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Association of Testosterone and Estradiol Deficiency with Osteoporosis and Rapid Bone Loss in Older Men

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Context: The clinical value of measuring testosterone and estradiol in older men with osteoporosis and of measuring bone mineral density (BMD) in older men with testosterone or estradiol deficiency is uncertain.

Objective: The objective of the study was to examine the association of testosterone and estradiol deficiency with osteoporosis and rapid bone loss in older men.

Design: This study was a cross-sectional and longitudinal analysis.

Setting: The study was conducted at six U.S. centers of the Osteoporotic Fractures in Men study.

Participants: The study population consisted of 2447 community-dwelling men aged 65 yr or older.

Main Outcome Measures: Total testosterone deficiency was defined as less than 200 ng/dl. Total estradiol deficiency was defined as less than 10 pg/ml. Osteoporosis was defined as femoral neck or total hip BMD T-score of −2.5 or less. Rapid bone loss was defined as 3%/yr or more.

Results: Prevalence of osteoporosis in men with deficient and normal total testosterone was 12.3 and 6.0% ($P = 0.003$) and 15.4 and 2.8% ($P < 0.0001$) in those with deficient and normal total estradiol. Among osteoporotic men and those with normal BMD, prevalence of total testosterone deficiency was 6.9 and 3.2% ($P = 0.01$), and prevalence of total estradiol deficiency was 9.2 and 2.4% ($P = 0.0001$). Incidence of rapid hip bone loss in men with deficient and normal total testosterone was 22.5 and 8.6% ($P = 0.007$) and in those with deficient and normal total estradiol was 14.3 and 6.3% ($P = 0.08$).

Conclusions: Older men with total testosterone or estradiol deficiency were more likely to be osteoporotic. Those with osteoporosis were more likely to be total testosterone or estradiol deficient. Rapid hip bone loss was more likely in men with total testosterone deficiency. BMD testing of older men with sex steroid deficiency may be clinically warranted. **(*J Clin Endocrinol Metab* 91: 3908–3915, 2006)**

INCREASINGLY, OSTEOPOROSIS IS recognized as an important disease in elderly men. With aging, older men experience bone loss and are at increased risk for osteoporosis-related fractures. Among factors reported to be associated with bone health in older men, a growing amount of literature has examined the role of sex steroids.

In older men, cross-sectional studies generally have found that estradiol correlates positively and more strongly with bone mineral density (BMD) than does testosterone (1–6), and that the respective bioavailable fractions are more strongly correlated with BMD than are total estradiol and testosterone (1, 4–7). A more sparse amount of literature of longitudinal studies has suggested that estradiol levels inversely correlate with bone loss but that an association between testosterone and bone loss is weaker or absent (3, 5, 8).

Although provocative, these studies provide little guidance regarding the clinical utility of measuring sex steroid levels in older men when BMD is known or measuring BMD in older men when sex steroid levels are known. Ultimately, the value of such additional testing may depend on not just the likelihood of identifying an abnormality but whether the new information may alter patient management.

There also is uncertainty regarding how to define osteoporosis (9, 10) and low sex steroid levels (11, 12) in older men. Because BMD is a continuous risk factor for fractures and sex steroids have a complex relationship with bone and other health outcomes, any categorical definition of abnormal is ultimately arbitrary. Nevertheless, cut points may assist clinical decision making if meaningfully related to patient prognosis or treatment (13).

In this context, this study examines, in a large geographically diverse cohort of older men, how applying a conventional definition of osteoporosis as a clinical screening threshold for measuring testosterone and/or estradiol levels identifies older men meeting *a priori* biochemical definitions for testosterone and/or estradiol deficiency. Analogously, this study examines how applying these definitions for testosterone and/or estradiol deficiency as screening thresholds

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Abbreviations: BMD, Bone mineral density; CI, confidence interval; CV, coefficient of variation; DXA, dual-energy x-ray absorptiometry; MrOS, Osteoporotic Fractures in Men Study; NHANES, National Health and Nutrition Examination Survey; OR, odds ratio.

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for BMD testing identifies older men with osteoporosis and/or subsequent rapid bone loss.

Subjects and Methods

Participants

Community-dwelling men aged 65 yr or older were recruited for the prospective Osteoporotic Fractures in Men Study (MrOS) at six U.S. sites: Birmingham, Alabama; Minneapolis, Minnesota; Palo Alto, California; Pittsburgh, Pennsylvania; Portland, Oregon; and San Diego, California. MrOS exclusion criteria included an inability to walk without assistance and a history of bilateral hip replacement. The MrOS study design and recruitment have been described in detail elsewhere (14, 15).

Of 5995 MrOS participants attending the baseline examination (March 2000 to April 2002), 5994 (>99.9%) completed technically adequate measurements of hip BMD by dual-energy x-ray absorptiometry (DXA). From September 2002 through May 2003, men enrolled at the Birmingham and Portland centers were invited to participate in a MrOS ancillary study to identify determinants of periodontal disease in older men. Endogenous sex steroid levels collected at baseline were assayed in a stratified sample of men, with strata consisting of race (white, nonwhite), completion of baseline hip and spine quantitative computed tomography (yes, no), and participation in the dental ancillary study (yes, no). Within each stratum, participants were sampled with known probability. All nonwhite participants were sampled, whereas those with complete quantitative computed tomography skeletal measurements or from the Birmingham and Portland sites were oversampled. The sample target was 2643 participants and a sample of 2623 (99%) was achieved. After exclusion of 173 men receiving testosterone supplementation or androgen suppression therapy at MrOS baseline and three men with incomplete sex steroid data, there remained 2447 participants with both baseline DXA hip BMD and complete sex steroid measurements. These men formed the cohort for the cross-sectional analyses.

