Effects of Potent Antiretroviral Therapy on Free Testosterone Levels and Fat-Free Mass in Men in a Prospective, Randomized Trial: A5005s, a Substudy of AIDS Clinical Trials Group Study 384

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Background. Low testosterone levels are commonly reported in patients with advanced human immunodeficiency virus disease. The effects of initiation of different antiretroviral regimens on testosterone levels and changes in fat-free mass have not been reported.

Methods. Antiretroviral-naive men ($n = 213$) were randomized to receive nelfinavir, efavirenz, or both plus either zidovudine and lamivudine or stavudine and didanosine. Patients underwent measurements of metabolic parameters, including determination of free testosterone level by equilibrium dialysis and bioelectrical impedance analysis, over a 64-week period.

Results. At baseline, the median free testosterone level was 92 pg/mL; the level was subnormal (i.e., <50 pg/mL) in 6%. Lower CD4 cell count at the time of study entry, higher weight, and greater age were independently associated with lower baseline free testosterone level. At week 64, the median free testosterone level increased more in zidovudine-lamivudine recipients (48 of whom had paired values available; change, +31 pg/mL) than in stavudine-didanosine recipients (57 of whom had paired values; change, +3 pg/mL; $P = .001$, by Wilcoxon rank sum test), and it increased more in efavirenz recipients (37 of whom had paired values; change, +30 pg/mL) than in nelfinavir recipients (28 of whom had paired values; change, −3 pg/mL; $P = .05$). The median fat-free mass for the entire group increased by 1.2 kg at week 64 (change, +2.0%; $P < .001$); the increase was greater in the zidovudine-lamivudine group ($n = 70$; change, +1.8 kg) than in the stavudine-didanosine group ($n = 79$; change, +0.5 kg; $P = .04$), and the increase was also greater for efavirenz recipients ($n = 53$; change, +2.1 kg) than among nelfinavir recipients ($n = 47$; change, +0.4 kg; $P = .003$). White race, lower CD4 cell count at study entry, assignment to the efavirenz treatment arm, and assignment to the zidovudine-lamivudine treatment arm independently predicted greater absolute change in fat-free mass at week 64.

Conclusions. Subnormal free testosterone levels occurred infrequently among these antiretroviral-naive men. Free testosterone and fat-free mass levels increased after initiation of antiretroviral therapy, with greater increases at 64 weeks among zidovudine-lamivudine recipients than among stavudine-didanosine recipients and among efavirenz recipients than among nelfinavir recipients.

Low serum testosterone levels have been commonly reported among men with HIV infection [1, 2], particularly among those with more advanced disease and lower CD4 cell counts [3]. Low levels of testosterone are associated with wasting [4–6], depression, bone loss, and increased morbidity. However, data on changes in testosterone levels after the initiation of potent anti-

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Members of the study group are listed at the end of the text.

retroviral therapy are sparse and derive from uncontrolled studies [7, 8]. Importantly, many studies have only reported total testosterone levels, which may not accurately reflect free bioactive testosterone concentrations because of increased sex hormone–binding globulin concentrations among men with HIV infection [3, 9].

Lean body mass primarily reflects muscle mass, and greater lean body mass correlates with greater serum testosterone concentrations in HIV-infected individuals [4]. Moreover, the use of physiologic testosterone supplementation in HIV-infected men with low testosterone levels has resulted in increases in lean body mass that correlated with increases in testosterone concentrations [2]. Use of potent antiretroviral therapy increases fat-free mass (FFM), as estimated by bioelectrical impedance analysis, particularly among persons with lower CD4 cell counts and higher HIV RNA levels before the commencement of treatment [10]. However, no data are currently available that compare the effects of initiation of different antiretroviral drug regimens on changes in FFM in patients who are initiating antiretroviral therapy.

The purpose of this analysis was to describe the prevalence of low free testosterone levels in men without antiretroviral experience and to compare the effects of different antiretroviral regimens on the changes in free testosterone levels and FFM over time. We report findings from a subset of participants enrolled in a large, randomized, prospective trial of initial antiretroviral therapy (AIDS Clinical Trials Group [ACTG] study 384 [11, 12]) who underwent serial evaluation of metabolic parameters and body composition at baseline and periodically after assignment to receive various antiretroviral regimens.

