Effect of Oral Androstenedione on Serum Testosterone and Adaptations to Resistance Training in Young Men

A Randomized Controlled Trial

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Context Androstenedione, a precursor to testosterone, is marketed to increase blood testosterone concentrations as a natural alternative to anabolic steroid use. However, whether androstenedione actually increases blood testosterone levels or produces anabolic androgenic effects is not known.

Objectives To determine if short- and long-term oral androstenedione supplementation in men increases serum testosterone levels and skeletal muscle fiber size and strength and to examine its effect on blood lipids and markers of liver function.

Design and Setting Eight-week randomized controlled trial conducted between February and June 1998.

Participants Thirty healthy, normotestosterogenic men (aged 19-29 years) not taking any nutritional supplements or androgenic-anabolic steroids or engaged in resistance training.

Interventions Twenty subjects performed 8 weeks of whole-body resistance training. During weeks 1, 2, 4, 5, 7, and 8, the men were randomized to either androstenedione, 300 mg/d (n = 10), or placebo (n = 10). The effect of a single 100-mg androstenedione dose on serum testosterone and estrogen concentrations was determined in 10 men.

Main Outcome Measures Changes in serum testosterone and estrogen concentrations, muscle strength, muscle fiber cross-sectional area, body composition, blood lipids, and liver transaminase activities based on assessments before and after short- and long-term androstenedione administration.

Results Serum free and total testosterone concentrations were not affected by short- or long-term androstenedione administration. Serum estradiol concentration (mean [SEM]) was higher ($P<.05$) in the androstenedione group after 2 (310 [20] pmol/L), 5 (300 [30] pmol/L), and 8 (280 [20] pmol/L) weeks compared with presupplementation values (220 [20] pmol/L). The serum estrone concentration was significantly higher ($P<.05$) after 2 (153 [12] pmol/L) and 5 (142 [15] pmol/L) weeks of androstenedione supplementation compared with baseline (106 [11] pmol/L). Knee extension strength increased significantly ($P<.05$) and similarly in the placebo (770 [55] N vs 1095 [52] N) and androstenedione (717 [46] N vs 1024 [57] N) groups. The increase of the mean cross-sectional area of type 2 muscle fibers was also similar in androstenedione (4703 [471] vs 5307 [604] mm^2; $P<.05$) and placebo (5271 [485] vs 5728 [451] mm^2; $P<.05$) groups. The significant ($P<.05$) increases in lean body mass and decreases in fat mass were also not different in the androstenedione and placebo groups. In the androstenedione group, the serum high-density lipoprotein cholesterol concentration was reduced after 2 weeks (1.09 [0.08] mmol/L [42 (3) mg/dL] vs 0.96 [0.08] mmol/L [37 (3) mg/dL]; $P<.05$) and remained low after 5 and 8 weeks of training and supplementation.

Conclusions Androstenedione supplementation does not increase serum testosterone concentrations or enhance skeletal muscle adaptations to resistance training in normotestosterogenic young men and may result in adverse health consequences.

JAMA. 1999;281:2020-2028 www.jama.com

Androgenic-anabolic steroids have been shown to enhance the gains in muscle size and strength associated with resistance training.[1-4] Androstenedione, a precursor to testosterone, is normally produced by the adrenal gland and gonads and is converted to testosterone through the action of 17β-hydroxysteroid dehydrogenase, which is found in most body tissues.[5-9] Androstenedione is also produced by some plants and has recently been marketed as a product for increasing blood testosterone concentrations to be used as a natural alternative to anabolic steroid use.

However, the interconversions of androstenedione and testosterone to other androgens, as well as to estrogens, are complex. In addition to serving as a precursor to testosterone, androstenedione may be converted into estrogens directly.[10,11] Since testosterone is also aromatized to estradiol,[11,12] it is also possible that increased production of testosterone following androstenedione administration may also result in increased aromatization, which would further attenuate any increase in the blood testosterone concentration. These considerations raise the question of

For editorial comment see p 2043.

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whether androstenedione supplementation increases the blood testosterone concentration and produces anabolic-androgenic effects.

To date only one study has investigated the effect of oral androstenedione administration on the blood testosterone concentration.[13] These authors observed 4- and 7-fold increases in the blood testosterone concentration in 2 healthy women, respectively, after the ingestion of a single dose of 100 mg of androstenedione. The effect of androstenedione administration on blood testosterone levels in healthy men is unknown. Therefore, one purpose of this study was to determine whether short- and long-term administration of oral androstenedione increases the blood testosterone concentration and enhances gains in muscle size and strength when combined with a resistance-training program.

Increased concentrations of testosterone in the blood have been associated with an increased risk of cardiovascular disease, due both to a lowering of the serum high-density lipoprotein cholesterol (HDL-C) concentration and an increased serum concentration of low-density lipoprotein (LDL) concentration.[3,14-19] Elevated blood testosterone concentrations may also result in significant alterations in liver function.[20,21] The effects on blood lipids and liver function appear to be more pronounced in oral anabolic steroids, compared with injectable agents. A second purpose of this study, therefore, was to examine the effect of androstenedione administration on blood lipids and on clinical markers of liver function.

METHODS

Subjects

A total of 30 healthy, normotestosterogenic young (aged 19-29 years) men were recruited for this experiment, approved by the Iowa State University Human Subjects Committee. These participants were screened to ensure that they were not consuming androstenedione or any other nutritional supplement prior to enrollment in the study and were not currently engaged in a resistance-training program. Subjects were also not taking illicit drugs or abusing alcohol consumption. All subjects were free of any cardiovascular or orthopedic condition that would contraindicate exercise testing or training.

