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Sex Steroids, Periodontal Health, and Tooth Loss in Older Men

ABSTRACT

Sex steroids have a significant effect on skeletal biology in men, with reduced levels being associated with lower skeletal bone mass and cortical thickness. The purpose of this study was to determine if sex steroids are associated with periodontitis and tooth loss in a cohort of 1210 older dentate men followed for 3 years. Periodontal measures included attachment loss, pocket depth, gingival bleeding, and number of teeth. Baseline serum testosterone and estradiol were measured by radio-immunoassay. Severe periodontitis was common at baseline (38%), and progression occurred in 32% of the cohort. Incident tooth loss occurred in 22% of the cohort. Testosterone, estradiol, and sex hormone binding globulin (SHBG) concentrations were not related to baseline periodontal status or number of teeth. Moreover, there was no relationship between sex steroid levels and periodontitis progression or incident tooth loss. Although periodontitis, progression of periodontitis, and tooth loss are common in older men, they were not associated with sex steroids.

KEY WORDS: periodontitis, testosterone, estradiol, men.

INTRODUCTION

Gonadal steroids have effects on skeletal biology in men. Hypogonadism is associated with bone loss and fracture risk (Shahinian *et al*., 2005), and both estrogens and androgens affect bone mass in men (Vanderscheuren *et al*., 1998; Khosla *et al*., 2001; Orwoll and Klein, 2001). With aging, estradiol and testosterone levels decline in men (Harman *et al*., 2001; Orwoll *et al*., 2006), and reduced levels have been linked to bone loss (Khosla and Riggs, 2005). Lower estradiol levels have been most clearly related to skeletal health and are associated with lower trabecular bone mass, cortical thickness, cortical density, and trabecular thickness (Khosla *et al*., 2006). In light of these data, it is reasonable that sex steroids may also affect oral bone metabolism, the likelihood of periodontal disease, and tooth loss.

In fact, sex steroids have been linked to oral bone health. Estrogen use may provide protection against tooth loss in menopausal women (Taguchi *et al*., 2004), and testosterone may be related to periodontal health in hypogonadotrophic men (Daltaban *et al*., 2006). Moreover, both estrogen and androgen may have direct effects on the periodontium (Mealey and Moritz, 2003; Soory and Tilakaratne, 2003a,b). A variety of potential mechanisms might explain the effects of sex steroids on periodontal disease, including modulation of immunological events (Taubman *et al*., 2005; Wactawski-Wende *et al*., 2005).

Despite this information, the relationship between periodontal health and sex steroid levels has not been examined in older men. Here we determine those relationships in a large population. Moreover, in a longitudinal evaluation, we examine the hypothesis that baseline sex steroid levels influence the progression of periodontitis and incident tooth loss.

MATERIALS & METHODS

The Osteoporotic Fractures in Men (MrOS) Study is a prospective, observational study of a cohort of community-dwelling ambulatory men ≥ 65 yrs old. During 2000-2002, MrOS recruited 5995 older men at six clinical sites. All participants completed a questionnaire concerning demographic characteristics, medical history, tobacco and alcohol use, and lifestyle. Participants also took part in a baseline visit that included measures of height and weight and collection of morning fasting blood. Information on MrOS recruitment strategies and study design has been published elsewhere (Blank *et al*., 2005; Orwoll *et al*., 2005). In 2002-2003, study participants at two of the clinical sites were enrolled in the MrOS Dental Study. Men who completed the first MrOS Dental Study visit (Visit 1) were invited to a second visit (Visit 2) in 2005-2006, an average of 2.7 yrs after Visit 1 (SD = 0.2, range = 1.9-3.5). This study was approved by the Institutional Review Boards at Oregon Health & Science

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University and the University of Alabama at Birmingham; informed consent was obtained from all study participants.

Periodontal Assessment

All examinations were completed by calibrated dentists and hygienists. Missing teeth and implants were recorded for the full mouth, while the periodontal examination was limited to a random half-mouth. Clinical attachment loss (CAL) and pocket depth (PD) were measured at 6 sites *per* tooth, as previously described (Phipps *et al*., 2007).