METHODS

Participants. HIV-infected individuals were eligible for entry into ACTG study 384 [11, 12] if they had prior antiretroviral experience of <7 days and an HIV-1 RNA level of >500 copies/mL. This metabolic substudy, A5005s [13], enrolled a total of 334 of the 980 ACTG 384 participants at 23 participating sites in the United States during the period 1998–1999; study follow-up continued through 2001. Participants were excluded from the A5005s substudy if they were known to have uncontrolled hypogonadism, were receiving supraphysiologic doses of androgens or glucocorticoids, had serum triglyceride levels >750 mg/dL, or had any history of Cushing disease or diabetes mellitus [13]. Two hundred thirteen male participants who had testosterone levels measured at entry and who were not receiving androgen supplementation were included in this intent-to-treat analysis. No participant was receiving androgen supplementation (either physiologic or supraphysiologic doses) at study entry. Five participants who were enrolled in the metabolic substudy started receiving androgens after entry and, thus, were not included in the current analysis; all 5 had normal levels of free testosterone at study entry. All participants provided written informed consent in accordance with the guidelines of each site's institutional review board. Participants were randomized to 1 of 6 treatment arms to receive nelfinavir, efavirenz or both, combined with zidovudine plus lamivudine or didanosine plus stavudine [11, 12].

Bioelectrical impedance analysis. Resistance and reactance were measured with a bioelectric impedance analyzer (RJL) on the right side of the body using techniques that were standardized across ACTG sites. Standardized measurements of height and weight obtained on the same visit. Participants were weighed on calibrated scales after removing shoes, outdoor clothing, and other heavy items. Analyzers were calibrated monthly. Calculations of fat mass and FFM were performed centrally, using published equations that have been validated in studies of HIV-infected subjects [14].

Assays. Free testosterone concentration (normal range for adult men, 50–210 pg/mL) was calculated as the product of total testosterone (as measured by radioimmunoassay) and free testosterone percentage. The free testosterone percentage was determined by equilibrium dialysis and was corrected for dilution using the formula of Vermeulen et al. [15]. Testosterone assays were performed at Quest Diagnostics' Nichols Institute (San Juan Capistrano, CA) at study entry and on weeks 16, 32, 48, and 64. Assays used serum samples that had been stored at −70°C, and all assays for each individual participant were conducted in a single batch. Other assays were performed as described elsewhere [13]. Serum samples were all collected while the patient was fasting and, with few exceptions, in the morning.

Statistical analyses. Data are summarized as medians (with interquartile ranges [IQRs]) or as counts and percentages, as appropriate. Changes over time were assessed within groups using the Wilcoxon signed rank test. Variables were compared between groups using the Wilcoxon rank sum test (continuous data) at individual time points. Mixed-models analysis of variance (MMANOVA) was used to assess the overall pattern of changes over time. Missing data were not imputed, because MMANOVA does not require complete data for each participant. Because few participants had week 48 testosterone measurements available, these results are not shown, but they were considered in the MMANOVA analyses when available. MMANOVA analysis used a heterogeneous Toeplitz correlation structure between measurements within the participant. This correlation structure allows for different variances at each time point, and it does not impose a structure on how the correlation between measurements changes as the interval between the measurements increases, but a single correlation is used for each interval. For comparisons between groups, our model adjusted for treatment group and time trend and considered a time trend to be a time × treatment group interaction, as ap-

Table 1. Baseline characteristics of the study population.

Characteristic	Value
Age, median years (IQR)	37 (32–44)
Race/ethnicity, percentage of patients	
White	52.1
Black	32.4
Hispanic	14.0
Other	1.4
Injection drug use, percentage of patients	
Never	93.4
Current	0.5
Previous	6.1
Median body mass index[a] (IQR)	23.7 (21.5–26.6)
FFM, median kg (IQR)	60.5 (54.8–66.2)
CD4 cell count, median cells/mm^3 (IQR)	263 (80–428)
HIV RNA level, median $\log_{10}$ copies/mL (IQR)	5.2 (4.6–5.6)
Free testosterone level, median pg/mL (IQR)	92 (70–120)

NOTE. FFM, fat-free mass; IQR, interquartile range.

