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Title: A population-level decline in serum testosterone levels in American men

Short title: Population-level declines in male serum T

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ABSTRACT

Context. Age-specific estimates of mean testosterone (T) concentrations appear to vary by year of observation and by birth cohort, and estimates of longitudinal declines in T typically outstrip cross-sectional decreases. These observations motivate a hypothesis of a population-level decrease in T over calendar time, independent of chronologic aging.

Objective. To establish the magnitude of population-level changes in serum T concentrations, and the degree to which they are explained by secular changes in relative weight and other factors.

Design. A prospective cohort study of health and endocrine functioning in randomly selected men of age 45-79 y. Three data collection waves: baseline (T1: 1987-89) and two follow-ups (T2: 1995-97, T3: 2002-04).

Setting. An observational study of randomly selected men residing in greater Boston, MA, USA.

Participants. Data obtained on 1374, 906 and 489 men at T1, T2, and T3, respectively, totaling 2769 observations taken on 1532 men.

Main outcome measures. Serum total testosterone and calculated bioavailable testosterone.

Results. We observe a substantial age-independent decline in T that does not appear to be attributable to observed changes in explanatory factors, including health and lifestyle characteristics such as smoking and obesity. The estimated population-level declines are greater in magnitude than the cross-sectional declines in T typically associated with age.

Conclusions. These results indicate that recent years have seen a substantial, and as yet unrecognized, age-independent population-level decrease in T in American men, potentially due to birth cohort differences or to health or environmental effects not captured in observed data.

INTRODUCTION

Considerable loss of serum testosterone (T) is thought to be a feature of male chronologic aging (1-9). Low serum T has been associated with numerous age-related adverse health conditions including abdominal obesity, diabetes and pre-diabetic states (such as insulin resistance, impaired glucose tolerance, and metabolic syndrome), dyslipidemia, low bone and muscle mass, impaired sexual function, depressed mood, frailty, and decreased quality of life (10-12). T decline across the life span therefore represents an issue of great concern for public health, but large studies of within-person decreases in T are rare.

A previous analysis of baseline (T1: 1987-89) and initial follow-up (T2: 1995-97) data from the Massachusetts Male Aging Study (MMAS) indicated that the mean longitudinal (within-subject) decline in serum total testosterone (TT) per year of aging was more than twice the baseline cross-sectional decrease in mean TT per year of age (13). Qualitative comparisons of other existing studies likewise indicates that longitudinal decline within subjects is generally of greater magnitude than corresponding cross-sectional trends. We have hypothesized (13) that this disparity may be due to rapid intra-subject declines in health among subjects enrolled in longitudinal studies. A competing hypothesis, however, asserts that a population-level decline in T concentrations confounds cross-sectional and longitudinal estimates of T decline with age. A population-level decrease in serum T levels could accelerate the longitudinal declines in T concentrations typically associated with subjects' aging and compress cross-sectional decreases associated with age. Completion of the latest follow-up wave of MMAS data collection (T3: 2002-04) allows us, for the first time, to formally investigate the possibility of an age-independent decline in serum T levels with calendar time.

To our knowledge, there exist no extensive published studies of changes in the age-matched distribution of T over time, but a population-level decline in serum T concentrations would be consistent with evidence of secular decreases in male fertility and sperm count (14, 15). In this analysis, we estimated differences in serum total testosterone and calculated bioavailable testosterone (BT) concentrations obtained from individuals of like age observed at different times (e.g. comparing TT in men who were 65 years old in 1988 to those in comparable men who were 65 years old in 2003). Our working hypothesis was that age-independent differences would be attributable to population-level changes in health and lifestyle observable during the nearly 20 years of study follow-up.

METHODS

The MMAS is a prospective cohort study of men's health and endocrine function. Its design and prior results are described elsewhere (1, 5, 13, 16). Briefly: from a randomly-chosen sample of 1709 men living in and around Boston, blood samples and interview data were obtained during in-home visits by trained staff, with data collection comprising a baseline (T1) and two follow-up (T2, T3) waves. All study activities, including informed consent protocol, were approved by the Institutional Review Board of the New England Research Institutes (NERI).