Of the 1360 men from Birmingham and Portland included in the cross-sectional analyses, 1236 (90.9%) attended the dental (second) examination, with 1235 (>99.9%) completing technically adequate DXA measurements of hip BMD. Change in hip BMD could not be calculated for eight men because the same hip was not scanned at both visits. There remained 1227 men who formed the cohort for the longitudinal analyses. The mean interval between examinations was 1.8 ± 0.4 yr (SD).

Written informed consent was obtained from all participants, and institutional review boards at all participating centers approved the study protocol.

Measurement of BMD

At the baseline and second examinations, participants underwent BMD (grams per square centimeter) measurement of total hip, femoral neck, and lumbar spine with DXA (QDR 4500W; Hologic, Inc., Waltham, MA). All hip BMD measurements were made on the right hip unless the participant reported a right hip replacement or metal objects in the right leg, in which case the left hip was measured. Lumbar spine BMD was measured in the anterior-posterior projection and calculated as the mean of the BMD from the first through fourth lumbar vertebrae. A standard phantom was used at all study clinics for cross-calibration. Interclinic coefficients of variation (CVs) were 0.9 (hip) and 0.6% (spine).

Measurement of sex steroids

At baseline, paired serum specimens were collected after an overnight fast and stored at −70 C until assayed. Assays were performed at the Oregon Health Sciences University General Clinical Research Center by a single technician. All testosterone and estradiol assays were completed in duplicate, and the average value was used in the analyses. Criteria were implemented to repeat the assay when replicates were highly discrepant within an assay; results from any repeat analyses were averaged with the original sample data. Pooled serum controls were used in every assay run.

Serum total testosterone was measured using a solid-phase ^{125}I RIA (Diagnostic Products Corp., Los Angeles, CA; detectable range 10–1600 ng/dl; intraassay CV 5.4%; interassay CV 8.2%). Serum total estradiol was measured with an ultrasensitive RIA (Diagnostic Systems Laboratories, Webster, TX; detectable range 2.5–750 pg/ml; intraassay CV 8.5%; interassay CV 13.3%). SHBG was measured using an immunometric assay (Diagnostic Products; detectable range 0.2–180 nM; intraassay CV 3.3%; total assay CV 5.3%). Bioavailable testosterone and estradiol were calculated using mass action equations (16, 17).

Other measurements

At study baseline, participants completed a self-administered questionnaire and were interviewed and examined by trained and certified clinical staff in the clinical center. Characteristics assessed included age, race, and weight.

Statistical analyses

Based on recommendations for a biochemical threshold to classify testosterone deficiency in older men ranging from 200 to 300 (18–22), participants were categorized as total testosterone deficient [<200 ng/dl (<6.9 nmol/liter)], possibly testosterone deficient [200–400 ng/dl (6.9–13.9 nmol/liter)], or normal [>400 ng/dl (> 13.9 nmol/liter), with secondary analyses categorizing participants as less than 200 ng/dl, 200 to less than 300 ng/dl, 300 to less than 400 ng/dl, 400 to less than 500 ng/dl, and 500 ng/dl or more. Considering published data suggesting a possible threshold for total estradiol deficiency in older men of 20–30 pg/ml (23) and the total estradiol distribution in MrOS, participants were categorized as total estradiol deficient [<10 pg/ml (< 36.7 pmol/liter)], possibly estradiol deficient [10–20 pg/ml (36.7–73.4 pmol/liter)], or normal [>20 pg/ml (> 73.4 pmol/liter)], with secondary analyses categorizing participants as less than 10 pg/ml, 10 to less than 15 pg/ml, 15 to less than 20 pg/ml, 20 to less than 25 pg/ml, and 25 pg/ml or more. Bioavailable testosterone and estradiol levels were categorized into quintiles, and then the cut point defining the lowest fifth percentile for each of these bioavailable sex steroid levels was identified.

With respect to skeletal status, participants were categorized according to their baseline femoral neck and total hip BMD T-scores using a young Caucasian male reference database from National Health and Nutrition Examination Survey (NHANES) III (24) as osteoporotic ($T \leq -2.5$ at either hip site), osteopenic (low bone mass) ($T > -2.5$ at both sites and ≤ -1 at either site), or normal ($T > -1$ at both sites). In secondary analyses, participants with T −2 or less at either hip site were defined as having low BMD, and alternatively participants were categorized according to their spine BMD T-scores, using the manufacturer's reference database, as osteoporotic ($T \leq -2.5$), low bone mass ($T > -2.5$ and ≤ -1), or normal ($T > -1$). Rapid bone loss between the baseline and second exam was defined as 3% or greater annualized loss in BMD at either hip site because this rate has been reported to predict increased risk of incident fractures independent of BMD (25). In secondary analyses, rapid bone loss was defined as 3% or greater annualized loss in spine BMD.

χ_2 tests were used to compare the prevalence of osteoporosis (or secondarily of BMD T-score ≤ -2) in participants: 1) with deficient or possibly deficient total testosterone or estradiol levels *vs.* those with normal levels of the respective sex steroid; 2) in each total testosterone category *vs.* a reference group with total testosterone 500 ng/dl or greater; 3) in each total estradiol category *vs.* a reference group with total estradiol 25 pg/ml or greater; and 4) in each quintile and then in the lowest fifth percentile of bioavailable testosterone or estradiol *vs.* reference groups in the highest quintile for the respective bioavailable sex steroid. χ_2 tests also were used to compare the prevalence of total testosterone or estradiol deficiency in participants with osteoporosis or low bone mass *vs.* a reference group with normal BMD.