Short-term Administration of Androstenedione

The effect of short-term administration of androstenedione on the serum concentration of androstenedione, free and total testosterone, luteinizing hormone (LH), and follicle-stimulating hormone (FSH) was studied in 10 of the men (mean age [SEM], 23 [4] years). On 2 separate days after an overnight fast, separated by 1 week, subjects ingested 100 mg of androstenedione or placebo (250 mg of rice flour), administered in a randomly assigned double-blind manner. This dose was chosen based on the previous report that 100 mg of androstenedione increases blood testosterone concentration by 4- to 7-fold in women.[13] Blood samples were obtained before and every 30 minutes after ingestion for 6 hours. Serum hormone concentrations were determined as described below.

Androstenedione Supplementation During Resistance Training

After screening, 20 of the men were randomly assigned in a double-blind manner to groups that consumed either androstenedione or placebo during weeks 1-2, 4-5, and 7-8, during the 8 weeks of resistance training. One subject in each group reported prior resistance-training experience, although none had performed resistance training during the preceding year. Supplementation was administered in a cyclic fashion as recommended by the manufacturer to simulate the supplementation regimen followed by many athletes. This cycle is believed by athletes to allow for a "washout" period and reduce the likelihood of adverse effects due to anabolic steroid administration. Subjects consumed 300 mg of androstenedione or a placebo (250 mg of rice flour) in capsule form each day. The 300-mg/d dosage was chosen to exceed the maximal dosage typically recommended by manufacturers (100-300 mg/d), as well as the dosage shown to increase blood testosterone concentrations in women.[13] Supplements were taken in unmarked white capsules in 3 equal doses before 9 AM, at 3 PM, and at bedtime. The androstenedione was derived from wild yams and was provided by Experimental and Applied Sciences Inc (Golden, Colo). Purity of the androstenedione contained in the capsules was assessed with high-performance liquid chromatography by 2 independent laboratories (Biomedical Laboratories Inc, Petaluma, Calif, and Integrated Biomolecule Corp, Tucson, Ariz). These analyses produced values for purity of 99% and 100%, respectively. To encourage compliance, subjects maintained a record of supplement ingestion and were required to return unused supplements at the completion of the study. At the conclusion of the study, when subjects were asked to identify which supplement they were taking, 2 subjects in the placebo group correctly identified the supplement they were taking.

Resistance Training

During the 8-week resistance-training program, subjects performed resistance training 3 days per week on nonconsecutive days. Subjects were instructed on proper lifting technique and supervised by 1 of the investigators (G.A.B., T.A.R., N.L.U., or K.A.P.) during all lifting sessions. The resistance-training program was designed to increase the strength of all major muscle groups. Subjects trained on bench press, shoulder press, knee extension, right and left knee flexion, vertical butterfly, leg press, calf press, biceps curl, triceps extension, and lattisimus dorsi pull-down. Subjects performed 3 sets of 10 repetitions for the first 2 weeks. For the final 6 weeks of training, subjects performed 3 sets of 8 repetitions. Resistance was set at 80% to 85% of 1 repetition maximum (1-RM). Following the determination of 1-RM after 4 weeks of training, the training intensity was adjusted to 80% to 85% of the

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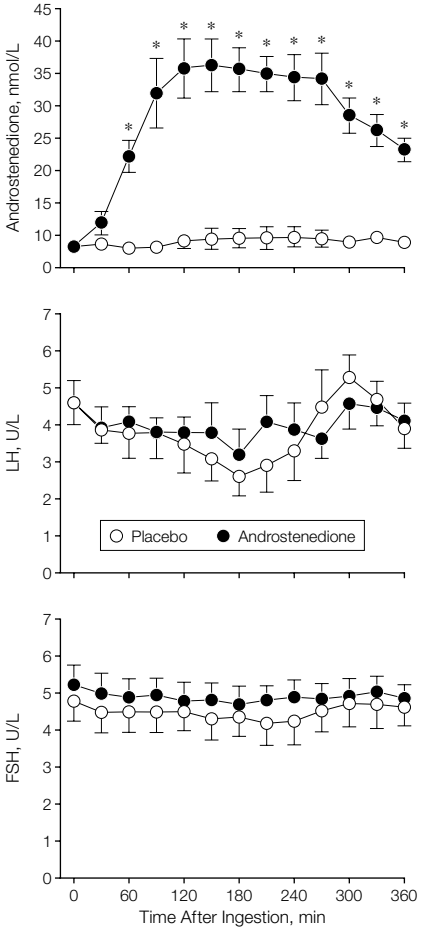
new 1-RM. All resistance training and 1-RM testing was performed on multistation isotonic resistance equipment (FTX; Paramount Fitness Equipment, Los Angeles, Calif).

Strength Testing

Muscle strength was assessed with the measurement of 1-RM before and after 4 and 8 weeks of resistance training. After a brief warm-up, subjects were encouraged to meet their 1-RM within 5 trials of progressing resistance. One repetition maximum was assessed on bench press, shoulder press, knee extension, right and left knee flexion, biceps curl, triceps extension, lattisimus dorsi pull-down, and vertical butterfly.

Figure 1. Serum Androstenedione, Luteinizing Hormone (LH), and Follicle-Stimulating Hormone (FSH) Concentrations With 100 mg of Androstenedione or Placebo



Data are mean (SEM) for n = 10. Asterisk indicates significantly different from time zero for androstenedione ($P<.05$).