T concentrations are subject to systematic variation due to components of study design (17-19). Accordingly, the MMAS took steps to minimize design bias. To counteract the effects of episodic secretion of hormones, two samples were obtained at each visit and pooled in equal aliquots at the time of assay. To control the effects of diurnal variation in hormone concentrations (20), samples were obtained within 4 hours of subjects' waking. Blood was kept in an ice-cooled container for transport and centrifuged within 6 hours. Serum was stored in 5 mL scintillation vials at -20°C, shipped to the laboratory within one week by same-day courier, and stored at -70°C until the time of assay. All

hormone values were obtained by a single technician at the Endocrine Laboratory, University of Massachusetts Medical Center, under the direction of Christopher Longcope, MD. TT concentrations were obtained by radioimmunoassay (Diagnostic Products Corporation, Los Angeles, CA). T1 assays were performed in 1994, while T2 and T3 samples were assayed shortly after in-home visits. TT inter-assay coefficients of variation were 8.0, 9.0, 8.3 at T1, T2, T3, respectively. TT concentrations obtained in the MMAS fall near the center of the distribution of concentrations obtained in other major epidemiologic studies (16), and quality control testing indicated negligible change in concentrations between T1 and T2 due either to sample storage or assay drift (5).

Serum sex hormone-binding globulin (SHBG) was measured using RIA kits at T1 and T2, and at T3 by chemiluminescent enzyme immunometric assay using the DPC Immulite technology. SHBG inter-assay CVs were 10.9%, 7.9%, and 3.0% at T1, T2, T3, respectively. BT was calculated using the mass action equations described by Södergard et al (21), with association constants taken from Vermeulen et al. (22)

Covariate Data. Demographic characteristics (age, education, income, marital status), health conditions (cancers, diabetes, heart disease, hypertension, and ulcer), self-assessed general health (a five-point ordinal scale), and smoking and daily alcohol consumption (23) were obtained via self-report. Self-reported diagnoses of prostate cancer were supplemented with examination of available medical records. Height, weight and waist and hip circumference were obtained using methods developed for large-scale epidemiologic field work (24). Body mass index (BMI) and waist-to-hip ratio were derived by calculation. A comprehensive inventory of all prescription medications used by subjects was obtained. Daily caloric intake was measured using the Willett 1-year food frequency questionnaire (25). Physical activity and energy expenditure were derived from subjects' seven-day

recall of duration and frequency of their activities (26). Depressive symptoms were measured using the Centers for Epidemiological Studies – Depression (CES-D) scale (27).

Analysis Sample. In order to enhance comparability of age distributions across study waves and to allow for analyses of T concentrations by subjects' birth cohorts, data were restricted to observations obtained on men of age 45 to 79 years born between 1916 and 1945, inclusive. This yielded potential samples of 1399, 975, and 579 observations at T1, T2, and T3, respectively. Of these, we excluded all observations on the seven men who had T1 serum total T < 100 ng/dL (3.5 nmol/L), and two outlying observations with total T > 1200 ng/dL (41.6 nmol/L). One hundred twenty-six observations were excluded because they were taken on subjects who, prior to the relevant study wave, had a diagnosis of prostate cancer, for which treatment via hormone suppression therapy could not be ruled out. An additional 44 observations were excluded because apparent health status could not be determined. This yielded samples of 1374, 906 and 489 observations at T1, T2, and T3, respectively, totaling 2769 observations taken on 1532 men.

Statistical Analysis. Exploratory analyses were conducted to assess the functional form of associations. We used mixed-effects linear regression (28) with random subject-level intercepts and slopes to estimate trends and test hypotheses. Hormone concentrations were log (base e) transformed to remove any effects of the mild skew in the data. For a covariate with associated regression estimate β^*, we approximated the corresponding percent change in mean hormone concentrations using the quantity $100 \times (e^{\beta^*}-1)$. Results were considered statistically significant if null hypotheses could be rejected at the 0.05 level. The significance of effects was evaluated using Wald and likelihood ratio (LR) tests. Confounders were employed in multivariate models if they had considerable theoretical importance or were significantly associated with T concentrations in the presence of other predictors.

All confounders were allowed to vary with time and were treated as internal time-dependent covariates (29).

RESULTS

A description of the analysis sample is given in Table 1. Median baseline age was 58 years, with interquartile range (IQR) 52 to 64 years. Seven hundred nineteen (52%) subjects reported at least one chronic illness, 340 (25%) were current smokers, 296 (22%) were obese (BMI ≥ 30), and 252 (18%) reported use of at least three prescription medications. Over the course of study follow-up, we observed marked increases in the proportion of subjects reporting at least one chronic illness or who were overweight or obese, as well as in the number of medications being used by subjects; there were dramatic decreases in the proportion of subjects who were current smokers or who were employed.