Age- and weight-adjusted logistic regression analyses were used to examine the likelihood of osteoporosis or low BMD at baseline and of subsequent rapid bone loss associated with each of the above categories of sex steroid levels and the likelihood of total testosterone or estradiol deficiency associated with osteoporosis. Results were expressed as odds ratios (ORs) with 95% confidence intervals (CIs). Bone loss analyses were also adjusted for baseline femoral neck BMD, and all models examining testosterone as a predictor variable were further adjusted for total estradiol level, whereas those examining estradiol as a predictor variable were further adjusted for total testosterone level. Then all logistical regression analyses were additionally adjusted for clinic site and the

stratified sex steroid sampling scheme. The statistical significance of these models was tested using χ_2 tests and Fischer's exact tests.

All analyses were performed with SAS (version 9.1; SAS Institute, Cary, NC).

Results

Study population

The 2447 participants in the cross-sectional analyses cohort had a mean age of 73 yr (SD 5.6; range 65–99) and were predominately Caucasian (Table 1). Of these men, 130 (5.3%) were categorized as osteoporotic at the hip and 145 (5.9%) as osteoporotic at the spine. Seventy-three men (3.0%) were total testosterone deficient and 78 (3.2%) were total estradiol deficient. Only 16 participants (0.7%) were deficient in both total testosterone and total estradiol. Of men in the longitudinal analysis cohort, 107 (8.7%) experienced rapid hip bone loss, whereas 42 (3.4%) experienced rapid spine bone loss.

TABLE 1. Baseline characteristics of participants (n = 2447)

Variable	
Mean age, yr (SD)	73.0 (5.6)
Race, % white	76.8
Mean weight, kg (SD)	82.7 (13.5)
Osteoporosis status, hip, % (n)[a]	
Osteoporosis	5.3 (130)
Low bone mass	51.3 (1255)
Normal	43.4 (1062)
Total testosterone category, % (n)[b]	
Deficient	3.0 (73)
Possibly deficient	44.1 (1080)
Normal	52.9 (1294)
<200 ng/dl	3.0 (73)
200 to <300 ng/dl	13.9 (340)
300 to <400 ng/dl	30.2 (738)
400 to <500 ng/dl	25.3 (618)
≥500 ng/dl	27.7 (678)
Total estradiol category, % (n)[c]	
Deficient	3.2 (78)
Possibly deficient	67.6 (1653)
Normal	29.3 (716)
<10 pg/ml	3.2 (78)
10 to <15 pg/ml	27.3 (668)
15 to <20 pg/ml	40.2 (983)
20 to <25 pg/ml	20.1 (491)
≥25 pg/ml	9.3 (227)
Bioavailable testosterone category, intraquintile range, ng/dl (nmol/liter)	
Lowest quintile	<168.6 (<5.8)
Second quintile	168.6 to <200.0 (5.8 to <6.9)
Third quintile	200.0 to <226.1 (6.9 to <7.8)
Fourth quintile	226.1 to <262.2 (7.8 to <9.1)
Highest quintile	≥262.2 (≥9.1)
Bioavailable estradiol category, intraquintile range, pg/ml (pmol/liter)	
Lowest quintile	<8.9 (<32.7)
Second quintile	8.9 to <10.7 (32.7 to <39.3)
Third quintile	10.7 to <12.5 (39.3 to <45.9)
Fourth quintile	12.5 to <15.0 (45.9 to <55.1)
Highest quintile	≥15.0 (≥55.1)

[a] Participants were categorized according to their baseline femoral neck and total hip BMD T-scores using a young Caucasian male reference database from NHANES III as either osteoporotic ($T \leq -2.5$ at either hip site), osteopenic (low bone mass) ($T > -2.5$ at both sites and ≤ -1 at either site), or normal ($T > -1$ at both sites). Participants were categorized according to their spine BMD T-scores using the manufacturer's young male Caucasian reference database as osteoporotic [$T < -2.5$; 5.9% (n = 145)], low bone mass [$T > -2.5$ and < -1; 27.8% (n = 681)], or normal [$T > -1$; 66.2% (n = 1620)].

[b] Total testosterone levels categorized as deficient [<200 ng/dl (< 6.9 nmol/liter)], possibly deficient [200–400 ng/dl (6.9–13.9 nmol/liter)], or normal [>400 ng/dl (> 13.9 nmol/liter)].

[c] Total estradiol levels categorized as deficient [<10 pg/ml (<36.7 pmol/liter)], possibly deficient [10–20 pg/ml (36.7–73.4 pmol/liter)], or normal [> 20 pg/ml (> 73.4 pmol/liter)].

Hip osteoporosis associated with sex steroid deficiency

The prevalence of hip osteoporosis in men with deficient, possibly deficient, and normal total testosterone levels was 12.3, 4.1, and 6.0%, respectively ($P = 0.003$) (Table 2). For every eight men in the MrOS population with total testosterone deficiency, approximately one was osteoporotic. The age and weight-adjusted odds of being categorized as osteoporotic at baseline were increased 3.5-fold in men with total testosterone deficiency, compared with those with normal total testosterone levels (OR 3.5, 95% CI 1.5–7.7). Further adjustment for total estradiol level appeared to somewhat attenuate this association (OR 2.6, 95% CI 1.1–6.1). Analyses categorizing men into narrower total testosterone groups suggested a threshold relationship in that risk for osteoporosis was increased only at total testosterone levels less than 200 ng/dl. The prevalence of osteoporosis and the age- and weight-adjusted odds of being osteoporotic appeared similar in men across different quintiles of bioavailable testosterone. Although men below the fifth percentile for bioavailable testosterone (<126.5 ng/dl, n = 117) appeared to have a modestly increased age- and weight-adjusted odds of osteoporosis (OR 2.0, 95% CI 0.9–4.8), these results were not statistically significant and were attenuated by additional adjustment for total estradiol level (OR 1.4, 95% CI 0.6–3.4).