We used the periodontitis and periodontitis progression case definitions proposed by the European Workshop in Periodontology (Tonetti *et al*., 2005), with one modification to account for our half-mouth design. Our criterion for a periodontitis case was the presence of proximal attachment loss of ≥ 5 mm in at least 30% of teeth examined, rather than teeth present, while the criterion for periodontitis progression was the presence of 2 or more teeth demonstrating a longitudinal loss of proximal attachment of ≥ 3 mm. Since we examined only a half-mouth in each participant, it is likely that we underestimated the prevalence and progression of periodontitis.

Incident Tooth Loss

If a participant lost a tooth between Visits 1 and 2, his dentist was contacted to collect information on date of and reason for extraction. Of the 371 men with at least 1 lost tooth, reason for extraction was obtained for 261 (69.6%).

Sex Steroid Measures

Morning serum samples were obtained for sex steroid measures. Total serum estradiol and testosterone levels and sex hormone binding globulin (SHBG) concentrations were measured by radioimmunoassay, and free estradiol and testosterone levels were calculated, as previously described in detail (Orwoll *et al*., 2006).

Statistical Analyses

Bivariate statistical comparisons were made with chi-square tests, *t* tests, or Pearson correlation coefficients. Linear and logistic regression models were used to estimate the relationship between sex steroid levels and periodontal measures adjusted for study site, examiner nested within study site, age at Visit 1, non-white race, college education, body mass index (BMI), diabetes, fair/poor/very poor self-reported general health status, and a history of 20 or more pack-years of smoking. Individual models were developed for total testosterone, total estradiol, SHBG, free testosterone, and free estradiol. As a further control for smoking, similar analyses were generated for never-smokers and current non-smokers. Given that 1019 men completed the follow-up visit, this study had 80% power to detect an R^2 of 0.007 attributed to each sex steroid variable, and an odds ratio of 0.80 *per* standard deviation increase in each sex steroid.

We assessed inter-examiner reliability for the periodontal examination on a subset of 56 participants who had a second periodontal examination administered by the study's periodontist. Nested models were used to summarize the percentage of total variability explained by the examiner. The inter-examiner correlation was 0.79 for CAL and 0.78 for PD. To account for inter-examiner error, all statistical models using periodontal examination data include 'examiner nested within site' as a covariate. For change and progression measures, the examiners at both the first and follow-up visits were included in models.

RESULTS

Participant Characteristics

The 1210 dentate men examined at Visit 1 ranged in age from 66-95 yrs, with a mean of 74.6 yrs. The mean number of teeth present was 22.5 (range, 1-28). The mean gingival index was 1.2 ± 0.5, and the prevalence of gingival bleeding was 53%. Mean CAL was 3.0 ± 0.8 mm; 82% had at least one site with CAL ≥ 5 mm. The mean PD was 2.5 ± 0.5 mm; 85% had at least one site with a PD ≥ 4 mm, while 34% had at least one site with PD ≥ 6 mm. Severe periodontitis was evident in 38% of the men.

Eighty-four percent (n = 1019) of the dentate men who completed Visit 1 attended Visit 2. Reasons for not attending Visit 2 included death (n = 76) and refusal (n = 115). The men who did not attend Visit 2 were older, less likely to have attended college, and had higher mean pack-years of smoking (Table 1). Of those who did not attend, 48% had periodontitis at baseline compared with 36% of those who did attend Visit 2 (Table 1). During follow-up, 9% of men with baseline periodontitis died compared with 5% without periodontitis (p = 0.02).

Sex Steroid Levels

Sex steroid and SHBG levels were similar to those reported in other cohorts of older men (Ferrini and Barrett-Connor, 1998; Travison *et al*., 2007). Increasing age was associated with slightly lower total and free testosterone levels (r = –0.1, p = 0.0002 and r = –0.2, p < 0.0001, respectively), lower total and free estradiol levels (r = –0.1, p = 0.0002 and r = –0.17, p < 0.0001, respectively), and higher SHBG levels (r = 0.17, p < 0.0001). Current smokers had higher SHBG levels (60.8 *vs*. 48.8, p = 0.02), and participants who rated their health as fair/poor had lower total testosterone (12.1 *vs*. 13.2 nmol/L, p = 0.02) and free testosterone (0.24 *vs*. 0.26 nmol/L, p = 0.03) levels than did those with excellent/good health status. Those with fair/poor health had similar levels of total (62.0 *vs*. 62.3 pmol/L, p = 0.9) and free estradiol (1.63 *vs*. 1.61 pmol/L, p = 0.8).