[a] Calculated as weight in kilograms divided by the square of height in meters.

propriate. Conceptually, the model with a time × treatment interaction is equivalent to fitting 2 nonparallel lines with different intercepts in an analysis of covariance. Absolute change from baseline was analyzed for testosterone level and FFM measurements. Spearman's correlation and the Wilcoxon rank sum test were used to screen continuous variables and categorical variables, respectively, as potential predictors of changes in the FFM at week 64. Stepwise backwards regression was used to construct multivariate models if only continuous variables were being considered. A similar approach was used in the generalized linear model framework when categorical variables were also included as potential predictors.

RESULTS

The baseline characteristics of the patients are shown in table 1. These variables were similar between the individual treatment groups (data not shown). Among 213 men at baseline, the median free testosterone level was 92 pg/mL (IQR, 70–120 pg/mL), and the level was subnormal (i.e., <50 pg/mL) in only 6%. Total testosterone levels (data not shown) were also subnormal (i.e., <260 ng/dL) in 6% of patients at baseline. However, results were frequently discordant: total testosterone levels were subnormal in only 5 of the 13 participants who had subnormal free testosterone levels. In multivariate analysis, lower CD4 cell count at entry, greater weight, and greater age each were independently associated with lower baseline free testosterone level, whereas race, baseline $\log_{10}$ HIV RNA level, and FFM were not.

For the group as a whole, the free testosterone level increased significantly at weeks 16, 32, and 64 (+17, +14, and +15 pg/mL, respectively; $P < .01$ for each, compared with the baseline level). The free testosterone level increased more in association with zidovudine and lamivudine (median change, +31 pg/mL; IQR, +7 to +46 pg/mL; $P < .001$ for comparison with baseline level) than with stavudine and didanosine (median change, +2 pg/mL; IQR, −21 to +32 pg/mL; $P = .8$) at week 64 ($P = .001$ for comparison between groups). Over time, there were greater increases in the free testosterone level among recipients of regimens with the zidovudine and lamivudine nucleoside backbone ($P = .01$) (figure 1*A*). Free testosterone increased level more at week 64 in efavirenz recipients (median change +30 pg/mL; IQR, −4 to +43 pg/mL) than in nelfinavir recipients (median change, −3 pg/mL; IQR, −37 to +34 pg/mL;