Table 2 presents descriptive statistics for age and T concentrations at all study waves. Median TT at baseline was 501 ng/dL (17.4 nmol/L), with IQR 392-614 ng/dL (13.6 – 21.3 nmol/L); the corresponding values at T3 were 391 ng/dL (13.6 nmol/L) and 310-507 ng/dL (10.7 – 17.6 nmol/L). Among subjects on whom follow-up data could be obtained, the median lag time between observations at T1 and T2 was 8.8 years, and between T2 and T3 was 6.4 years.

Exploratory Analyses. We used graphical displays to assess three interrelated quantities: first, the cross-sectional association between T concentrations and age at any study wave; second, the longitudinal decline of T over time associated with subjects' aging; third, the *age-matched* difference between, for instance, mean T concentrations obtained from 65 year-old men in 1988 and concentrations obtained from 65 years old men in 2003 (equivalently, we sought to compare T

concentrations obtained in 1988 from men born circa 1923 to concentrations obtained in 2003 from men born circa 1938). A depiction of mean TT concentrations is given in Figure 1, which displays nonparametric locally weighted estimates of TT by age separately for each study wave. The negative slopes of the wave-specific fits correspond to the relatively modest cross-sectional decline of mean TT with age. The age-matched difference by time (denoted by the vertical distance between the fitted curves in overlapping age ranges) is likewise evident. The data suggest that the cross-sectional decline of TT within T1 is smaller than the age-matched difference between concentrations taken at T2 versus T1, which are separated by only about nine years in time; simple linear regression estimates indicate cross-sectional TT decreases of 17 and 20 ng/dL (0.6 and 0.7 nmol/L) per 10 years of age at T1 and T2, respectively, whereas the mean difference between subjects age 65 at T1 versus subjects age 65 at T2 is roughly 50 ng/dL (1.7 nmol/L).

To more carefully explore trends associated with age and time, it is useful to depict subjects by birth cohort. Figure 2 displays all (log-transformed) TT concentrations in the analysis sample versus age, and includes mixed-effects regression (28) estimates of the average within-subject TT decline by 5-year birth cohort. A display fitting nonparamentric locally weighted regression smooths (not shown) was similar. We refer to five-year birth cohorts as Cohort I (men born in the years 1916-19), Cohort II (1920-24), … , Cohort VI (1940-45). Although the design of the MMAS precludes all cohorts from being observed over all ages, the pattern of decreasing TT concentrations across adjacent cohorts is compelling. That the regression lines are approximately parallel indicates that the age-matched decline over time (again indicated by vertical distances between pairs of fitted lines) is consistent across age groups.

Detailed exploratory analyses provide additional evidence of an age-matched time trend. Table 3 provides an illustrative example. Here we restrict our attention to Cohorts II and IV and their

147 associated TT concentrations at T1 and T2. Calculation indicates that among subjects in Cohort IV
148 (born 1930-34), the proportionate decline in mean TT from T1 to T2 was 16.1% (the median age at T1
149 was 56 years and at T2 was 64 years). By contrast, a cross-sectional comparison at baseline indicates
150 that Cohort II (median age 65 years) T levels are only 5.5% lower than those of Cohort IV (median age
151 56 years). The age-matched time difference (comparing observations on men of similar age separated
152 by time: Cohort IV at T2 versus Cohort II at T1) is roughly 11.2%, approximately the difference
153 between the cross-sectional and longitudinal trends. Similar effects may be observed in other
154 combinations of birth cohorts and study waves.
155
156 **Formal Results: Total Testosterone.** An analysis of all data yields results in agreement with our
157 exploratory observations. In order to estimate cross-sectional and longitudinal trends, we partition
158 subjects' ages into two pieces: age at baseline and subsequent aging, the latter defined as calendar
159 time since study entry. The per-year age-matched time trend was estimated as the difference between
160 the associated longitudinal and cross-sectional regression estimates (30-32). Mean cross-sectional,
161 longitudinal and age-matched trends derived from mixed-effects models of TT as a function of age and
162 aging are depicted on the left side of Table 4. The estimated cross-sectional decline in TT is -0.4% per
163 year of age, with a corresponding 95% confidence interval (CI) of (-0.6%, -0.2%). The longitudinal
164 within-subject decline is approximately -1.6% per year (CI: -1.8%, -1.4%). The age-matched time
165 trend is -1.2% per year (CI: -1.4%, -1.0%).
166
167 We hypothesized that the presence of the age-matched time trend could be accounted for by observable
168 secular changes in health status or lifestyle characteristics. This hypothesis relies upon an assertion
169 that for men of, say, 65 years of age, health/lifestyle characteristics vary by observation time. For
170 instance, the well-known and ongoing secular increase in obesity might explain the fact that the typical
171 blood sample taken from a 65-year-old man in 2003 exhibited lower TT concentrations than a sample

