ANDROSTENEDIONE IS A STEROID HORMONE produced in the gonads and adrenal glands of both sexes. It is synthesized from dehydroepiandrosterone and then converted to testosterone by the enzyme 17β-hydroxysteroid dehydrogenase or to estrone by the aromatase enzyme complex.1-3 It is currently available without prescription and marketed primarily to athletes and bodybuilders. The number of people regularly using androstenedione is not known. It has been estimated that 4.9% of male and 2.4% of female adolescents in the United States have used illegal androgenic/anabolic steroids.4 Because androstenedione is readily available as a dietary supplement, its use may be even greater.

Unsubstantiated claims have been made that orally administered androstenedione increases testosterone levels and has anabolic effects in men. To determine whether androstenedione increases serum testosterone concentrations, we administered 100 or 300 mg/d for 7 days (vs controls) to young, healthy men and made detailed measurements of serum sex steroid hormone concentrations.

Context Androstenedione, a steroid hormone and the major precursor to testosterone, is available without prescription and is purported to increase strength and athletic performance. The hormonal effects of androstenedione, however, are unknown.

Objective To determine if oral administration of androstenedione increases serum testosterone levels in healthy men.

Design Open-label randomized controlled trial conducted between October 1998 and April 1999.

Setting General clinical research center of a tertiary-care, university-affiliated hospital.

Participants Forty-two healthy men aged 20 to 40 years.

Intervention Subjects were randomized to receive oral androstenedione (either 100 mg/d [n = 15] or 300 mg/d [n = 14]) or no androstenedione (n = 13) for 7 days.

Main Outcome Measures Changes in serum testosterone, androstenedione, estrone, and estradiol levels, measured by frequent blood sampling, compared among the 3 treatment groups.

Results Mean (SE) changes in the area under the curve (AUC) for serum testosterone concentrations were −2% (7%), −4% (4%), and 34% (14%) in the groups receiving 0, 100, and 300 mg/d of androstenedione, respectively. When compared with the control group, the change in testosterone AUC was significant for the 300-mg/d group (P <.001) but not for the 100-mg/d group (P = .48). Baseline testosterone levels, drawn 24 hours after androstenedione administration, did not change. Mean (SE) changes in the AUC for serum estradiol concentrations were 4% (6%), 42% (12%), and 128% (24%) in the groups receiving 0, 100, and 300 mg/d of androstenedione, respectively. When compared with the control group, the change in the estradiol AUC was significant for both the 300-mg/d (P <.001) and 100-mg/d (P = .002) groups.

There was marked variability in individual responses for all measured sex steroids.

Conclusions Our data suggest that oral androstenedione, when given in dosages of 300 mg/d, increases serum testosterone and estradiol concentrations in some healthy men.

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ing. All subjects had normal liver function, renal function, and serum testosterone concentrations. The study was approved by the human studies committee at Massachusetts General Hospital. All subjects gave written informed consent.

Subjects were randomly assigned to 1 of 3 groups: no androstenedione (n = 13) or 100 (n = 15) or 300 mg/d (n = 14) of androstenedione (Sports One, Klein Laboratories, Wallingford, Conn) for 7 days. Androstenedione capsules were dispensed at the general clinical research center at the same time each day. Subjects were instructed to ingest nothing by mouth except water for 1 hour after androstenedione administration.

Serum androstenedione, testosterone, estrone, and estradiol concentrations were measured at 0, 15, 30, 45, 60, 90, 120, 180, 240, 360, and 480 minutes after administration on days 1 and 7. On days 2 to 6, hormone concentrations were measured just prior to administration. Baseline serum luteinizing hormone, follicle-stimulating hormone, liver function, creatinine, and total cholesterol concentrations were measured each day. Hematocrit and serum sex hormone–binding globulin concentrations were measured on days 1 and 7.

Analysis of Androstenedione

The amount of androstenedione in 13 different capsules, each of which was purported to contain 100 mg of androstenedione, was determined by comparison to a 100-mg reference standard of Δ⁴-androstene-3,17-dione (Lot 18-C0226; Sigma Chemical Co, St Louis, Mo) by means of high-performance liquid chromatography equipped with a diode array detector. The mean amount of androstenedione was 99.8 mg (range, 83.9-113.9 mg; coefficient of variation, 8.7%). All capsules contained androstenedione as determined by high-performance liquid chromatography and by liquid chromatography–tandem mass spectrometry. The mass spectrum of the androstenedione peak was identical to that of the reference standard. All other peaks were less than 1% of the androstenedi-