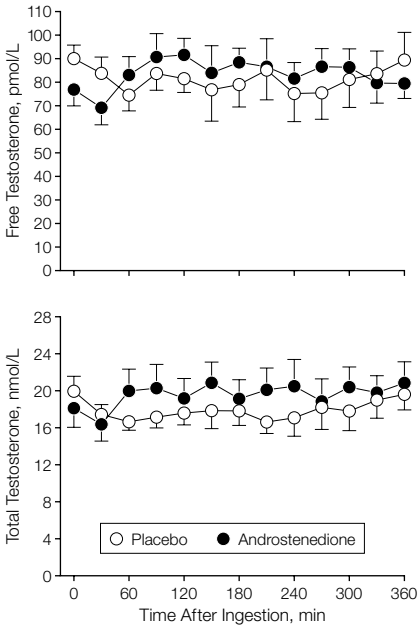
Body Composition

Body mass and circumference measures were obtained before training and after 4 and 8 weeks of training. All circumference measurements were performed by the same investigator (T.A.R) and were obtained for the following sites: biceps, shoulder, chest, abdomen, waist, hips, gluteal, thigh, and calf. Body density and percent body fat were determined with hydrostatic weighing before and after 8 weeks of training using a computer-interfaced load cell and custom computer program. Body fat percent was calculated using the Siri equation[22] after estimation of the residual volume.[23]

Dietary Analysis

To assess diet, subjects kept a food-intake record for 3 days prior to beginning resistance training and supplementation. Subjects were instructed to maintain their typical dietary intake during the course of the study. Diet records were analyzed for composition using a food analysis software package (Food Comp; Iowa State University, Ames). Mean (SEM) daily energy intake was not different in placebo (9983 [214] kJ/d) and androstenedione (9660 [198] kJ/d) groups prior to supplementation and resistance training. Daily protein intake was also not different in placebo (83 [5] g/d) and androstenedione (98 [4] g/d) groups and exceeded the recommended daily allowance for all subjects, suggesting adequate nitrogen balance. Although it was not possible to directly assess diet compliance during the study, subjects were queried at the end of training, and all indicated that their diet did not change during the 8 weeks.

Figure 2. Serum Free and Total Testosterone Concentrations With 100 mg of Androstenedione or Placebo



Data are mean (SEM) for n = 10.

Clinical Blood Chemistry and Hormonal Analyses

Blood samples were obtained after an overnight fast for a standard blood chemistry and hormonal analyses before training and after 2, 5, and 8 weeks of training. Blood samples were drawn without stasis from a catheter inserted into an antecubital vein. Clinical blood chemistry analyses were performed by a commercial laboratory (Labcorp Inc, Kansas City, Mo). Another sample was centrifuged and serum was frozen at −80°C until analysis. Serum concentrations of free and total testosterone, androstenedione, LH, FSH, estradiol, estrone, and estriol were measured with radioimmunoassay using commercially available kits (Diagnostic Products, Los Angeles, Calif, and Diagnostic Systems Laboratories Inc, Webster, Tex). All samples for each subject were assayed in the same run. The intra-assay coefficients of variation were 7.3%, 7.7%, 6.7%, 4.7%, 3.9%, 6.0%, 8.2%, and 7.3% for free testosterone, total testosterone, androstenedione, LH, FSH, estradiol, estrone, and estriol, respectively.

Muscle Histochemistry

Muscle samples (about 100 mg) were obtained from the lateral aspect of the vastus lateralis muscle using the needle

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biopsy technique described by Bergstrom.[24] Muscle specimens were placed in mounting medium and immediately frozen in isopentane cooled to the temperature of liquid nitrogen for later sectioning and staining. Frozen transverse sections (about 10 µm) were cut on a cryostat (Histostat Microtome; AO Scientific Instruments, Buffalo, NY) at –20°C and mounted on cover glasses. Muscle sections were stained for adenosine triphosphatase activity at pH 9.4 after a preincubation at pH 4.3. Samples were then counterstained with eosin Y (Sigma-Aldrich, St Louis, Mo) for color enhancement to aid in image analysis. Muscle-fiber type distribution and muscle-fiber areas were determined using a computer-operated image analysis system (Neosis Visilog Image Analysis Software; SGI-Computer; Sony DXC-3000A-Camera). The system captures the light microscope image, traces the muscle-fiber boundaries, counts the light and dark muscle fibers, and measures the cross-sectional areas. For muscle-fiber type–distribution, all type 1 and type 2 muscle fibers were counted. When the data from the placebo and androstenedione groups before and after training are combined, fiber–type distribution was determined on 337 fibers.[25] For determination of mean cross-sectional area of type 1 and type 2 fibers, groupings of clearly delineated fibers were highlighted, and 20 fibers of each type were randomly selected by a technician blinded to the treatments.

Statistical Analyses

Data were analyzed using commercial software (SPSS Inc, Chicago, Ill). Statistical analyses were performed using 2-factor (time and treatment) analyses of variance (ANOVA) with repeated measures. When ANOVA revealed a significant interaction ($P<.05$), specific mean differences were assessed with *t* tests, using the Bonferroni α correction for multiple comparisons.

RESULTS

One subject assigned to the androstenedione group for the training study had elevated fasting glucose levels, was referred to a physician, and was diagnosed as having diabetes mellitus. This subject's data were therefore excluded from the analysis.