taken from a different 65-year-old subject in 1988 (the latter subject having been born approximately 15 years earlier than the former). In this analysis we observed little evidence of age-independent trends with respect to most covariate factors; notable exceptions to this rule, however, were the aforementioned increases in relative weight, as well as population-level changes in the prevalence of smoking and the concurrent use of multiple medications (polypharmacy). There were substantial age-specific increases in obesity and polypharmacy over the course of study follow-up, whereas the proportion of subjects who smoked cigarettes declined dramatically in all age groups. These trends are potentially important in accounting for an apparent secular decline in TT levels, as weight gain, smoking cessation, and the use of medications have been associated with decreases in serum T (33-37). However, while controlling for these and other factors significantly associated with TT concentrations was sufficient to substantially decrease the estimates of cross-sectional and longitudinal decline in TT, the estimate of the age-matched time trend was only slightly reduced (see Table 4). Results were essentially unchanged when all covariate effects (see Methods) were included in multivariate analyses.

Bioavailable Testosterone. As noted above, the technology by which SHBG was measured at T1 and T2 (RIA) differed from that employed at T3 (Immulite). Because of this, observed variation in calculated BT concentrations between T2 and T3 could be artificially inflated. We therefore restricted formal estimation of cross-sectional, longitudinal and age-matched time trends in BT to values obtained at T1 and T2.

In the resulting models, as is consistent with other published results, cross-sectional and longitudinal age trends in BT were substantially sharper than those in TT. However, the age-matched time trend was similar in magnitude to that in TT, and was likewise robust to control for all covariates. When only the effects of age and aging were controlled, the estimated age-matched time trend in BT values was approximately -1.4% per calendar year (95% CI: -1.8%, -1.1%), whereas when the effects of all

other covariates were accounted for, the estimated age-matched trend was -1.3% per year (95% CI: -1.7%, -1.1%).

Sensitivity Analyses. In order to test the robustness of all findings, we performed a number of additional analyses. Analyses including effects for town of residence, assay batch, month of interview, and time of study visit yielded results nearly identical to those described above. Results did not change substantially when analyses of TT were restricted to data from any two of the three study waves. Likewise, results were similar when analyses of either TT or BT were restricted to men above or below certain ages, to men with complete data at all three waves, or to men in particular birth cohorts. In addition, we examined the distribution of baseline TT and BT concentrations among those subjects who had complete data versus those who did not, and found that they were comparable.

DISCUSSION

These findings indicate that the past twenty years have seen substantial age-independent decreases in male serum T concentrations, decreases that do not appear to be the consequence of the contemporaneous trends in health and lifestyle considered here. It remains unclear to what these apparent population-level decreases in T are attributable.

Because age, birth year and observation time are perfectly confounded (that is, knowledge of any two determines the third), their influences are not separable through data analysis. Age-matched time differences cannot, therefore, be definitively attributed to historical (pre-study) trends affecting different birth cohorts in different ways or, rather, to contemporary secular changes in the underlying population (e.g., to age-independent increases in obesity beyond those captured in the analyses described here). As noted previously, there is little evidence that the association between T and age

(that is, the slope of a line depicting the relationship between the two) depends upon birth year, so that irrespective of birth cohort, decreases in T with age are constant (see Figure 2). This evidence is consistent with - but does not prove - the notion that the linear T/age association is consistent across different generations, and implies that the age-matched declines in T levels associated with each year of calendar time apply equally to men from 45 to 80 years of age.

The presence of the age-matched trend itself, however, suggests that neither cross-sectional nor longitudinal investigations may properly describe the true effect of aging *per se* on T (30-32). Suppose, for instance, there were an unmeasured but persistent environmental exposure associated with decreased T levels, affecting recent generations of men at birth. In that case the cross-sectional decline in T with age might be underestimated, as younger men could have lower T levels than their historic counterparts and appear more like their older contemporaries (born prior to the advent of the exposure) than one would normally expect in the absence of such a hypothetical exposure.