The prevalence of hip osteoporosis in men with deficient, possibly deficient, and normal total estradiol levels was 15.4, 5.9, and 2.8%, respectively ($P < 0.0001$) (Table 2). For every seven men in the MrOS population with total estradiol deficiency, approximately one was osteoporotic. The age- and weight-adjusted odds of being categorized as osteoporotic at baseline were increased 4.8-fold in men with total estradiol deficiency, compared with men with normal total estradiol levels (OR 4.8, 95% CI 2.1–10.6). Further adjustment for total testosterone level did not alter these results. Additional analyses suggested that risk for osteoporosis increased in a graded fashion for each lower total estradiol category. Similarly, the prevalence of osteoporosis and the age- and weight-adjusted odds of being osteoporotic appeared to increase monotonically with each lower bioavailable estradiol quintile without evidence for a threshold effect. When compared with men in the highest quintile, those below the fifth percentile for bioavailable estradiol (<6.7 pg/ml, n = 117) had a greater than 4-fold increased age- and weight-adjusted risk of osteoporosis (OR 4.4, 95% CI 1.8–10.6). Further adjustment for total testosterone level did not alter these results.

Sex steroid deficiency associated with hip osteoporosis

The prevalence of total testosterone deficiency among men with hip osteoporosis, low bone mass, and normal BMD was 6.9, 2.4, and 3.2%, respectively ($P = 0.01$) (Table 3). For every 14 men in the MrOS population with osteoporosis, approx-

TABLE 2. Proportion and odds of osteoporosis at hip as a function of sex steroid category[a]

	Proportion osteoporotic at hip, % (n/N)	*P* value	Odds of osteoporosis at hip, OR (95% CI)[b]
Total testosterone category[c]			
Deficient	12.3 (9/73)	0.003	3.5 (1.5, 7.7)
Possibly deficient	4.1 (44/1080)		0.9 (0.6, 1.4)
Normal	6.0 (77/1294)		1.0 (ref)
<200 ng/dl	12.3 (9/73)	0.002	3.1 (1.3, 7.0)
200 to <300 ng/dl	2.6 (9/340)		0.6 (0.3, 1.2)
300 to <400 ng/dl	4.7 (35/738)		0.9 (0.6, 1.5)
400 to <500 ng/dl	4.7 (29/618)		0.7 (0.5, 1.2)
≥500 ng/dl	7.1 (48/678)		1.0 (ref)
Bioavailable testosterone category[d]			
Lowest quintile	6.0 (28/470)	0.58	1.4 (0.8, 2.7)
Second quintile	4.0 (19/471)		0.8 (0.4, 1.6)
Third quintile	6.2 (29/471)		1.4 (0.8, 2.5)
Fourth quintile	5.7 (27/471)		1.2 (0.7, 2.2)
Highest quintile	4.9 (23/470)		1.0 (ref)
Total estradiol category[e]			
Deficient	15.4 (12/78)	<0.0001	4.8 (2.1, 10.6)
Possibly deficient	5.9 (98/1653)		1.8 (1.1, 2.9)
Normal	2.8 (20/716)		1.0 (ref)
<10 pg/ml	15.4 (12/78)	<0.0001	5.4 (1.8, 16.6)
10 to <15 pg/ml	6.3 (42/668)		2.0 (0.8, 5.2)
15 to <20 pg/ml	5.7 (56/983)		2.0 (0.8, 5.2)
20 to <25 pg/ml	3.1 (15/491)		1.2 (0.4, 3.4)
≥25 pg/ml	2.2 (5/227)		1.0 (ref)
Bioavailable estradiol category[f]			
Lowest quintile	9.1 (43/470)	<0.0001	2.8 (1.3, 6.0)
Second quintile	7.2 (34/471)		2.9 (1.3, 6.1)
Third quintile	5.1 (24/470)		2.3 (1.0, 5.0)
Fourth quintile	3.4 (16/471)		1.5 (0.6, 3.4)
Highest quintile	1.9 (9/471)		1.0 (ref)

[a] Osteoporosis defined as femoral neck or total hip BMD T-score −2.5 or less.
[b] Age and weight adjusted.
[c] Total testosterone levels categorized as normal [>400 ng/dl (> 13.9 nmol/liter)], possibly deficient [200–400 ng/dl (6.9–13.9 nmol/liter)], or deficient [<200 ng/dl (<6.9 nmol/liter)].
[d] Bioavailable testosterone quintiles in nanograms per deciliter were as follows: less than 168.6, 168.6 to less than 200.0, 200.0 to less than 226.1, 226.1 to less than 262.2, and 262.2 or more.
[e] Total estradiol levels categorized as normal [>20 pg/ml (>73.4 pmol/liter)], possibly deficient [10–20 pg/ml (36.7–73.4 pmol/liter)], or deficient [<10 pg/ml (<36.7 pmol/liter)].
[f] Bioavailable estradiol quintiles in picograms per milliliter were as follows: less than 8.9, 8.9 to less than 10.7, 10.7 to less than 12.5, 12.5 to less than 15.0, and 15.0 or less.

imately one was total testosterone deficient. The age- and weight-adjusted odds of being categorized as total testosterone deficient were increased 3.7-fold in men with osteoporosis (OR 3.7, 95% CI 1.6–8.3). Additional adjustment for total estradiol level appeared to somewhat attenuate this association (OR 2.9, 95% CI 1.2–6.7).