Acute Hormonal Response to Androstenedione Administration

Ingestion of 100 mg of androstenedione increased the serum androstenedione concentration by 175% during the first 60 minutes following ingestion (FIGURE 1; $P<.05$). Between 90 and 270 minutes after ingestion, the serum androstenedione concentration was increased by 325% to 350% with androstenedione. Although the serum androstenedione concentration tended to decrease from 270 to 360 minutes after ingestion, serum levels remained elevated above baseline for androstenedione. Serum concentrations of LH and FSH did not change during the 360 minutes following ingestion of either androstenedione or placebo (Figure 1). Ingestion of 100 mg of androstenedione did not affect the serum concentrations of either free or total testosterone (FIGURE 2).

Hormonal Response to Androstenedione Administration During Resistance Training

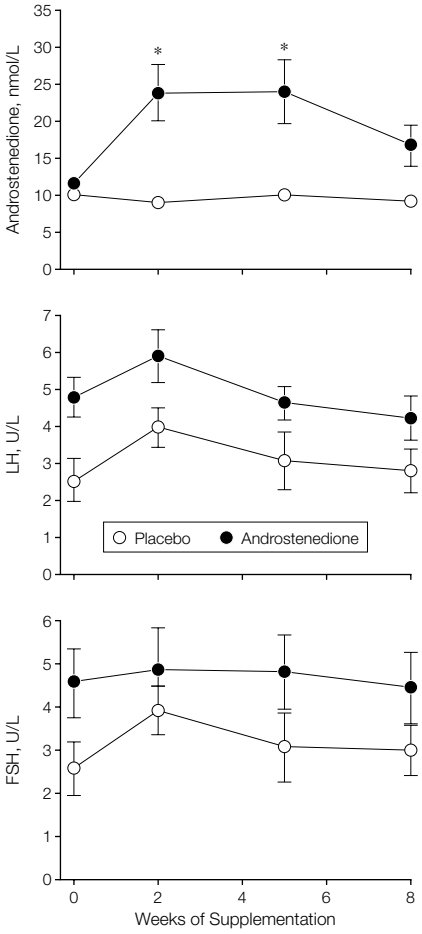
The serum androstenedione concentration (FIGURE 3) increased 100% in the androstenedione group after 2 and 5 weeks of training and supplementation ($P<.05$) and tended to be elevated after 8 weeks ($P=.07$). Serum concentrations of LH and FSH were unaffected by supplementation and training in either androstenedione or placebo groups (Figure 3).

The serum free testosterone concentration (FIGURE 4) was significantly higher in the androstenedione group than in the placebo group before and following supplementation (significant main effect, $P=.01$). The serum free testosterone concentration was not significantly altered by the 8-week period of training and supplementation in either placebo or androstenedione groups. The serum total testosterone concentration was not different in placebo and androstenedione groups prior to supplementation and did not change in either group during the period of training and supplementation.

The calculated effect size for the comparison of the serum free testosterone concentrations between week 0 and week 8 for androstenedione was 0.28. Assuming a power of 80% and $P=.05$, a sample size of 160 would have been required to detect an effect of this size. These calculations highlight the lack of effect of androstenedione supplementation on serum testosterone concentrations.

The serum estradiol concentration was higher prior to supplementation in

Figure 3. Serum Androstenedione, Luteinizing Hormone (LH), and Follicle-Stimulating Hormone (FSH) Concentrations With 300 mg/d of Androstenedione (n = 9) and Placebo (n = 10)



Data are mean (SEM). Asterisk indicates significantly different from time zero for androstenedione ($P<.05$).

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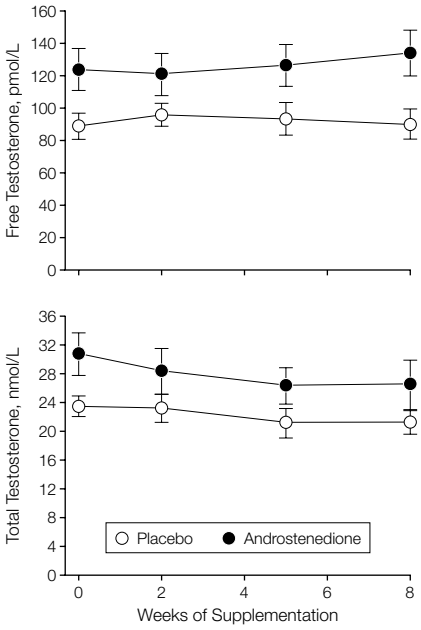
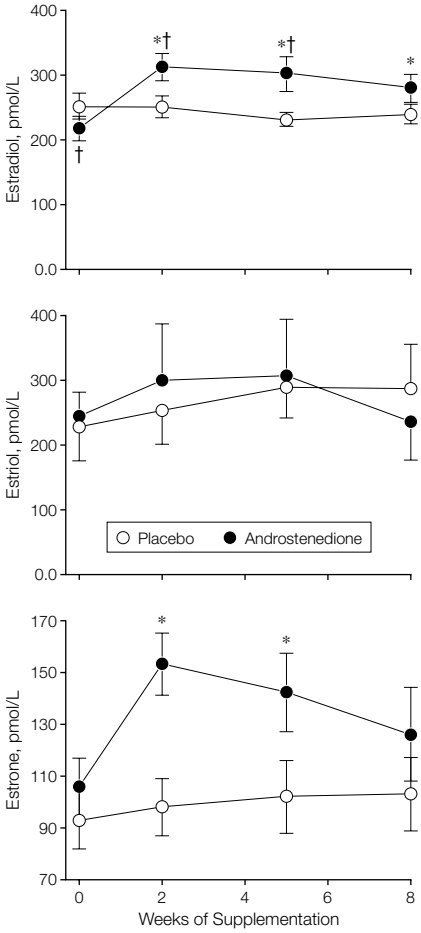
placebo, due to a very high initial value for 1 subject (460 pmol/L). Figure 1 shows the serum estradiol concentration, after eliminating the data of this subject. The serum estradiol concentration prior to supplementation was not different in placebo and androstenedione groups. The serum estradiol concentration did not change significantly during the 8-week experimental period for the placebo group (FIGURE 5). The serum estradiol concentration increased significantly ($P<.05$) in the androstenedione group after 2 weeks (310 [20] pmol/L), 5 weeks (300 [30] pmol/L), and 8 weeks (280 [20] pmol/L) of supplementation compared with presupplementation values (220 [20] pmol/L). The serum estradiol concentration was significantly higher for the androstenedione group compared with the placebo group after 2 and 5 weeks of training and supplementation ($P<.05$). The serum estriol concentration did not change during the 8 weeks of training and supplementation in either placebo or androstenedione groups. In contrast, the serum estrone concentration was significantly ($P<.05$) elevated in the androstenedione group after 2 weeks (153 [12] pmol/L) and 5 weeks (142 [15] pmol/L) of training and supplementation compared with presupplement values (106 [11] pmol/L). The serum estrone concentration did not change during the training and supplementation period in the placebo group. The increases in serum estradiol and estrone concentrations observed after 2 weeks of supplementation were observed in all subjects ingesting androstenedione.