On the other hand, if the age-matched trend is not historic but rather indicative of population-level changes occurring during the time subjects were under study, the age-matched trend denotes a secular trend in T concentrations over that time. Under this scenario it is easy to see that longitudinal estimates of change in T concentrations may in fact overstate the true effect of aging, because the observed effect of a year of aging would include not only the true age-related decreases in T but also whatever decreases the population-level secular trend imposed on all men simultaneously. Such a secular trend in T might be attributable to parallel population-level changes in the distribution of health and lifestyle factors, independent of age. We have observed, however, that while baseline and evolving health states in the study sample successfully account for a substantial proportion of the cross-sectional and longitudinal associations between age and T, they do not explain the age-matched decline in T concentrations.

We therefore hypothesize that the observed age-matched decline in serum testosterone is due to some undocumented historical or contemporary influence, health-related or environmental, which manifests in observable age-matched differences in T concentrations separated either by time of observation or by birth cohort.

It is interesting to note that the estimated age-matched time trends in TT and BT are of comparable magnitude. This may not in itself be surprising, as the time trends are explicitly intended to remove the effects of aging itself, leaving only secular changes in other factors as contributors to changes in T levels with time. We can currently offer, however, no additional speculation as to whether one would expect a secular trend in BT to differ markedly from that in TT.

Some limitations of this study should be acknowledged. Though the consistency of the methods by which TT concentrations were obtained - as well as that of the age-matched time trend across all pairs of study waves - indicates that design artifacts are likely not the cause of these observations, they cannot be completely discounted as contributors to the age-matched time trends, as relatively subtle changes in measurement may contribute substantially to differences between observations separated by time. Likewise, though the evidence suggests that subject loss to follow-up has not influenced our result, we must acknowledge the possibility of biases arising from subject dropout. However, under the assumption of such a survival bias, those subjects who remain in the study, being younger (and presumably more healthy) than those lost to follow-up, would be likely to exhibit higher mean T concentrations during follow-up than would the complete sample had it been fully observed. Under such a scenario, it is likely that the estimates of longitudinal and age-matched decline described here would be too low, rather than too high.

An added concern is that the covariates considered in this analysis cannot account for all known causes of T decline. Indeed it is exceedingly unlikely that population-level T concentrations would decline with calendar time - independently of age - of their own accord. Rather, if such declines exist, they have one or several causes which themselves may be evolving over time. We have observed that several candidate causes observable at the level of the individual subject, most notably the well-known secular declines in smoking rates and increases in relative weight, do not appear to completely explain the observed age-matched trends in T. It remains possible, however, that more detailed and comprehensive measurement of such factors could fully account for the age-matched trends in T.

If the trends observed in the MMAS are real and continue, the prevalence of low T in American men will exhibit increases in excess of those to be expected given the projected aging of the population (38). As such, it is important that future research endeavors to confirm or disprove the existence of age-independent T declines, and to discover their causes, environmental or otherwise, so that they may be addressed through prevention.

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FIGURE LEGENDS

Figure 1. Crude mean total testosterone concentrations, by MMAS study wave (T1, T2, T3), with confidence bands (dotted lines). Estimates are obtained from a generalized additive model with a lowess smoothing term.

Figure 2. MMAS mean total testosterone (TT) versus age, by five-year birth cohort. Fitted lines are obtained from cohort-specific mixed-effects regression of the log of TT on centered age, with random effects for each subject. Data points in the analytic sample are also depicted; each subject contributes up to three observations. Models are fit using maximum likelihood.

Table 1. Descriptive statistics by MMAS study wave, Mean (Standard Deviation) or Count (%).