TABLE 3. Proportion and odds of sex steroid deficiency as a function of hip osteoporosis category[a]

	Proportion total testosterone deficient, % (n/N)[b]	*P* value	Odds of total testosterone deficiency, OR (95% CI)[c]
Hip osteoporosis category			
Normal	3.2 (34/1062)	0.01	1.0 (ref)
Low bone mass	2.4 (30/1255)		1.0 (0.6, 1.6)
Osteoporotic	6.9 (9/130)		3.7 (1.6, 8.3)
	Proportion total estradiol deficient, % (n/N)[d]	*P* value	Odds of total estradiol deficiency, OR (95% CI)[c]
Hip osteoporosis category			
Normal	2.4 (25/1062)	0.0001	1.0 (ref)
Low bone mass	3.3 (41/1255)		1.4 (0.8, 2.3)
Osteoporotic	9.2 (12/130)		3.9 (1.8, 8.4)

[a] Participants were categorized according to their baseline femoral neck and total hip BMD T-scores using a young Caucasian male reference database from NHANES III as osteoporotic (T ≤ −2.5 or less at either hip site), osteopenic (low bone mass; T > −2.5 at both sites and ≤ −1 at either site), or normal (T > −1 at both sites).
[b] Total testosterone deficiency defined as less than 200 ng/dl (6.9 nmol/liter).
[c] Age and weight adjusted.
[d] Total estradiol deficiency defined as less than 10 pg/ml (36.7 pmol/liter).

The prevalence of total estradiol deficiency among men with hip osteoporosis, low bone mass, and normal BMD was 9.2, 3.3, and 2.4%, respectively ($P = 0.0001$) (Table 3). For every 11 men in the MrOS population with osteoporosis, approximately one was total estradiol deficient. The age- and weight-adjusted odds of being categorized as total estradiol deficient were increased 3.9-fold in men with osteoporosis (OR 3.9, 95% CI 1.8–8.4). Additional adjustment for total testosterone level did not alter these results.

Rapid hip bone loss associated with sex steroid deficiency

The incidence of rapid hip bone loss in men with deficient, possibly deficient, and normal total testosterone levels was 22.5, 7.9, and 8.6%, respectively ($P = 0.007$). In results adjusted for age, weight and baseline BMD, when compared with participants with normal total testosterone, the odds of subsequent rapid bone loss were 3.2-fold greater in those with total testosterone deficiency (OR 3.2, 95% CI 1.4–7.3) (Table 4). Additional analyses suggested a threshold relationship in that risk for rapid bone loss was increased only at total testosterone levels less than 200 ng/dl. After adjustment for age, weight, and baseline BMD, differences in risk of rapid bone loss were statistically significant across quintiles of bioavailable testosterone but without a clear direction. However, when compared with men in the highest quintile, those below the fifth percentile for bioavailable testosterone appeared at greater risk of rapid bone loss (OR 2.6, 95% CI 1.1–6.1). Additional adjustment for total estradiol level did not alter these results.

The incidence of rapid hip bone loss in men with deficient, possibly deficient, and normal total estradiol levels was 14.3, 9.7, and 6.3%, respectively ($P = 0.08$). In results adjusted for age, weight, and baseline BMD, when compared with participants with normal total estradiol levels, the odds of rapid bone loss appeared 2.1-fold greater in those with total estradiol deficiency, although these results were not statistically significant (OR 2.1, 95% CI 0.7–6.0) (Table 4). Analyses categorizing men into narrower total estradiol groups showed no clear association between total estradiol level and risk of rapid bone loss. Incidence and odds of rapid bone loss appeared to increase modestly but monotonically for each lower bioavailable estradiol quintile

TABLE 4. Proportion and odds of rapid hip bone loss as a function of baseline sex steroid category[a]

	Proportion with rapid hip bone loss, % (n/N)	*P* value	Odds of rapid hip bone loss, OR (95% CI)[b]
Total testosterone category[c]		0.007	
Deficient	22.5 (9/40)		3.2 (1.4, 7.3)
Possibly deficient	7.9 (41/522)		0.9 (0.6, 1.4)
Normal	8.6 (57/665)		1.0 (ref)
<200 ng/dl	22.5 (9/40)	0.03	3.0 (1.3, 7.0)
200 to <300 ng/dl	8.6 (14/163)		0.9 (0.5, 1.9)
300 to <400 ng/dl	7.5 (27/359)		0.8 (0.5, 1.4)
400 to <500 ng/dl	8.0 (25/314)		0.8 (0.5, 1.5)
≥500 ng/dl	9.1 (32/351)		1.0 (ref)
Bioavailable testosterone category[d]		0.02	
Lowest quintile	13.0 (30/230)		1.7 (0.9, 3.2)
Second quintile	8.1 (19/236)		1.0 (0.5, 2.0)
Third quintile	4.9 (11/226)		0.6 (0.3, 1.3)
Fourth quintile	10.6 (27/256)		1.4 (0.8, 2.7)
Highest quintile	7.4 (18/245)		1.0 (ref)
Total estradiol category[e]		0.08	
Deficient	14.3 (5/35)		2.1 (0.7, 6.0)
Possibly deficient	9.7 (77/797)		1.4 (0.9, 2.3)
Normal	6.3 (25/395)		1.0 (ref)
<10 ng/dl	14.3 (5/35)	0.12	1.3 (0.4, 4.2)
10 to <15 pg/ml	9.0 (27/301)		0.8 (0.4, 1.7)
15 to <20 pg/ml	10.1 (50/494)		1.0 (0.5, 2.0)
20 to <25 pg/ml	5.1 (14/276)		0.5 (0.2, 1.1)
≥25 pg/ml	9.1 (11/121)		1.0 (ref)
Bioavailable estradiol category[f]		0.38	
Lowest quintile	11.6 (25/215)		1.6 (0.8, 3.2)
Second quintile	9.1 (20/219)		1.3 (0.7, 2.6)
Third quintile	9.0 (22/244)		1.3 (0.7, 2.6)
Fourth quintile	8.5 (21/247)		1.3 (0.7, 2.6)
Highest quintile	6.3 (17/268)		1.0 (ref)

[a] Rapid bone loss defined as 3% or greater annualized rate of femoral neck or total hip BMD loss between first and second MrOS exams (mean interval 1.8 yr).