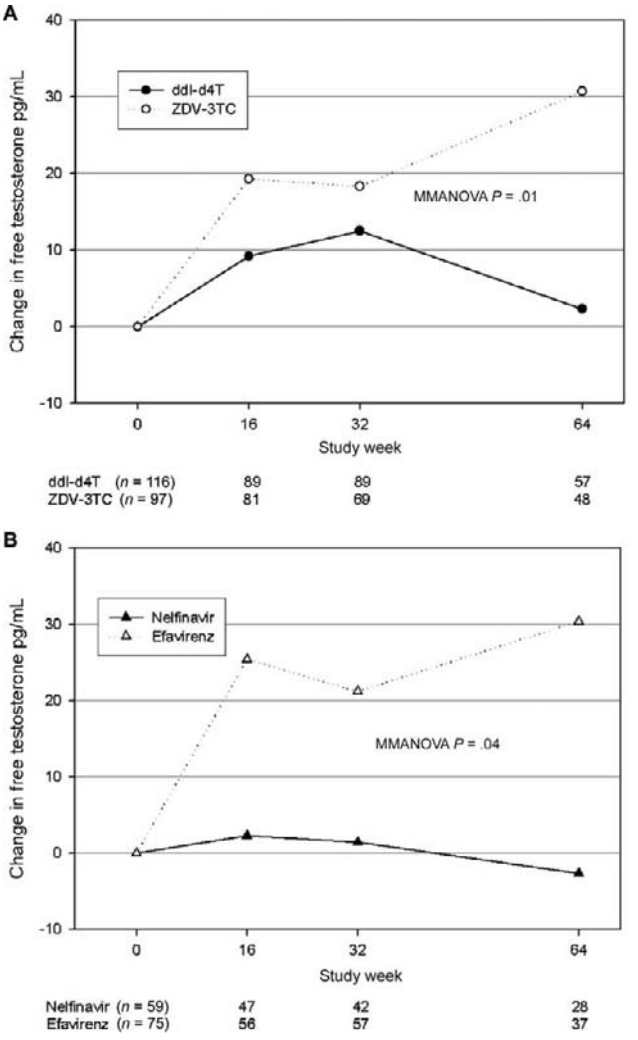

Figure 1. Median change in free testosterone concentration, by nucleoside assignment (*A; P* = .001 for the difference between groups at week 64, determined by Wilcoxon rank sum test) and by nelfinavir or efavirenz assignment (*B; P* = .05 for the difference between groups at week 64, determined by Wilcoxon rank sum test). ddI, didanosine; d4T, stavudine; MMANOVA, mixed-models analysis of variance; 3TC, lamivudine; ZDV, zidovudine.

$P = .05$), and the increases were greater over time ($P = .04$) (figure 1*B*).

The FFM increased by a median of 1.2 kg for the whole group at week 64 (median change, +2.0%; $P < .001$, by Wilcoxon signed rank test). The FFM increased significantly in association with receipt of both nucleoside backbones, but the week 64 increase was greater for zidovudine and lamivudine assignment (median change, +1.8 kg; IQR, −0.3 to +3.3 kg) than for stavudine and didanosine (median change, +0.5 kg; IQR, −1.3 to +2.6 kg; $P = .04$). Over time, there were greater increases in FFM associated with zidovudine and lamivudine ($P = .007$) (figure 2*A*). FFM increased more at week 64 with efavirenz (change, +2.1 kg; IQR, +0.3 to +4.3 kg) than with nelfinavir (change, +0.4 kg; IQR, −1.1 to +1.8; $P = .003$). Over time, there were greater increases in the FFM associated with efavirenz than with nelfinavir ($P = .003$) (figure 2*B*).

Assignment to the efavirenz arm, assignment to the zidovudine-lamivudine arm, white race, higher HIV RNA level at study entry, and lower CD4 cell count at entry were all independent predictors of increases in FFM at week 64, whereas age, weight, FFM at baseline, and baseline free testosterone level were not. In adjusted analyses, participants who received efavirenz without nelfinavir gained a mean (±SE) of 1.4 ± 0.5 kg more FFM than did participants receiving nelfinavir (with or without efavirenz; $P = .008$); those assigned to receive zidovudine and lamivudine gained 1.1 ± 0.5 kg more FFM than did those assigned to receive stavudine and didanosine ($P = .02$); white subjects gained 1.3 ± 0.5 kg more FFM than did nonwhite subjects ($P = .01$); and a lower CD4 cell count at baseline was associated with a greater gain in the FFM (0.5 ± 0.1 kg per 100 CD4 cells; $P < .001$).

There were modest correlations between change in free testosterone level at week 64 and changes both in fat level ($r = 0.33$; $P < .001$) and in FFM ($r = 0.21$; $P = .05$). The correlation tended to be greater with fat mass than with FFM, but the difference between these 2 characteristics was not statistically significant. There was also a modest correlation of intermediate magnitude between change in the free testosterone level at week 64 and change in total body weight ($r = 0.26$; $P = .007$).