	T1 (1987-1989) N = 1374	T2 (1995-1997) N = 906	T3 (2002-2004) N = 489
Age (y)	57.7 (7.2)	63.2 (7.8)	67.3 (6.5)
Chronic Illness			
Non-Prostate Cancers	89 (6%)	124 (14%)	85 (17%)
Diabetes	120 (9%)	80 (9%)	62 (13%)
Heart Disease	196 (14%)	155 (17%)	114 (23%)
Hypertension	449 (33%)	340 (38%)	248 (51%)
Ulcer	146 (11%)	117 (13%)	64 (13%)
Any	719 (52%)	545 (60%)	349 (71%)
Depressive Symptoms (CES-D ≥ 16)[a]	149 (11%)	96 (11%)	43 (9%)
Self-assessed General Health			
Excellent	417 (30%)	280 (31%)	127 (26%)
Very good	475 (35%)	336 (37%)	190 (39%)
Good	360 (26%)	219 (24%)	110 (27%)
Fair / Poor	120 (9%)	71 (8%)	42 (9%)
Prescription Medications			
0	517 (38%)	196 (22%)	0 (0%)
1-2	557 (41%)	351 (39%)	170 (37%)
3-5	252 (18%)	270 (30%)	178 (38%)
6+	48 (3%)	89 (10%)	116 (25%)
Education			
< High school	173 (13%)	83 (9%)	34 (7%)
High School Graduate	263 (19%)	137 (15%)	81 (17%)
> High School	938 (68%)	680 (76%)	374 (76%)
Marital Status			
Single / Never Married	108 (8%)	63 (7%)	40 (8%)
Married	1044 (76%)	701 (77%)	367 (75%)
Divorced/Separated	171 (12%)	97 (11%)	55 (11%)
Widowed	51 (4%)	45 (5%)	27 (5%)
Household Income			
< $40,000 / y	546 (41%)	271 (31%)	122 (26%)
$40,000 – 79,000 / y	530 (40%)	299 (34%)	153 (32%)
> $80,000 / y	250 (19%)	302 (35%)	199 (42%)

Currently Employed	1032 (75%)	565 (62%)	257 (53%)
Weight and Body Shape			
Body mass index (kg/m^2)	27.4 (4.4)	27.6 (4.4)	28.3 (4.8)
Waist-to-hip ratio	.95 (.06)	.96 (.06)	.97 (.06)
Cigarette smoking	340 (25%)	118 (13%)	45 (9%)
Dietary Intake			
Total kcal / day	2069 (817)	2006 (720)	1911 (743)
Animal fat (g /day)	40.3 (22)	36.6 (19)	38.0 (20)
Sedentary Activity Levels	488 (36%)	285 (31%)	139 (28%)

Table 2: Total and calculated bioavailable testosterone concentrations, by study wave and corresponding age range.

Study Wave	Observation Years	Age Range (y)	N	Total Testosterone (ng/dL[a])		Bioavailable Testosterone (ng/dL[a])	
				Median	Interquartile Range	Median	Interquartile Range
T1	1987-89	45 – 71	1383	501	392 – 614	237	179 – 294
T2	1995-97	50 – 80	955	435	350 – 537	188	150 – 234
T3	2002-04	57 – 80	568	391	310 – 507	130	101 – 163

[a] May be converted to nmol/L via multiplication by 0.03467.

Table 3: Age-matched trends: illustrative example.

Crude cross-sectional, longitudinal and age-matched trends in mean total testosterone (TT) per year age or time, restricted to men born 1920-24 (Cohort II) or 1930-34 (Cohort IV). Men in Cohort II have comparable age when observed at T1 (upper left) to that of men in Cohort IV when observed at T2 (lower right); the disparity between these measurements approximates the unadjusted age-matched time trend in TT. Median time between observation at T1 and T2 is roughly 8.8 years.

		T1: 1987-89		**T2: 1995-97**		
Cohort	**Birth Years**	**Median Age (y)**	**Mean (SD) Total Testosterone (ng/dL[d])**	**Median Age (y)**	**Mean (SD) Total Testosterone (ng/dL[d])**	
II	**1920 – 24**	65	500 (161)			**Longitudinal Difference[b]: -16.0 %**
IV	**1930 – 34**	56	529 (183)	64	444 (145)	
	Cross-Sectional Difference[a]:		**-5.5%**			
					Age-Matched Time Difference[c]: -11.2%	

[a] T1: Cohort II versus Cohort IV, estimates mean cross-sectional decrease per nine years age.

[b] Cohort IV: T2 versus T1; estimates mean longitudinal decline per nine years aging.

[c] Cohort IV, T2, versus Cohort II, T1; estimates mean age-matched decline per nine years time.

[d] May be converted to nmol/L via multiplication by 0.03467.

Table 4: Longitudinal regression results. Though apparent cross-sectional and longitudinal associations with age are reduced by statistical control for health and lifestyle, the age-matched time trend remains large.

