |  |
| $<0.18$ | 205 | 98.3 | 1.41 (1.19-1.66) |  |
| $\geq 0.18$ | 572 | 66.4 | 1.00 (Ref.) |  |

${ }^{\text {a }}$ Model 1 is adjusted for age, race, clinic site, smoking status, alcohol consumption, systolic BP, antihypertensive use, HDL cholesterol, BMI, and waist circumference. Model 2 is adjusted for model 1 covariates plus SHBG.
${ }^{\mathrm{b}} \mathrm{SD}=181 \mathrm{ng} / \mathrm{dL}$ for total $\mathrm{T}, 2.3 \mathrm{ng} / \mathrm{dL}$ for free testosterone, $0.23 \mathrm{ng} / \mathrm{mL}$ for DHT , and $0.14 \mathrm{ng} / \mathrm{dL}$ for free DHT.
${ }^{c} P$ values test the statistical significance of the linear and quadratic terms for the hormone analyte.
medications that affect DHT levels may adversely affect men's health. Such medications include the $5 \alpha$-reductase inhibitors finasteride and dutasteride, which decrease DHT levels by $60 \%$ to $95 \%$ (29) and are used to treat benign prostatic hypertrophy and male pattern baldness. A meta-analysis of dutasteride and a large clinical trial of finasteride did not report associations with CV events, whereas a trial of dutasteride reported increased episodes of cardiac failure associated with dutasteride ( $30-32$ ). These studies are limited, however, because they were not designed to assess CV outcomes and relied on self-report for adverse CV outcomes.

We found no association between total T and cFT and incident CVD, which is in contrast to other studies (9-16). However, the results of previous studies appeared to be influenced by whether the cohort contained men with prevalent CVD. In cohorts that included prevalent CVD, $62 \%$ of the studies ( 8 of 13 ) reported an association between lower T and CVD ( $9-16$ ). In contrast, in cohorts with no prevalent CVD or unknown CVD, only $33 \%$ of the studies ( 4 of 12 ) found an association with T and CVD (15, 33-35) (see Supplemental Table 3). Therefore, the lack of an association between T and CVD in the current
study is consistent with most studies of men without CVD that found no association between T and CVD. In contrast, most studies that included men with CVD reported an association between T and CVD. This may be because men with prevalent CVD are more vulnerable to adverse effects of low T or are at greater risk for recurrent CVD or that prevalent CVD is a confounder in the association between T and CVD. Although several studies adjusted for baseline CVD, some did not ( $36-38$ ), whereas others adjusted only for coronary heart disease $(10,19)$ or cerebrovascular disease (9) but not for both.

We also found no association between low total T and all-cause mortality, which conflicts with several previous studies, including an earlier study of ours, which found that low total T increased the risk of all-cause mortality (39). The results of the current study may differ from the previous study because the previous study examined a younger population of middle-aged male veterans who had high medical morbidity.

Limitations of this study are that we had only a single T measurement, whereas current guidelines (40) recommend repeated T levels. However, a single T level may be adequate for epidemiologic studies (41). Furthermore, the

ICC estimates based on repeated hormone measures indicated good to excellent reproducibility. Another limitation is that time of day for blood collection was not standardized. The effects of varying blood collection times on study results is likely to be minimal, however, because the circadian fluctuation in T levels is blunted in older men and DHT levels do not exhibit significant circadian variation (42). Another limitation is that calculated free DHT values by the Mazer method have not yet been validated via equilibrium dialysis. Finally, we were unable to measure estradiol, another major active metabolite of T , because we had insufficient sera to do so.

Strengths of this study are that we examined a cohort of elderly men in the CHS who had well-characterized CV risk factors, baseline CVD, and adjudicated CV outcomes. There are few other cohort studies with such well-characterized CV outcomes, and several previous studies relied on self-reported CV outcomes, which may be inaccurate. An additional strength is that we used a tandem mass spec-trometry-based assay to measure total T and DHT levels, which is the gold standard method for assaying hormone levels. Finally, measured levels of T and DHT were comparable to mean hormone levels measured by mass spectrometry reported in another similarly aged population of community-dwelling men in their 70s (43).

## Summary and conclusions

This is one of the first studies to examine the association of DHT with incident CVD and all-cause mortality in elderly men without a previous history of CVD. We found that DHT and calculated free DHT had curvilinear associations with incident CVD and all-cause mortality. The curvilinear associations of DHT with adverse outcomes suggest that there may be an ideal physiological range for DHT. However, the associations found in this study do not establish a causal relationship between DHT and adverse outcomes because this cannot be ascertained from an observational study. Further studies are needed to confirm these results and to clarify the physiologic mechanisms underlying the association of DHT with CVD and allcause mortality.

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[^1]:    a Unless indicated otherwise, results are shown as mean (SD).
    ${ }^{\text {b }}$ Calculated using height measured in 1992 and 1993 and weight measured in 1994 and 1995.
    ${ }^{c}$ Measured in 1992 and 1993.

[^2]:    ${ }^{\text {a }}$ Model 1 is adjusted for age, race, clinic site, smoking status, and alcohol consumption. Model 2 is adjusted for model 1 covariates plus systolic BP, antihypertensive use, HDL cholesterol, BMI, and waist circumference. Model 3 is adjusted for model 2 covariates plus diabetes. Model 4 is adjusted for model 2 covariates plus SHBG.
    ${ }^{\mathrm{b}} \mathrm{SD}=181 \mathrm{ng} / \mathrm{dL}$ for total $\mathrm{T}, 2.3 \mathrm{ng} / \mathrm{dL}$ for free testosterone, $0.23 \mathrm{ng} / \mathrm{mL}$ for DHT, and $0.14 \mathrm{ng} / \mathrm{dL}$ for free DHT.
    ${ }^{c} P$ values test the statistical significance of the linear and quadratic terms for the hormone analyte.