Figure 4. Serum Free and Total Testosterone Concentrations With 300 mg/d of Androstenedione (n = 9) and Placebo (n = 10)



Data are mean (SEM). Serum free testosterone was significantly higher in the androstenedione group vs placebo group (significant main effect, $P<.01$).

Figure 5. Serum Estradiol, Estriol, and Estrone Concentrations With 300 mg/d of Androstenedione (n = 9) and Placebo (n = 10)



Data are mean (SEM). Asterisk indicates significantly different from week zero for androstenedione ($P<.05$). Dagger indicates androstenedione significantly different from placebo ($P<.05$).

The observed values for serum estradiol concentrations appear to be somewhat (20%) higher than those typically reported in the literature. However, there appears to be considerable variability between laboratories, as well as between and within subjects. In addition, these estradiol values obtained at the lower end of the standard curve create more error in the calculation between defined and calculated dose. Serum estriol concentrations were also somewhat higher than those reported in the literature. However, the levels of estriol found in the current study are below the minimal reportable range as indicated by the manufacturer of the radioimmunoassay kits and, therefore, are considered to be within normal limits for men. Regardless of the explanation for these data, comparisons within these subjects over time are valid, since all samples for each subject were analyzed in the same assay.

Clinical Blood Chemistry

The 8-week period of training and supplementation did not affect serum concentrations of total cholesterol, LDL cholesterol, very LDL cholesterol, or triglycerides (TABLE 1). The serum HDL cholesterol concentration was significantly reduced by 12% ($P<.05$) after 2 weeks and remained reduced after 5 and 8 weeks of training and supplementation with androstenedione. Serum concentrations of liver function enzymes were within normal limits for all subjects throughout the study and were unaffected by training or supplementation. Training or supplementation did not significantly affect total iron, hematocrit, and hemoglobin concentrations.

Resistance Training

There was no significant difference between placebo and androstenedione groups in the number of repetitions per training session, amount of force produced, or relative intensity expressed as a percentage of maximal force produc-

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Table 1. Clinical Blood Chemistry Results*

	Androstenedione Group (n = 9)				Placebo Group (n = 10)			
	Week 0	Week 2	Week 5	Week 8	Week 0	Week 2	Week 5	Week 8
Blood lipids, mmol/L								
Total cholesterol	4.24 (0.26)	4.11 (0.28)	4.11 (0.34)	4.11 (0.31)	4.06 (0.26)	4.09 (0.31)	4.34 (0.23)	4.14 (0.23)
HDL cholesterol	1.09 (0.08)	0.96 (0.08)†	0.96 (0.08)†	0.96 (0.08)†	1.01 (0.05)	0.98 (0.08)	1.01 (0.08)	1.01 (0.08)
LDL cholesterol	2.45 (0.28)	2.40 (0.28)	2.61 (0.31)	2.59 (0.31)	2.46 (0.18)	2.51 (0.21)	2.72 (0.21)	2.61 (0.21)
VLDL cholesterol	0.70 (0.10)	0.72 (0.10)	0.52 (0.05)	0.54 (0.08)	0.65 (0.13)	0.57 (0.13)	0.59 (0.13)	0.49 (0.13)
Triglycerides	1.54 (0.20)	1.60 (0.25)	1.14 (0.10)	1.23 (0.19)	1.31 (0.30)	1.25 (0.29)	1.34 (0.26)	1.10 (0.26)
Liver function enzymes, U/L								
γ-Glutamyltransferase	26 (3)	24 (3)	25 (3)	23 (3)	19 (4)	19 (5)	25 (9)	20 (5)
Aspartate aminotransferase	19 (2)	20 (2)	23 (3)	21 (2)	22 (4)	25 (4)	27 (5)	24 (3)
Alanine aminotransferase	19 (2)	20 (3)	21 (3)	18 (2)	22 (5)	27 (7)	34 (15)	24 (7)
Iron and red blood cell status								
Total iron, μmol/L	14.32 (2.33)	16.65 (2.33)	15.93 (1.61)	22.02 (4.83)	15.04 (1.25)	14.50 (1.07)	14.50 (1.43)	13.07 (1.25)
Hematocrit	0.45 (0.007)	0.45 (0.006)	0.44 (0.007)	0.45 (0.006)	0.40 (0.007)	0.41 (0.01)	0.41 (0.008)	0.41 (0.007)
Hemoglobin, g/L	153 (3)	155 (2)	151 (3)	152 (2)	140 (2)	140 (4)	141 (3)	140 (2)