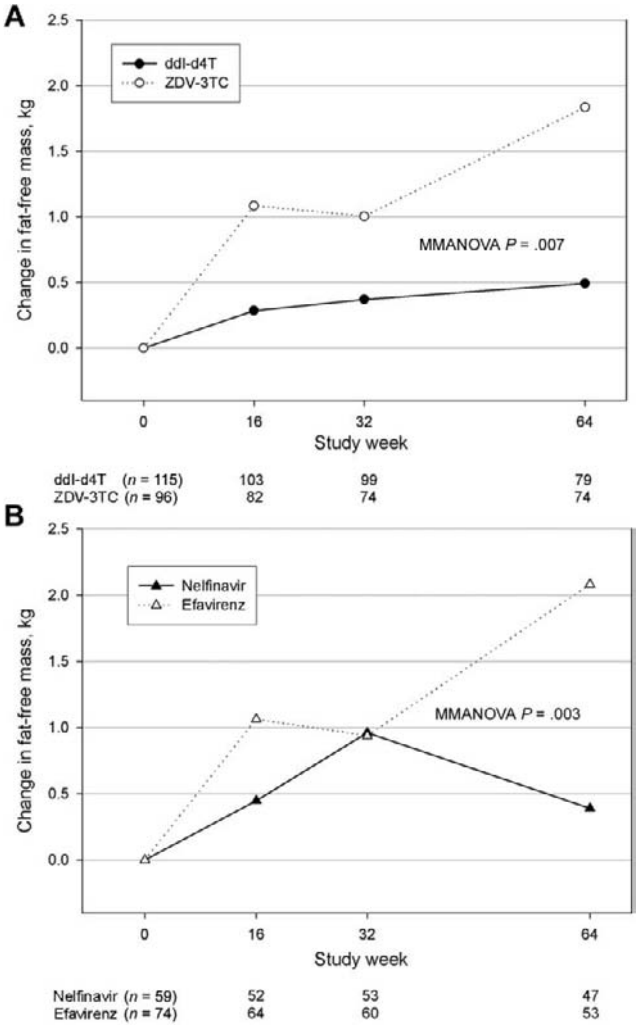

Figure 2. Median change in fat-free mass, as assessed using bioelectrical impedance analysis, by nucleoside assignment (*A; P* = .04 for the difference between groups at week 64, determined by Wilcoxon rank sum test) and by nelfinavir or efavirenz assignment (*B; P* = .003 for the difference between groups at week 64, determined by Wilcoxon rank sum test). ddI, didanosine; d4T, stavudine; MMANOVA, mixed-models analysis of variance; 3TC, lamivudine; ZDV, zidovudine.

DISCUSSION

In men who initiated potent antiretroviral therapy, the free testosterone level and FFM increased significantly after commencement of treatment. At week 64, there were significantly greater increases in subjects randomized to receive the nucleoside pair of zidovudine and lamivudine, compared with recipients of stavudine and didanosine; the same was true for patients assigned to receive efavirenz, compared nelfinavir recipients. Few of these antiretroviral-naive participants had low free or total testosterone levels at entry.

Previous studies have reported a high prevalence of hypogonadism and low testosterone levels in HIV-infected men [2], particularly in those who have experienced wasting [4, 5]. More recent data suggest that the prevalence of low testosterone levels in outpatients with stable infection, most of whom were receiving potent antiretroviral therapy, was lower [7]. Subnormal free testosterone levels occurred infrequently among our antiretroviral-naive participants, who were not selected on the basis of weight loss. Five participants were excluded from this analysis because they received androgen supplements; even if these 5 patients were considered to have low free testosterone levels, the overall prevalence would still be low (8%). Because earlier studies tended to focus on individuals with wasting and lower CD4 cell counts [2, 4, 6], we speculate that our study

population, which was not selected on the basis of weight loss or a clinical diagnosis of wasting and which had higher CD4 cell counts, may be more representative of antiretroviral-naive men in the era of potent antiretroviral therapy. Importantly, free testosterone and total testosterone levels were frequently discordant. Our data support obtainment of free testosterone levels when hypogonadism is suspected in patients with conditions such as HIV infection, for which sex hormone–binding globulin levels are commonly elevated, as has been suggested elsewhere [16]. It is also important to note that determination of a single testosterone level at each time point alone, as was done in our study, is not sufficient to diagnose hypogonadism [16]. Although it is difficult to attribute causality, lower entry CD4 cell count, greater weight, and greater age—but not race, HIV RNA level, or FFM at the time of study entry—independently predicted lower baseline free testosterone levels.

In the group as a whole, free testosterone levels increased significantly after initiating antiretroviral therapy. A cross-sectional study found that, compared with no antiretroviral therapy, total testosterone levels were greater in patients who were receiving NNRTI or protease inhibitor–based antiretroviral therapy [7]. In addition, levels were significantly higher among protease inhibitor–treated participants, but the specific NNRTI and protease inhibitors used were not reported. Because the metabolic effects of different NNRTIs and protease inhibitors can vary widely within these classes of drugs, our results must be considered specific to the particular agents studied and not necessarily a class effect.