Table 1. Baseline Clinical and Laboratory Characteristics of Study Subjects

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>0 (n = 13)</th>
<th>100 (n = 15)</th>
<th>300 (n = 14)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>27.8 (4.0)</td>
<td>26.3 (5.2)</td>
<td>31.5 (6.6)</td>
<td>.07</td>
</tr>
<tr>
<td>Height, cm</td>
<td>177.3 (5.6)</td>
<td>178.5 (8.4)</td>
<td>182.1 (6.6)</td>
<td>.25</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>84.6 (10.3)</td>
<td>76.7 (8.4)</td>
<td>78.6 (7.8)</td>
<td>.11</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>26.9 (2.5)</td>
<td>24.2 (3.4)</td>
<td>23.8 (3.4)</td>
<td>.005</td>
</tr>
<tr>
<td>Androstenedione, ng/dL†</td>
<td>167 (77)</td>
<td>184 (83)</td>
<td>145 (67)</td>
<td>.42</td>
</tr>
<tr>
<td>Estrone, pg/mL‡</td>
<td>41 (19)</td>
<td>44 (21)</td>
<td>31 (11)</td>
<td>.15</td>
</tr>
<tr>
<td>Estradiol, pg/mL‡</td>
<td>31 (13)</td>
<td>41 (21)</td>
<td>29 (9)</td>
<td>.10</td>
</tr>
<tr>
<td>Testosterone, ng/dL†</td>
<td>500 (237)</td>
<td>541 (192)</td>
<td>493 (180)</td>
<td>.66</td>
</tr>
</tbody>
</table>

*Data are presented as mean (SD) unless otherwise indicated.
†To convert values for androstenedione and testosterone to nmol/L, multiply by 0.0349 and 0.0347, respectively.
‡To convert values for estrone and estradiol to pmol/L, multiply by 37 and 3.67, respectively.

Figure 1. Mean Serum Testosterone and Estradiol Concentrations, Day 7

Figure 2. Mean Percentage Change in Area Under the Curve for Serum Testosterone and Estradiol Concentrations, Days 1 and 7

Values are presented as the averages for days 1 and 7. Asterisk indicates P value for comparison vs control and 100-mg/d groups. Dagger indicates P value for comparison vs control group. Error bars represent SE.
one peak and none were testosterone, 19-norandrostenedione, dehydroepiandrosterone, or 19-norandrostosterone.

### Serum Measurements
Serum testosterone, estradiol, estrone, and androstenedione concentrations were measured by radioimmunoassay. Serum luteinizing hormone, follicle-stimulating hormone, and sex hormone–binding globulin concentrations were measured using chemiluminescent immunometric assays. The cross-reactivity of androstenedione in the testosterone assay was 0.5%. All samples for an individual subject were analyzed in the same assay.

### Statistical Analysis
The primary end point was area under the curve (AUC) on days 1 and 7 for each steroid hormone, expressed in units of concentration-hours. The mean values of AUC for each hormone were compared after log transformation by means of repeated measures analysis of covariance, adjusting for baseline level of the hormone and study day. Changes in serum luteinizing hormone, follicle-stimulating hormone, cholesterol, and daily baseline sex-steroid hormone concentrations were compared using a mixed-model analysis of covariance. Baseline clinical characteristics were compared using the Kruskal-Wallis test.

### RESULTS
There were no differences in most of the baseline clinical characteristics between study groups, except for a statistically significant difference in body mass index (Table 1).

In the control group, serum androstenedione, testosterone, estrone, and estradiol levels were stable during frequent blood sampling (testosterone and estradiol only shown in Figure 1). The mean (SE) changes in AUC for serum testosterone concentrations during the frequent blood sampling period were −2% (7%), −4 (4%), and 34% (14%) in the groups receiving 0, 100, and 300 mg/d of androstenedione, respectively (Figure 2). For the 300-mg/d group, the change in testosterone AUC was significant when compared with both the control (P < .001) and 100-mg/d (P < .001) groups. There was no difference between the control and 100-mg/d groups (P = .48). In the 100-mg/d group, the mean AUC for serum androstenedione, estrone, and estradiol concentrations increased 72% (18%), 74% (20%), and 42% (12%), respectively (P < .001 for androstenedione and estrone, and P = .002 for estradiol vs control). In the 300-mg/d group, the mean AUC for serum androstenedione, estrone, and estradiol concentrations increased 697% (136%), 196% (28%), and 128% (24%), respectively (P < .001 vs control for each comparison). Increases in mean serum androstenedione (P < .001), estrone (P = .002), and estradiol (P = .001) concentrations were greater in the subjects who received 300 mg/d of androstenedione vs the 100-mg/d group.