Sex Steroids and Periodontal Health at Baseline

In analyses adjusted for relevant covariates, sex steroid and SHBG levels were not associated with mean CAL, mean PD, or gingival bleeding (Appendix Table 1, Appendix Figs. 1, 2). Although men with severe periodontitis were slightly older, were more often non-white, and were more likely to smoke, the levels of sex steroids were not different in men with and without severe periodontitis (Appendix Table 2). After adjusting for covariates, we found no significant difference in the proportions of men with severe periodontitis among participants with low testosterone (testosterone ≤ 8.7 nmol/L) *vs*. higher testosterone

Table 1. Comparison of Baseline Characteristics of Men Who Did or Did Not Attend Visit 2

Characteristic	Attended Visit 2		
	No (N = 191)	Yes (N = 1019)	p-value
Mean age (yrs)	76.4	74.3	< 0.01
Non-Hispanic White (%)	86.9	90.0	0.20
Education—college or higher (%)	52.4	61.4	0.02
Mean pack-years of smoking	21.9	15.2	< 0.01
Mean body mass index (kg/m^2)	27.4	27.4	0.94
Mean clinical attachment loss (mm)	3.1	3.0	0.04
Mean pocket depth (mm)	2.5	2.5	0.89
Periodontal disease (%)	47.6	36.4	< 0.01

levels (43% *vs*. 37%, p = 0.13). Sex steroid levels were not associated with periodontal measures in models containing total estradiol, total testosterone, and SHBG simultaneously with covariates (data not shown). In addition, we found no significant association ($p \leq 0.01$) between periodontal health and sex steroids when the analyses were restricted to never-smokers or current non-smokers (data not shown).

We examined whether sex steroid levels were lower in edentulous men at Visit 1 (n = 136) to assess whether men with low levels had early tooth loss. We found no significant differences in mean total or free testosterone or estradiol between edentulous and dentate men (data not shown).

Sex Steroids and Progression of Periodontitis

There was progression of periodontal disease in 32% of men. Periodontal progression differed significantly by site (Portland, 19%; Birmingham, 46%) and by examiner. In multivariable analyses, there was no association between progression of periodontitis and sex steroid or SHBG levels measured at Visit 1 after adjustment for covariates including site and examiner (Table 2). Similarly, there was no association between sex steroid levels and the rate of change in CAL (Table 2).

Tooth Loss

Eighty-five percent of participants were missing at least 1 tooth at baseline, and additional tooth loss occurred in 22% of men during follow-up. Periodontitis was cited as the cause of extraction in 16%. There was no association between sex steroid levels and number of teeth at baseline, and multivariate analyses with incident tooth loss as the dependent variable found that sex steroids were not independently associated with tooth loss (Table 3).

DISCUSSION

In a large population of community-dwelling older men, periodontitis was common, as were progression of periodontal disease and tooth loss. However, sex steroid and SHBG levels were not associated with baseline periodontal measures, worsening of periodontal disease, history of tooth loss, or incident tooth loss.

Table 2. Relationship between Periodontitis Progression and Longitudinal Change in CAL *per* Standard Deviation Difference in Sex Steroid Levels

Sex Steroid		Slope of Change in Mean CAL (*per* SD) mm (p-value)	Odds Ratio of Periodontitis (*per* SD) Progression (95% CI)
Total	Estradiol	0.018 (0.34)	1.00 (0.85, 1.16)
	Testosterone	–0.006 (0.74)	0.97 (0.83, 1.14)
	SHBG	0.003 (0.87)	0.99 (0.85, 1.16)
Free	Estradiol	0.016 (0.40)	0.99 (0.84, 1.15)
	Testosterone	–0.012 (0.54)	0.93 (0.78, 1.09)

Linear regression and logistic models adjusted for clinic site, examiner, age, non-white race, college education or higher, BMI, diabetes, fair/poor/very poor general health, 20+ pack-years of smoking.
SD = Standard deviation.
CAL = Clinical attachment loss, mm.
95% CI = 95 percent confidence interval.

The study had considerable power to detect associations, and it is unlikely that meaningful relationships were not detected. These findings strongly suggest that sex steroid levels do not influence periodontal health in older men.