We found that greater increases in free testosterone level and FFM occurred in persons assigned to receive zidovudine and lamivudine, compared with those assigned to receive stavudine and didanosine. The reasons for these findings are unclear, but we speculate that mitochondrial dysfunction may play a role. In a previous report of regional body fat from this study, receipt of stavudine and didanosine was associated with significantly greater limb fat loss—a toxicity associated with mitochondrial dysfunction—than was receipt of zidovudine and lamivudine [13]. Participants with lipoatrophy have tended to reduce muscle mitochondrial DNA content as well [17, 18], suggesting a link between lipoatrophy and skeletal muscle toxicity that potentially results in lesser gains in FFM. Although it has not been studied, it is possible that testicular mitochondrial function may also be impaired by stavudine and didanosine use, resulting in a more modest increase in free testosterone level after the initiation of therapy. However, there was no association between a lower total testosterone level and stavudine use, compared with zidovudine use, in a cross-sectional study in which stavudine recipients experienced more cases of lipoatrophy [8].

Greater increases in free testosterone level and FFM also occurred in persons assigned to receive efavirenz, compared with nelfinavir recipients. Similarly, receipt of nelfinavir was associated with significantly greater limb fat loss than was receipt of efavirenz [13]. In the Australian lipodystrophy survey, lower total testosterone levels were associated with a greater degree of lipoatrophy [19]. We speculate that subcutaneous fat loss may lead to lesser increases in the testosterone level, both for nelfinavir and for stavudine plus didanosine.

White race, higher HIV RNA level at study entry, lower CD4 cell count at study entry, assignment to the efavirenz arm, and assignment to the zidovudine-lamivudine arm were all independent predictors of increases in FFM at week 64. The association between markers of disease severity (CD4 cell count and HIV RNA level) and greater increases in FFM during potent antiretroviral therapy has been reported previously [10] and presumably represents a return-to-health phenomenon in persons with more advanced HIV disease.

This study was limited by the inclusion of only men. Too few women were enrolled in the parent study to provide meaningful results for that subgroup, for which results may have been different. We used the "gold standard" method of equilibrium dialysis for measuring free testosterone level [20] for our primary analyses, but this technique is technically demanding and expensive and values obtained in different laboratories may not be comparable [20]. Although they are, in general, technically less demanding, total testosterone assays are not standardized across platforms and often overestimate testosterone concentrations; similarly, values obtained in different laboratories may not be comparable [20]. Another limitation is that use of bioelectrical impedance analysis may be inferior to dual x-ray absorptiometry for estimating lean body mass in participants with HIV infection [21]. However, we attempted to optimize the results obtained with bioelectrical impedance analysis, including standardized measurements of height and weight, which are major sources of variability in FFM measured by this method, and we used central training in electrode placement for these longitudinal assessments. It is also possible that our calculated values for FFM may have been affected by treatment-induced alterations in regional body fat associated with lipoatrophy in our participants.

We conclude that, in this antiretroviral-naive cohort, subnormal levels of free testosterone are relatively uncommon among men at the time that antiretroviral therapy is initiated. Increases in free testosterone level and FFM occur after initiation of potent antiretroviral therapy, but the magnitude of these increases may vary with different antiretroviral drug regimens. Studies of the effects of different antiretroviral agents on lean body mass are needed to help identify the most optimal regimens.