Mean baseline and peak serum testosterone levels on days 1 and 7 are shown in Table 2. There was considerable individual variability in the changes in serum sex steroid levels (Figure 3). In the men who received 100 mg/d of androstenedione, no subjects had serum testosterone concentrations that exceeded the upper limit of normal (1000 ng/dL [34.7 nmol/L]), whereas 4 subjects who received 300 mg/d of androstenedione had serum testosterone concentrations above the normal range. Twelve of 15 subjects in the 100-mg/d group and 10 of 14 subjects in the 300-mg/d group had estradiol levels above the upper limit of normal for men (50 pg/mL [184 pmol/L]).

Mean daily baseline serum androstenedione and estradiol concentrations increased significantly during the 7-day period in the 300-mg/d group (P = .01 for androstenedione and P = .003 for estradiol). Mean daily baseline serum testosterone, estrone, follicle-stimulating hormone, and luteinizing hormone concentrations did not change. Serum sex-hormone–binding globulin concentrations decreased in both treated groups (data not shown).

No subjects reported adverse effects. There were no changes in liver function tests or serum creatinine, serum total cholesterol, or hematocrit levels.

### COMMENT
This study shows that oral androstenedione administration increases serum testosterone, androstenedione, estradiol, and estrone concentrations in healthy men. Few studies have examined the effects of oral androstenedione administration on testosterone production in humans. A 100-mg dose of androstenedione increased testosterone concentrations in 2 women. In a recent study, in which an-
dione to testosterone, 17b-hydroxysteroid dehydrogenase, and aromatase, the enzyme complex that converts steroid dehydrogenase, and aromatase, thus, it is possible to increase testosterone and estrogen concentrations in healthy men. However, our data also demonstrate that a higher dose does increase serum testosterone concentrations.

The enzyme that converts androstenedione to testosterone, 17b-hydroxysteroid dehydrogenase, and aromatase, the enzyme complex that converts androstenedione and testosterone to estrogen and estradiol, is expressed in many human tissues including skeletal muscle and fat. Thus, it is possible that increases in local tissue levels of testosterone, estrone, or estradiol are even greater than the increases in their circulating concentrations.

That oral androstenedione administration increases serum testosterone levels suggests that it could have androgenic or anabolic effects. High doses of testosterone increase muscle size and strength in healthy men. It is unclear if smaller increases in serum testosterone allow one to have anabolic effects. Muscle size and strength did not change when 100 mg of androstenedione was administered 3 times daily to healthy men without prior weight-lifting experience. As this dose was not sufficient to raise testosterone levels, it remains unknown if doses of androstenedione that increase testosterone levels would have significant effects on muscle size and function. Finally, because androstenedione itself is a weakly androgenic steroid, increases in androstenedione itself could have anabolic effects.

While testosterone levels increased in the subjects receiving the 300-mg/d doses, muscle and fat gain also increased in subjects receiving the 300-mg/d dose. Our data suggest that orally administered androstenedione increases serum testosterone and estrogen levels in healthy men, particularly at higher doses. These increases could lead to anabolic or untoward effects in susceptible populations. Long-term studies of androstenedione use are needed.

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**REFERENCES**

1. Horton R, Tate JF. Androstenedione production and interconversion rates measured in peripheral blood and studies on the possible site of conversion to testoster-


4. Yesalis CE, Barukiewicz CK, Kopstein AN, Bahre MS. Trends in anabolic-androgenic steroid use among ado-


10. Simpson ER, Zhao Y, Agarwal VR, et al. Aro-


16. Anderson RA, Wallace EM, Wu FC. Effect of testoste-

17. Prader A, Zachmann M. Treatment of excess-


19. Forney JP, Milewich L, Chen GT, et al. Aromati-

20. Simpson ER, Zhao Y, Agarwal VR, et al. Aro-


24. Anderson RA, Wallace EM, Wu FC. Effect of testoste-

25. Prader A, Zachmann M. Treatment of excess-


27. Forney JP, Milewich L, Chen GT, et al. Aromati-

28. Simpson ER, Zhao Y, Agarwal VR, et al. Aro-


32. Anderson RA, Wallace EM, Wu FC. Effect of testoste-

33. Prader A, Zachmann M. Treatment of excess-