[b] Age, weight, and baseline femoral neck BMD adjusted.

[c] Total testosterone levels categorized as normal [>400 ng/dl (>13.9 nmol/liter)], possibly deficient [200–400 ng/dl (6.9–13.9 nmol/liter)], or deficient [<200 ng/dl (<6.9 nmol/liter)].

[d] Bioavailable testosterone quintiles in nanograms per deciliter were as follows: less than 168.6, 168.6 to less than 200.0, 200.0 to less than 226.1, 226.1 to less than 262.2, and 262.2 or greater.

[e] Total estradiol levels categorized as normal [>20 pg/ml (>73.4 pmol/liter)], possibly deficient [10–20 pg/ml (36.7–73.4 pmol/liter)], or deficient [<10 pg/ml (<36.7 pmol/liter)].

[f] Bioavailable estradiol quintiles in picograms per milliliter were as follows: less than 8.9, 8.9 to less than 10.7, 10.7 to less than 12.5, 12.5 to less than 15.0, and 15.0 or greater.

without evidence of a threshold effect. When compared with men in the highest quintile, those below the fifth percentile for bioavailable estradiol appeared to have a 1.9-fold increase in the age-, weight-, and BMD-adjusted odds of rapid bone loss (OR 1.9, 95% CI 0.7–5.1). However, none these results were statistically significant. The above results were not altered by additional adjustment for total testosterone level.

Additional analyses

The prevalence of hip BMD T-score of −2 or less was approximately triple that of osteoporosis [n = 403 (16.5%)], and associations between sex steroid deficiency categories and hip T-score of −2 or less appeared attenuated, compared with their respective associations with osteoporosis. Specifically, the age- and weight-adjusted odds of hip T-score of −2 or less were increased 2.1-fold in men with total testosterone deficiency (OR 2.1, 95% CI 1.1–3.9) and 3.3-fold in men with total estradiol deficiency (OR 3.3, 95% CI 1.8–5.8).

The age- and weight-adjusted odds of being osteoporotic at the spine at baseline were not increased in men with either total testosterone or estradiol deficiency and did not differ across quintiles of bioavailable testosterone (data not shown). However, risk for osteoporosis was increased in the lowest bioavailable estradiol quintile (OR 1.9, 95% CI 1.1–3.4). The odds of total testosterone or estradiol deficiency were not increased in men with spine osteoporosis. Last, the age-, weight-, and baseline BMD-adjusted odds of rapid spine bone loss appeared possibly increased by either deficient total testosterone (OR 2.4, 95% CI 0.7–8.9) or deficient total estradiol (OR 3.4, 95% CI 0.9–13.6), although neither result was statistically significant.

Additional adjustment of logistical regression analyses for clinic site and the sex steroid stratified sampling scheme did not alter our results.

Discussion

In the present study, we found that older men with total testosterone or total estradiol deficiency were more likely to be osteoporotic at the hip. Conversely, older men with hip osteoporosis were more likely to be total testosterone or total estradiol deficient. Older men with total testosterone deficiency, and possibly men with total estradiol deficiency, were at increased risk of subsequent rapid hip bone loss. There was little association of testosterone or estradiol deficiency with baseline spine osteoporosis, but results suggested that both sex steroid deficiencies may increase risk of subsequent rapid spine bone loss.

Our analyses, in suggesting that total estradiol deficiency was possibly a stronger predictor of hip osteoporosis than was total testosterone deficiency and that low bioavailable estradiol was associated with osteoporosis whereas low bioavailable testosterone was not, are generally consistent with other studies in older men (1–6). However, our finding that total testosterone deficiency was associated with osteoporosis independent of estradiol levels contrasts with findings of previous studies (5, 8). Similarly, our findings that low total and bioavailable testosterone appeared to be possibly stronger predictors of rapid hip bone loss than were low total and bioavailable estradiol are not consistent with previous reports (3, 5, 8). Whereas previous studies have used correlations to compare the relative strengths of association of total sex steroid levels and their respective bioavailable fractions with BMD and bone loss, this was not the purpose of the present study given its cut point-based analysis approach and the lack of clearly established thresholds for bioavailable testosterone or estradiol deficiency.

The present study also appears to contrast with previously published work in its failure to identify a threshold association of bioavailable estradiol with risk for osteoporosis and rapid bone loss. Previous longitudinal analyses conducted in 130 men aged 60–90 yr had suggested that men with total estradiol levels less than 31 pg/ml (and bioavailable estradiol levels < 11 pg/ml) had higher rates of bone loss than men with levels above these thresholds (8). Conclusions drawn by authors of this prior study appear to have been based primarily on observed rates of bone loss at the radius and ulna. However, the elderly men in this earlier study experienced a net gain in BMD at the total hip and net loss in BMD at the spine, and authors reported that there was no threshold effect of bioavailable estradiol at these skeletal sites. The current study examined the prevalence and odds of osteoporosis and rapid bone loss at the hip and spine but did not collect BMD data at other skeletal sites.