	Unadjusted Results			Adjusted Results[a]		
	Mean Decline (% / y)	**95% CI**	**p-value[b]**	**Mean Decline (% / y)**	**95% CI**	**p-value[b]**
Cross-sectional trend (per y age)	- 0.4	(- 0.6, - 0.2)	< .001	- 0.1	(- 0.3, 0.1)	0.42
Longitudinal trend (per y aging)	- 1.6	(- 1.8, - 1.4)	< .001	- 1.1	(- 1.3, - 0.9)	< .001
Age-matched time trend (per y time)	- 1.2	(- 1.5, - 1.0)	< .001	- 1.0	(- 1.3, - 0.8)	< .001

[a] Adjusted for chronic illness, general health, medications, smoking, BMI, employment, marital status.

[b] Wald test of regression effect.

Figure 1

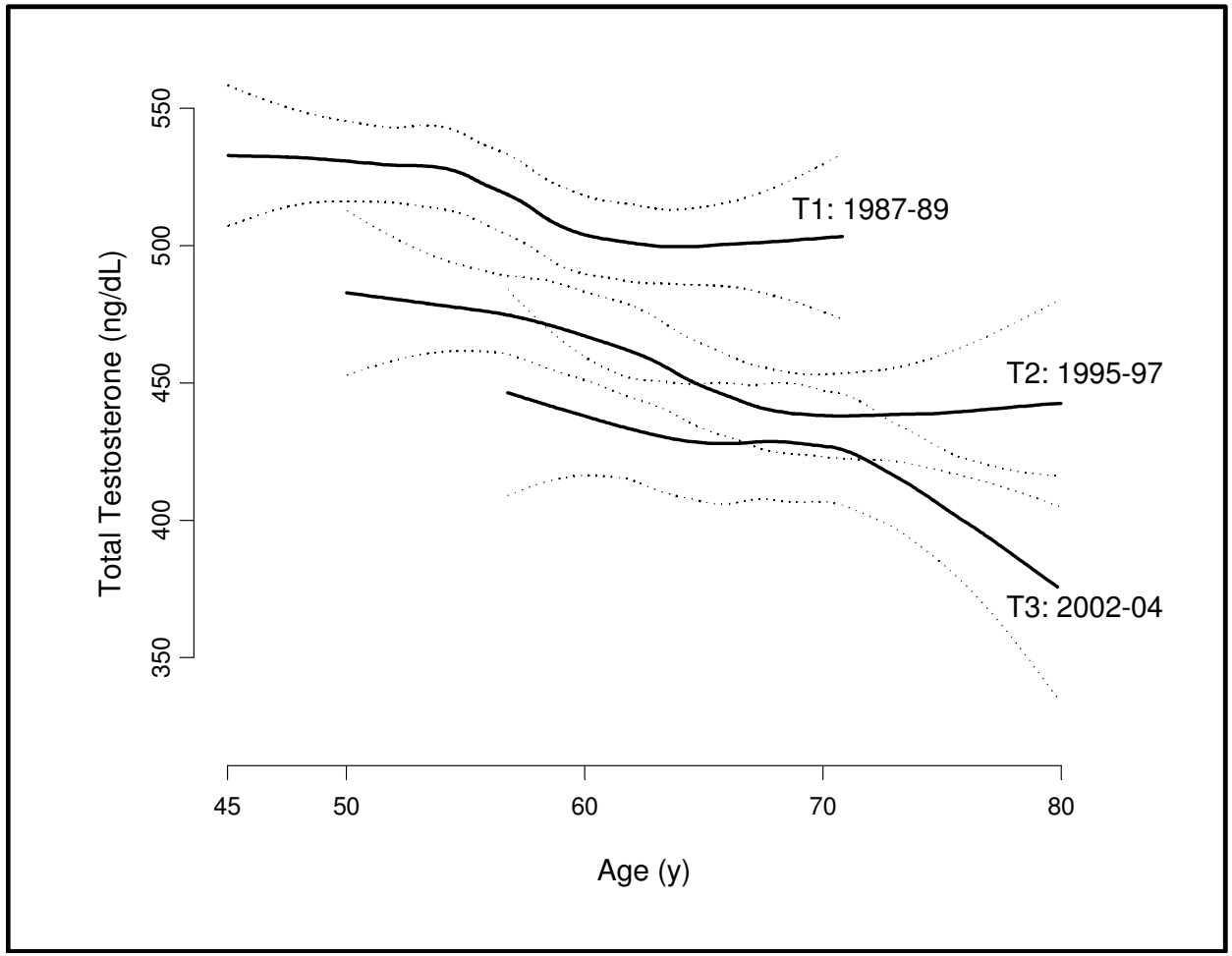
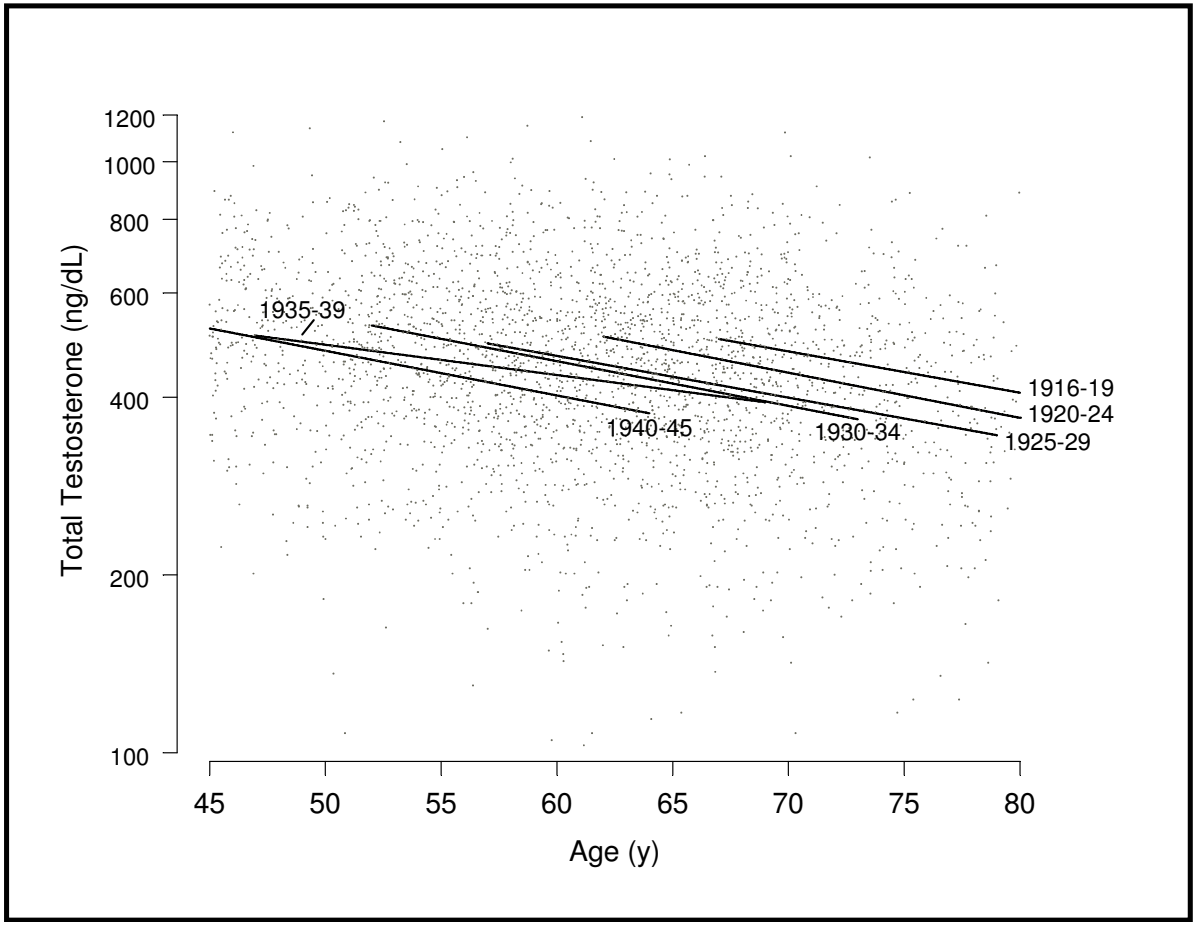

Total Testosterone (ng/dL)
550
500
450
400
350
T1: 1987-89
T2: 1995-97
T3: 2002-04
45
50
60
70
80
Age (y)

Figure 2


Total Testosterone (ng/dL)
1200
1000
800
600
400
200
100
45
50
55
60
65
70
75
80
Age (y)
1935-39
1940-45
1930-34
1925-29
1920-24
1916-19