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References

1. Dobs A. Endocrine disorders in men infected with human immunodeficiency virus. Am J Med **1988**; 84:611–6.
2. Arver S, Sinha-Hikim I, Beall G, Guerrero M, Shen R, Bhasin S. Serum dihydrotestosterone and testosterone concentrations in human immunodeficiency virus-infected men with and without weight loss. J Androl **1999**; 20:611–8.
3. Laudat A, Blum L, Guechot J, et al. Changes in systemic gonadal and adrenal steroids in asymptomatic human immunodeficiency virus-infected men: relationship with the CD4 cell counts. Eur J Endocrinol **1995**; 133:418–24.
4. Grinspoon S, Corcoran C, Lee K, et al. Loss of lean body mass and muscle mass correlates with androgen levels in hypogonadal men with acquired immunodeficiency syndrome and wasting. J Clin Endocrinol Metab **1996**; 81:4051–8.
5. Rietschel P, Corcoran C, Stanley T, Basgoz N, Klibanski A, Grinspoon S. Prevalence of hypogonadism among men with weight loss related to human immunodeficiency virus infection who were receiving highly active antiretroviral therapy. Clin Infect Dis **2000**; 31:1240–4.
6. Dobs AS, Few WL 3rd, Blackman MR, Harman SM, Hoover DR, Graham NM. Serum hormones in men with human immunodeficiency virus-associated wasting. J Clin Endocrinol Metab **1996**; 81:4108–12.
7. Collazos J, Martinez E, Mayo J, Ibarra S. Sexual hormones in HIV-infected patients: the influence of antiretroviral therapy. AIDS **2002**; 16:934–7.
8. Domingo P, Sambeat MA, Perez A, Ordonez J, Rodriguez J, Vazquez G. Fat distribution and metabolic abnormalities in HIV-infected patients on first combination antiretroviral therapy including stavudine or zidovudine: role of physical activity as a protective factor. Antivir Ther **2003**; 8:223–31.
9. Martin ME, Benassayag C, Amiel C, Canton P, Nunez EA. Alterations in the concentrations and binding properties of sex steroid binding protein and corticosteroid-binding globulin in HIV+ patients. J Endocrinol Invest **1992**; 15:597–603.
10. Shikuma CM, Zackin R, Sattler F, et al. Changes in weight and lean body mass during highly active antiretroviral therapy. Clin Infect Dis **2004**; 39:1223–30.
11. Robbins GK, De Gruttola V, Shafer RW, et al. Comparison of sequential three-drug regimens as initial therapy for HIV-1 infection. N Engl J Med **2003**; 349:2293–303.
12. Shafer RW, Smeaton LM, Robbins GK, et al. Comparison of four-drug regimens and pairs of sequential three-drug regimens as initial therapy for HIV-1 infection. N Engl J Med **2003**; 349:2304–15.
13. Dubé MP, Parker RA, Tebas P, et al. Glucose metabolism, lipid, and

body fat changes in antiretroviral-naive subjects randomized to nelfinavir or efavirenz plus dual nucleosides. AIDS **2005**; 19:1807–18.
14. Kotler DP, Burastero S, Wang J, Pierson RN Jr. Prediction of body cell mass, fat-free mass, and total body water with bioelectrical impedance analysis: effects of race, sex, and disease. Am J Clin Nutr **1996**; 64: 489S–97S.
15. Vermeulen A, Stoica T, Verdonck L. The apparent free testosterone concentration, an index of androgenicity. J Clin Endocrinol Metab **1971**; 33:759–67.
16. Bhasin S, Cunningham GR, Hayes FJ, et al. Testosterone therapy in adult men with androgen deficiency syndromes: an endocrine society clinical practice guideline. J Clin Endocrinol Metab **2006**; 91: 1995–**2010**.
17. Zaera MG, Miro O, Pedrol E, et al. Mitochondrial involvement in antiretroviral therapy-related lipodystrophy. AIDS **2001**; 15:1643–51.
18. McComsey GA, Paulsen DM, Lonergan JT, et al. Improvements in lipoatrophy, mitochondrial DNA levels and fat apoptosis after replacing stavudine with abacavir or zidovudine. AIDS **2005**; 19:15–23.
19. Miller J, Carr A, Emery S, et al. HIV lipodystrophy: prevalence, severity and correlates of risk in Australia. HIV Med **2003**; 4:293–301.
20. Rosner W, Auchus RJ, Azziz R, Sluss PM, Raff H. Position statement: utility, limitations, and pitfalls in measuring testosterone. An Endocrine Society position statement. J Clin Endocrinol Metab **2007**; 92:405–13.
21. Corcoran C, Anderson EJ, Burrows B, et al. Comparison of total body potassium with other techniques for measuring lean body mass in men and women with AIDS wasting. Am J Clin Nutr **2000**; 72:1053–8.