The biochemical criteria used to define total testosterone and estradiol deficiency were based on previously published recommendations (18–23), although within the range of suggested cut points we chose those that classified only a small proportion of MrOS men as abnormal. By comparison, because there are no conventional definitions for bioavailable testosterone or estradiol deficiency, we categorized men into quintiles for these variables. Then, because even the lowest quintiles classified a large proportion of MrOS men as abnormal and could have attenuated any association of low bioavailable testosterone or estradiol with osteoporosis and rapid bone loss, we further examined the risk for osteoporosis and rapid bone loss in men in the lowest fifth percentile of these bioavailable sex steroid fractions.

Our findings have several implications for clinical practice and future research. These data suggest that for older men with documented total testosterone or estradiol deficiency, measurement of BMD should be considered. In MrOS, for every eight men with total testosterone deficiency and every seven men with total estradiol deficiency, approximately one was identified as osteoporotic. Clinician awareness of osteoporosis in these men is relevant because bisphosphonate treatment may reduce their subsequent risk of fractures (26).

While study data suggest that older men with osteoporosis have an increased prevalence of total testosterone or estradiol deficiency, it is uncertain whether measurement of sex steroid levels in osteoporotic men provides additional information that should change their clinical management. By virtue of being osteoporotic, these men may already be candidates for bisphosphonate therapy (26), and there is as yet no evidence that sex hormone treatment provides additional fracture protection or that sex steroid levels predict BMD response to bisphosphonates (27). Whereas clinical practice in this area remains controversial, individuals in this population may be candidates for randomized trials examining

the effect of testosterone, selective androgen receptor modulators, or selective estrogen receptor modulators in comparison with or in combination with bisphosphonates or other therapies on changes in BMD and fracture. Planned clinical trials to establish whether potential nonbone benefits of testosterone treatment outweigh risks in older men will further inform clinical decision making (28).

Finally, our findings suggest that the cut points used here for defining osteoporosis, total testosterone deficiency, and total estradiol deficiency may be useful in the clinical setting, at least with respect to the association between sex steroids and bone. Our results indicate that less extreme cut points for total testosterone deficiency and total estradiol deficiency are likely to be weaker predictors of osteoporosis and rapid bone loss. Future studies, including an ongoing analysis of MrOS data (29), may determine whether there are better clinical thresholds for defining total testosterone and estradiol deficiency in older men and whether there are clinically useful cut points for defining deficiency of bioavailable estradiol and testosterone. It also will be important to examine whether these measures can be shown to predict incident fractures.

This study has numerous strengths. To our knowledge, it is the largest study to examine the association of sex steroids with BMD in older men and one of relatively few to examine the longitudinal association of sex steroids with bone loss in older men. However, its greater value may be that it examines the association between sex steroids and bone in older men from a clinical perspective, using defined cut points that may guide decision making.

This study also has several limitations. First, we used unextracted RIA methods to determine total sex steroid levels. Although stringent quality control procedures were used to increase assay precision, this technique is somewhat less reliable in measuring very low total estradiol levels [for men with total estradiol $\leq$ 10 pg/ml (n = 78), intraassay CV 22.3%]. Second, although men were classified as total testosterone or estradiol deficient based solely on biochemical criteria and without consideration of clinical symptoms, the cut points used were stringent and should have identified men who were more severely hypogonadal (20). Third, our definition for rapid bone loss was based on the rate of bone loss that predicted increased risk of incident fracture in a population of postmenopausal women (25) because no such data have been reported in older men. Fourth, associations between sex steroid measurements and spine BMD could have been partially confounded by vertebral osteoarthritis and/or aortic calcification. Finally, because MrOS participants are community dwelling, and predominately Caucasian, study findings may not apply to other population groups.

In conclusion, we found that older men with total testosterone or estradiol deficiency were more likely to have osteoporosis. Conversely, older men with osteoporosis were more likely to have total testosterone or estradiol deficiency. Older men with total testosterone deficiency were more likely to experience subsequent rapid bone loss. BMD testing of older men with sex steroid deficiency may be warranted to identify those with osteoporosis who potentially may benefit from bisphosphonate therapy. At present, there is insufficient evidence that identifying sex steroid deficiency in older osteoporotic men warrants changes in patient management, but data regarding the prevalence of sex steroid deficiency in this population may aid in planning future clinical trials.