*HDL indicates high-density lipoprotein; LDL, low-density lipoprotein; and VLDL, very low-density lipoprotein. To convert total, HDL, LDL, and VLDL cholesterol values from millimoles per liter to milligrams per deciliter, divide by 0.02586. To convert triglycerides from millimoles per liter to milligrams per deciliter, divide by 0.01129. To convert total iron values from micromoles per liter to micrograms per deciliter, divide by 0.179. All values are presented as mean (SEM).
†Significantly different from week 0 ($P<.05$).

tion (1-RM). When the data from all exercises are combined, the total amount of force production (SE) during the resistance-training program was 343.2 (16.9) kN and 317.8 (30.3) kN for the placebo group and the androstenedione group, respectively. During the first 4 weeks of training, the mean exercise intensity (SEM) for all exercises was 85% (1%) and 86% (1%) of 1-RM for placebo and androstenedione groups, respectively. During the final 4 weeks of training, the mean exercise intensity for all exercises was 82% (1%) and 84% (1%) of 1-RM for placebo and androstenedione groups, respectively.

Table 2. Maximal Muscle Strength*

	Androstenedione Group (n = 9)			Placebo Group (n = 10)		
Exercise	Week 0	Week 4	Week 8	Week 0	Week 4	Week 8
Bench press	647 (68)	766 (69)	840 (73)	851 (95)	925 (106)	1047 (119)
Biceps curl	287 (19)	316 (20)	331 (22)	351 (21)	378 (22)	391 (25)
Knee extension	717 (46)	865 (44)	1024 (57)	770 (45)	1000 (57)	1095 (52)
Lateral pull-down	657 (25)	722 (33)	787 (44)	672 (35)	761 (34)	876 (50)
Left leg flexion	277 (17)	376 (22)	415 (22)	307 (11)	387 (14)	440 (18)
Right leg flexion	292 (14)	385 (24)	430 (28)	311 (10)	391 (14)	445 (16)
Shoulder press	420 (28)	465 (25)	463 (27)	494 (51)	543 (50)	569 (50)
Triceps extension	311 (24)	366 (26)	390 (19)	329 (31)	405 (33)	454 (30)
Vertical butterfly	774 (107)	957 (108)	1081 (94)	927 (99)	1144 (94)	1268 (89)

*All data are presented as mean (SEM), in newtons. All weeks were significantly different from each other (main effect, $P<.05$).

Muscle Strength

Muscle strength (TABLE 2) did not differ between placebo and androstenedione groups before training or after 4 and 8 weeks of resistance training and supplementation. The resistance training resulted in significant increases in strength for each exercise after 4 weeks of resistance training ($P<.05$). The final 4 weeks of training further increased muscle strength for each of these exercises. When the data from placebo and androstenedione groups are combined, gains in strength ranged from 14% (3%) for the biceps curl to 47% (4%) for the left leg curl. Knee extension strength increased ($P<.05$) by 43% and 42% in androstenedione and placebo groups, respectively.

Muscle Histochemistry

Due to an accidental thawing of 1 sample from the placebo group and 2 samples from the androstenedione group due to freezer failure, muscle-fiber–type distribution and cross-sectional areas were determined in 9 placebo and 7 androstenedione subjects. The percentage of type 1 fibers prior to resistance training and supplementation was similar in placebo (44% [4%]) and androstenedione (48% [2%]) groups. Muscle fiber–type distribution did not change as a consequence of resistance training and supplementation in either androstenedione (44% [3%]) or placebo (44% [4%]) groups. The mean (SEM) cross-sectional area of type 1 fibers was not altered with resistance training and supplementation in placebo (3980 [411] vs 4102 [604] μm^2) or androstenedione (3310 [308] vs 3812 [398] μm^2). The mean cross-sectional area of type 2 fibers increased similarly (significant main effect; $P<.05$) in placebo (5271 [485] vs 5728 [451] μm^2) and androstenedione (4703 [471] vs 5307 [604] μm^2) subjects.

Body Composition

Although the resistance-training program (TABLE 3) significantly affected body composition, there were no significant differences between androstenedione and placebo subjects. When the data for both groups are combined, the resistance-training pro-

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Table 3. Body Composition in Newtons*