Although there are few data available concerning this issue, our findings are somewhat surprising. Testosterone and estradiol levels decline in men with aging, and many of the common sequelae of aging, including loss of bone density and the appearance of frailty, have been linked to a relative deficiency of sex hormone action (Matsumoto, 2002). For example, in the overall MrOS study, low levels of estradiol and testosterone were associated with reduced bone mineral density (Mellström *et al*., 2006) and loss of bone density (Fink *et al*., 2006). Moreover, MrOS men with lower sex steroid levels were at increased risk of fracture (Mellström *et al*., 2006, 2008). Attention has been drawn to the possible relationships among periodontal disease, oral bone loss, and post-menopausal osteoporosis (Krall, 2001; Jeffcoat, 2005). Estrogen inhibits the expression of inflammatory cytokines important in bone resorption, and estrogen deficiency may contribute to more intense gingival inflammation during periodontitis and subsequent oral bone loss, and may result in bone loss at both oral and skeletal sites (Lerner, 2006). Several studies have suggested that the risk of post-menopausal tooth loss is reduced by estrogen replacement (Lerner, 2006). Furthermore, lower estrogen levels have been linked to gingival inflammation (Norderyd *et al*., 1993) and reduced clinical attachment levels (Reinhardt *et al*., 1999). Nevertheless, in this population of older men, we found no relationship between estrogen levels and periodontitis or tooth loss. The reasons for this finding and the previous associations between oral health and estrogen in older women are not clear. One possibility is that older men have considerably higher levels of estrogen than do post-menopausal women, and there may be a threshold estrogen concentration needed for the prevention of oral bone loss. In fact, it has been suggested that there is a threshold level of estradiol in older men above which there is protection against skeletal bone loss (Khosla *et al*., 2002).

Testosterone also has a positive effect on bone in men (Wiren and Orwoll, 1999; Vanderschueren *et al*., 2004), but the

Table 3. Relationship between Baseline Sex Steroid Levels and Number of Teeth at Baseline and Incident Tooth Loss

Sex Steroid		Slope of Number of Teeth *per* Standard Deviation (SD) Difference in Sex Steroid Levels (p-value)	Odds Ratio for Incident Tooth Loss *per* SD Difference in Sex Steroid Levels (95% CI)
Total	Estradiol	–0.034 (0.67)	0.92 (0.83, 1.10)
	Testosterone	0.227 (0.18)	1.01 (0.87, 1.18)
	SHBG	–0.054 (0.75)	1.06 (0.91, 1.23)
Free	Estradiol	–0.090 (0.60)	0.90 (0.76, 1.06)
	Testosterone	0.164 (0.33)	0.95 (0.82, 1.11)

Linear regression and logistic models adjusted for clinic site, age, non-white race, college education or higher, BMI, diabetes, fair/poor/very poor general health, 20+ pack-years of smoking.

relationships between testosterone levels and oral health have rarely been studied (Daltaban *et al*., 2006). Our findings provide no support for a relationship between serum testosterone and periodontal or oral bone health.

This study had considerable strengths. The MrOS Dental population was large, and was studied with carefully controlled dental measures. Sex steroid measurements were similarly carefully performed, and there was a wide range of concentrations of testosterone and estradiol present in the cohort. Assessments of multiple potential covariates were available. There were also several potential limitations. The men studied were volunteers, and their general health and socio-economic status were generally good. Thus, these results may not be representative of the whole population of older men. Moreover, there were different rates of progression of periodontitis in Portland and Birmingham. This difference may reflect real geographical variation in the nature of periodontitis, but, despite careful cross-training and calibration, it also may represent site differences in measurement techniques. Nevertheless, periodontitis was common at baseline, and progression of disease occurred in many participants at both clinic sites. It is highly likely that a meaningful effect of sex steroids on the course of periodontal disease would have been detected. The population studied was primarily Caucasian, and these results may not be generalizable to other racial groups. Whereas we found no associations between sex steroid levels and periodontal status, this cannot be taken as evidence that sex steroid supplementation would have no effect.

In summary, this is the first large study of the associations between periodontal disease and sex steroid levels in men. We found no evidence that serum sex steroid concentrations were associated with measures of periodontal disease, subsequent progression of disease, the number of teeth at baseline, or incident tooth loss.

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