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References

1. **Khosla S, Melton III LJ, Atkinson EJ, O'Fallon WM, Klee GG, Riggs BL** 1998 Relationship of serum sex steroid levels and bone turnover markers with bone mineral density in men and women: a key role for bioavailable estrogen. J Clin Endocrinol Metab 83:2266–2274
2. **Center JR, Nguyen TV, Sambrook PN, Eisman JA** 1999 Hormonal and biochemical parameters in the determination of osteoporosis in elderly men. J Clin Endocrinol Metab 84:3626–3635
3. **Van Pottelbergh I, Goemaere S, Kaufman JM** 2003 Bioavailable estradiol and an aromatase gene polymorphism are determinants of bone mineral density changes in men over 70 years of age. J Clin Endocrinol Metab 88:3075–3081
4. **Szulc P, Munoz F, Claustrat B, Garnero P, Marchand F, Duboeuf F, Delmas PD** 2001 Bioavailable estradiol may be an important determinant of osteoporosis in men: the MINOS study. J Clin Endocrinol Metab 86:192–199
5. **Gennari L, Merlotti D, Martini G, Gonnelli S, Franci B, Campagna S, Lucani B, Dal Canto N, Valenti R, Gennari C, Nuti R** 2003 Longitudinal association between sex hormone levels, bone loss, and bone turnover in elderly men. J Clin Endocrinol Metab 88:5327–5333
6. **Greendale GA, Edelstein S, Barrett-Connor E** 1997 Endogenous sex steroids and bone mineral density in older women and men: the Rancho Bernardo Study. J Bone Miner Res 12:1833–1843
7. **van den Beld AW, de Jong FH, Grobbee DE, Pols HA, Lamberts SW** 2000 Measures of bioavailable serum testosterone and estradiol and their relationships with muscle strength, bone density, and body composition in elderly men. J Clin Endocrinol Metab 85:3276–3282
8. **Khosla S, Melton III LJ, Atkinson EJ, O'Fallon WM** 2001 Relationship of serum sex steroid levels to longitudinal changes in bone density in young versus elderly men. J Clin Endocrinol Metab 86:3555–3561
9. **Faulkner KG, Orwoll E** 2002 Implications in the use of T-scores for the diagnosis of osteoporosis in men. J Clin Densitom 5:87–93
10. **Vallarta-Ast N, Krueger D, Binkley N** 2002 Densitometric diagnosis of osteoporosis in men: effect of measurement site and normative database. J Clin Densitom 5:383–389
11. **Mohr BA, Guay AT, O'Donnell AB, McKinlay JB** 2005 Normal, bound and nonbound testosterone levels in normally ageing men: results from the Massachusetts Male Ageing Study. Clin Endocrinol (Oxf) 62:64–73

12. **Barrett-Connor E** 2005 Male testosterone: what is normal? Clin Endocrinol (Oxf) 62:263–264
13. **Kanis JA, Gluer CC** 2000 An update on the diagnosis and assessment of osteoporosis with densitometry. Committee of Scientific Advisors, International Osteoporosis Foundation. Osteoporos Int 11:192–202
14. **Orwoll E, Blank JB, Barrett-Connor E, Cauley J, Cummings S, Ensrud K, Lewis C, Cawthon PM, Marcus R, Marshall LM, McGowan J, Phipps K, Sherman S, Stefanick ML, Stone K** 2005 Design and baseline characteristics of the osteoporotic fractures in men (MrOS) study—a large observational study of the determinants of fracture in older men. Contemp Clin Trials 26:569–585
15. **Blank JB, Cawthon PM, Carrion-Petersen M, Harper L, Johnson JP, Mitson E, Delay RR** 2005 Overview of recruitment for the osteoporotic fractures in men study (MrOS). Contemp Clin Trials 26:557–568
16. **Sodergard R, Backstrom T, Shanbhag V, Carstensen H** 1982 Calculation of free and bound fractions of testosterone and estradiol-17β to human plasma proteins at body temperature. J Steroid Biochem 16:801–810
17. **Vermeulen A, Verdonck L, Kaufman JM** 1999 A critical evaluation of simple methods for the estimation of free testosterone in serum. J Clin Endocrinol Metab 84:3666–3672
18. **Liu PY, Swerdloff RS, Veldhuis JD** 2004 Clinical review 171: the rationale, efficacy and safety of androgen therapy in older men: future research and current practice recommendations. J Clin Endocrinol Metab 89:4789–4796
19. **Conway AJ, Handelsman DJ, Lording DW, Stuckey B, Zajac JD** 2000 Use, misuse and abuse of androgens. The Endocrine Society of Australia consensus guidelines for androgen prescribing. Med J Aust 172:220–224
20. **Snyder PJ** 2004 Hypogonadism in elderly men—what to do until the evidence comes. N Engl J Med 350:440–442
21. **Report of National Institute on Aging Advisory Panel on Testosterone Replacement in Men** 2001 J Clin Endocrinol Metab 86:4611–4614
22. **Cunningham GR, Swerdloff RS** 2006 Summary from the Second Annual Andropause Consensus Meeting. 2001. Available at: http://www.endo-society.org/publicpolicy/legislative/upload/andro_cc_summary.pdf. Accessed January 24
23. **Khosla S, Melton III LJ, Riggs BL** 2002 Clinical review 144: estrogen and the male skeleton. J Clin Endocrinol Metab 87:1443–1450
24. **Looker AC, Wahner HW, Dunn WL, Calvo MS, Harris TB, Heyse SP, Johnston Jr CC, Lindsay R** 1998 Updated data on proximal femur bone mineral levels of US adults. Osteoporos Int 8:468–489
25. **Hansen MA, Overgaard K, Riis BJ, Christiansen C** 1991 Role of peak bone mass and bone loss in postmenopausal osteoporosis: 12 year study. BMJ 303:961–964
26. **Orwoll E, Ettinger M, Weiss S, Miller P, Kendler D, Graham J, Adami S, Weber K, Lorenc R, Pietschmann P, Vandormael K, Lombardi A** 2000 Alendronate for the treatment of osteoporosis in men. N Engl J Med 343:604–610
27. **Drake WM, Kendler DL, Rosen CJ, Orwoll ES** 2003 An investigation of the predictors of bone mineral density and response to therapy with alendronate in osteoporotic men. J Clin Endocrinol Metab 88:5759–5765
28. **Liverman CT, Blazer DG, eds.** 2004 Testosterone and aging: clinical research directions. Washington, D.C.: National Academies Press
29. **Cauley J, Taylor B, Fink H, Ensrud K, Bauer D, Barrett-Connor E, Marshall L, Nevitt M, Stefanick M, Orwoll E** 2004 Sex steroid hormones in older men: cross-sectional and longitudinal associations with bone mineral density (BMD). The Osteoporotic Fracture in Men Study (MrOS). J Bone Miner Res 19(Suppl 1):S16

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