Parameter	Androstenedione Group (n = 9)			Placebo Group (n = 10)		
	Week 0	Week 4	Week 8	Week 0	Week 4	Week 8
Height, cm	176.8 (2.7)	. . .	. . .	178.0 (1.5)	. . .	. . .
Body mass, kg*†	80.6 (4.1)	81.5 (4.3)	81.2 (4.2)	81.1 (5.2)	83.3 (5.1)	83.2 (4.9)
Lean body mass, kg†	61.2 (2.5)	. . .	64.1 (2.4)	63.1 (2.6)	. . .	66.0 (2.5)
Fat mass, kg†	19.3 (2.9)	. . .	17.1 (3.4)	18.0 (2.9)	. . .	17.2 (2.9)
Body fat,%†	23.5 (2.4)	. . .	20.3 (3.0)	21.3 (1.9)	. . .	19.9 (2.1)
Circumferences, cm						
Biceps*†	31.9 (0.8)	32.2 (0.9)	32.4 (0.8)	32.6 (1.4)	33.2 (1.4)	33.4 (1.3)
Shoulder†‡	119.3 (1.3)	120.3 (1.6)	121.1 (1.4)	120.3 (3.4)	120.2 (3.5)	120.7 (3.3)
Chest†‡	99.6 (2.0)	99.1 (1.9)	99.2 (1.9)	98.0 (3.8)	99.9 (3.8)	100.9 (3.4)
Abdomen*†‡	89.5 (5.2)	88.3 (5.1)	87.7 (5.1)	87.2 (3.8)	86.5 (4.0)	85.9 (3.6)
Waist*†	86.6 (4.2)	84.6 (3.6)	83.7 (3.6)	84.9 (3.9)	83.6 (3.9)	83.7 (3.7)
Hips*†‡	91.3 (3.5)	89.4 (3.9)	88.1 (3.7)	89.2 (2.5)	87.7 (2.1)	85.7 (2.3)
Gluteal*†‡	101.7 (3.1)	98.9 (3.4)	98.2 (3.1)	100.4 (2.5)	98.6 (2.5)	97.4 (2.3)
Thigh	54.2 (1.3)	54.7 (1.9)	54.4 (1.8)	54.7 (1.2)	55.1 (1.4)	55.1 (1.5)
Calf	37.8 (0.7)	37.8 (0.9)	37.6 (0.8)	37.4 (1.7)	38.4 (0.9)	38.6 (0.9)

*Values are mean (SEM). Ellipses indicate parameter not measured at this point. Week 4 was significantly different from week 0 (main effect, $P<.05$).
†Week 8 was significantly different from week 0 (main effect, $P<.05$).
‡Week 8 was significantly different from week 4 (main effect, $P<.05$).

gram significantly ($P<.05$) increased mean body mass (SEM) (80.9 [3.2] vs 82.3 [3.1] kg), mean lean body mass (SEM) (62.2 [1.7] vs 65.1 [1.6] kg), and mean reduced fat mass (SEM) (18.6 [1.9] vs 17.2 [2.1] kg). Significant increases in circumferences occurred for the biceps, shoulder, and chest sites ($P<.05$), while the abdominal, waist, hip, and gluteal circumferences decreased during resistance training in both androstenedione and placebo subjects ($P<.05$).

COMMENT

A major finding of this study is that short- and long-term androstenedione supplementation did not increase the serum testosterone concentration in young men with normal serum testosterone levels. The only prior report on androstenedione administration in humans demonstrated substantial elevations in the blood testosterone concentration in 2 healthy women.[13] In these women, 100 mg of androstenedione produced increases in the blood androstenedione concentration from 0 to 5 nmol/L and increased the blood total testosterone from 3 to 18 nmol/L. The results of the present study are in striking contrast, since the 36-nmol/L increase in the serum androstenedione concentration observed after short-term intake of androstenedione was not accompanied by any increase in the serum testosterone concentration. In the German patent[26] for androstenedione, it is claimed that ingestion of androstenedione increases the serum testosterone concentration by as much as 237% within 15 minutes, followed by a secondary increase of 48% to 97% occurring 3 to 4 days later, and persisting for an additional 6 to 7 days. However, interpretation of this claim is impossible, since the subject population was not described with respect to age, sex, or hormonal status, and no data are presented.

The unchanged serum testosterone concentration with androstenedione supplementation in the present study, coupled with significant elevations in the serum estrone and estradiol concentrations, suggests that a significant proportion of the ingested androstenedione underwent aromatization to these estrogens.[10,11] Anabolic steroid administration has previously been shown to suppress endogenous testosterone production, secondary to decreased serum levels of LH and FSH.[27] In our study, serum concentrations of LH and FSH were unaffected by supplementation, suggesting that hypothalamic-pituitary function was not modified by androstenedione supplementation. Therefore, the unchanged serum testosterone concentration, in spite of the approximately 2.5 times higher androstenedione concentration, appears to be related to an increased formation of estrogens from the exogenous androstenedione.

The quantitative contribution of different tissues to the aromatization of androstenedione is unknown. However, aromatizing activity has been reported in most body tissues, and it is clear that there is ample capacity to support the increased estrone and estradiol concentrations reported in the present study. For example, adipose tissue has a maximal aromatizing activity of 0.072 pmol/g per hour with a Michaelis constant of 25 nmol/L.[28] Since serum androstenedione concentrations were increased to approximately 24 nmol/L, aromatizing activity would have been at half the maximum rate (½ V_{max} or 0.036 pmol/g per hour). With a fat mass of 19.3 kg, calculated total adipose tissue aromatizing activity is 695 pmol/h. If plasma volume is assumed to equal 20% of body weight, or about 4.0 L, the 47-pmol/L increase in the serum estrone concentration observed from week 0 to week 2 would reflect an increase of 188 pmol in the total increase in circulating estrone concentration. Thus, the aromatizing activity of adipose tissue alone could theoretically account for the increased serum estrone concentration observed with androstenedione supplementation. It has also been reported that muscle converts tritiated androstenedione to estrone at a rate almost as great as adipose tissue.[29] Because of its large mass, muscle is also, therefore, a quantitatively significant source of estrogens. Since it has been estimated that muscle and adipose tissue combined account for only 35% to 45% of total extragonadal aromatization to estrogens,[30] it is clear that whole-body aromatizing activity is sufficient to account for the observed increase in the serum estrone concentration.

Since many androstenedione users undoubtedly ingest amounts in excess

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of the 300 mg/d taken in our study, it could be argued that the dose of androstenedione was insufficient to raise serum testosterone levels. This dose exceeds the 100- to 200-mg/d intake recommended by most manufacturers and the dose (100 mg) observed to increase the blood testosterone concentration in women.[13] The lack of any significant increase in the serum testosterone concentration, despite the 175% and 100% increases in the serum androstenedione concentration observed with short- and long-term administration of androstenedione, however, suggests than any putative increase in serum testosterone with higher doses would be associated with additional elevations in the serum estrogen concentration and lowering of the serum HDL concentration.

The significantly higher serum free testosterone concentrations observed in androstenedione both before and during resistance training and supplementation were unexpected, and difficult to explain, given the random assignment of subjects to each treatment group. However, values for all subjects were in the normal range, and it is unlikely that the initial differences influenced the response to the supplementation period.

Although androstenedione supplementation did not enhance the serum testosterone concentration in these young normotestosterogenic men, the reported increase in serum testosterone levels in women[13] may suggest that androstenedione supplementation increases the serum testosterone concentration in hypotestosterogenic populations, such as women and older men.[31,32]

The resistance-training program used in this investigation was effective in enhancing lean body mass, the cross-sectional area of type 2 muscle fibers, and muscle strength. Gains in muscle size and strength are markedly enhanced when androgenic-anabolic steroids are taken in conjunction with a resistance-training program.[1-4] In our study, the increases in lean body mass, muscle fiber cross-sectional area, and muscle strength were not enhanced with androstenedione supplementation. These results are not surprising, since serum testosterone concentrations were not affected by androstenedione supplementation, and since androstenedione has only weak anabolic-androgenic activity in comparison with testosterone.[33] The large increases in strength observed in both experimental groups suggest that the lack of any improvement in strength with androstenedione supplementation is not due to an inadequate training stimulus but instead is due to lack of efficacy of androstenedione as an anabolic-androgenic supplement.

A significant lowering of the serum HDL-C concentration was observed with androstenedione administration, a finding in agreement with prior work demonstrating a lowering of the HDL-C concentration with anabolic steroid use.[3,15-19] The reduction in HDL-C appears to be due primarily to a reduction in the HDL_2 subfraction, secondary to an induction of hepatic triacylglycerol lipase activity.[21,34] The serum HDL-C concentration did not reach a level (<0.91 mmol/L [<35 mg/dL]) typically considered to constitute a risk factor for cardiovascular disease.[35] However, the finding that cardiovascular disease risk increases 2% to 3% with every 0.03-mmol/L (1-mg/dL) decrease in HDL-C suggests that the significant reduction in HDL-C observed with androstenedione supplementation is clinically relevant.[36] Since serum testosterone concentrations were unaffected by androstenedione supplementation, the decrease in HDL-C may be due to the approximately 2.5 times higher serum androstenedione concentration. Previous research has reported that anabolic steroid administration lowers the HDL-C concentration by as much as 27% to 70%.[3,15-19] One possible explanation for the significant, but smaller (12%), decrease in the HDL-C concentration in our study is the lower metabolic potency of androstenedione compared with testosterone.[33] In addition, subjects in prior studies typically consumed high doses of more metabolically active anabolic steroids for more prolonged periods.

Elevated serum liver transaminase concentrations are frequently observed during clinical steroid therapy using 17α-alkylated or other oral compounds.[37,38] Although the serum concentration of liver transaminases has been reported to be significantly elevated with anabolic steroid administration in athletes,[20,21] this is not a universal finding.[3,25,39] In our study, serum liver enzyme levels were unaffected by the 8-week period of androstenedione administration. However, significant impairment of liver function following more prolonged androstenedione supplementation or with higher dosages cannot be ruled out.

The hormonal milieu induced by androstenedione supplementation may predispose the user to adverse consequences in addition to those documented in this study. Increased serum estrogen levels have been known for some time to be associated with the development of gynecomastia.[40] Increased concentrations of estrogens may also increase the risk of cardiovascular disease.[41] Elevated estradiol concentrations have been related to increased risk of breast cancer in women[42] and pancreatic cancer in men.[43] Furthermore, elevated serum androstenedione concentrations have been observed to increase the risk for prostate cancer in some[44] but not all[45] previous studies, and increased serum androstenedione concentrations may also be associated with pancreatic cancer.[46] Taken together, these previous findings suggest that androstenedione supplementation may predispose the user to additional health risks.

In summary, androstenedione administration during resistance training did not significantly alter the serum testosterone concentration in normotestosterogenic young men. The increased muscle size and strength observed with resistance training were also not augmented with androstenedione administration. The use of androstenedione increased the serum concentrations of estradiol and estrone, suggesting an increased aromatization of the ingested androstenedione and/or

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testosterone derived from the exogenous androstenedione. The use of androstenedione was associated with decreased levels of HDL-C. These data provide evidence that androstenedione does not enhance adaptations to resistance training and may result in potentially serious adverse health consequences in young men.

Funding/Support: This research was supported by Experimental and Applied Sciences Inc, Golden, Colo, a manufacturer of oral androstenedione.

Acknowledgment: We appreciate the technical assistance of Emily Martini, Trina Radske, and Vicki Strissel, without whom the project would not have been accomplished. We also thank Murray Kaplan, PhD, for the use of the gamma counter in his laboratory, Roger A. Fielding, PhD, for his insightful comments, and Jerry Thomas, PhD, and Kathi Thomas, PhD, for their help with the statistical analyses.

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