Dihydrotestosterone is elevated following sprint exercise in healthy young men

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EXERCISE IS KNOWN TO TRIGGER acute elevations in circulating androgens, with responses that are dependent on historical training status and workload design (11, 35). These hormonal responses have been implicated both in the execution of acute workout performance and in the accrual of adaptive training gains (9). A variety of mechanisms have been proposed to support these actions, including the activation of cell signaling pathways promoting the mobilization of energy reserves via the glutamine transporter 4 (GLUT4) (33) and the accretion of protein for skeletal muscle hypertrophy (15) via mTor (38), modulation of the excitability of neuromotor units (6), and more complex influences on behavioral motivation (4) and cognition (3). However, the precise nature of the interaction between hormonal response and functional outcome in a given exercise setting remains poorly understood. In particular, the reliance on testosterone as a ubiquitous marker of the androgen system has come under scrutiny with recent attention widening to include another bioactive androgen, dihydrotestosterone (DHT) (40).

DHT is considered the terminal active product of androgen biosynthesis, being produced via both testosterone-dependent and -independent pathways (40). Compared with testosterone, DHT has greater affinity to the androgen receptor (5). DHT has been described as an autocrine/paracrine hormone because it is formed in local tissue by the irreversible 5α-reduction of testosterone or androstenedione and is locally regulated in its action by an enzymatically controlled, reduction-oxidation equilibrium with a series of inactive metabolites (30). As a result, it is typically present in circulation in smaller quantities than other androgens (25, 28). This regulation may serve in part to limit adiogenic actions that have seen chronically elevated DHT levels linked to metabolic and cardiovascular disease risk (14). Given its heightened androgenic potency, the enzymatic machinery required for biocconversion of androgens to DHT is not universally distributed, allowing tissue-specific effects only in selected androgen-sensitive tissues (37).

Emerging evidence suggests that exercise can acutely trigger local muscular conversion of testosterone to DHT in rats (2). DHT has further been shown to enhance force production in isolated fast-twitch muscle fiber bundles from mice via rapid nongenomic signaling (21) and to promote a variety of transcriptional events associated with anabolism via classical genomic signaling (41). Historically, human skeletal muscle was not thought to contain significant levels of the 5α-reductase responsible for conversion of testosterone to DHT (37). However, this view has recently been challenged by the discovery of a third isoform of 5α-reductase that is highly expressed in human skeletal muscle (16).

Given that the role of DHT in the acute adaptive response to exercise in humans is not known, it would be prudent to establish whether exercise provokes an increase in circulating DHT before any detailed exploration of its potential role in exercising muscle function. The purpose of the present study was therefore to determine whether circulating DHT is acutely elevated in a group of exercise-trained men performing repeat sprint exercise that involves sustained recruitment of fast-twitch muscle fibers. We further aimed to establish whether this exercise response was related to training status or characteristics of the sprint performance.

MATERIALS AND METHODS

Participants

Fourteen healthy men participated in this study [age 28.3 ± 4.1 yr; body mass (BM) 75.6 ± 8.5 kg; \( V\dot{O}_{2\text{max}} \) 61.0 ± 8.1 ml·kg BM\(^{-1}\)·min\(^{-1}\)]. All participants completed a self-report training ques-
tionnaire and were categorized as either a recreational (n = 7) or a competitive (n = 7) athlete with a minimum training history of 6 yr. These criteria were established to isolate the effects of training and competition status without the confounding effects of years of training history, as both have been implicated in the modulation of the hormonal response to exercise (11). Sports participation included running, cycling, swimming, triathlon, combat sports, ball sports, and team sports. Among the reported activities, all competitive athletes participated in cycling or triathlon, and all but one of the recreational athletes were engaged in resistance training. The study received approval by the University of Bath Ethics Committee, and each participant was provided with a written and verbal briefing regarding the nature of the testing and provided written, informed consent before participation.

Protocol

Incremental exercise trial. To determine participant fitness and to set workloads for subsequent sprint exercise, all participants undertook an incremental exercise test to exhaustion on a stationary cycle ergometer (Schoberer Rad Messtechnik; SRM, Jülich, Germany). Expired gases were collected in the final minute of exercise using a Douglas bag and subsequently analyzed for volume of air expired using a dry gas meter (Harvard Apparatus, Kent, UK), temperature of expired gases via a digital thermometer (model C; Edale Instruments, Cambridge, UK), and fractional concentrations of O2 and CO2 using paramagnetic and infrared methods, respectively (Servoflex MiniMP; Servomex, Crowborough, UK). These analyzers were calibrated before each test with gases of known composition and volume within the physiological range, as certified by prior gravimetric analysis (British Oxygen, Guildford, UK), and measured values were used to determine the maximal rate of oxygen consumption (VO2max). Starting load was self-selected based on warm up intensity, and the exercise protocol consisted of 30-W increments every 3 min for 15 min followed by self-selected based on warm up intensity, and the exercise protocol consisted of 30-W increments every 3 min for 15 min followed by 20-W increments per minute until exhaustion. Power output during the final minute was averaged to represent work capacity (Wmax).

Repe t sprint exercise trial. On a subsequent visit, at least 48 h later, participants completed a bout of sprint interval cycle exercise on the stationary ergometer (SRM, Germany), consisting of 10 repetitions of 30 s sprinting at a target load of 150% of the Wmax determined from the incremental test, interspersed with 90 s of low-intensity recovery cycling at or below 100 W. To normalize for circadian variation in hormonal state (24), all sprint testing took place in the morning between 2 and 4 h after wakening. Workload was self-paced, and participants were given real-time numerical and graphical feedback on their current power output, cycling cadence, and time elapsed. Where participants were unable to sustain the target load, they were encouraged to perform maximally during the remaining sprints.

Blood sampling. On arrival at the laboratory, a trained phlebotomist collected 5 ml of venous blood from a superficial antecubital vein without stasis. Following this preexercise sample, further samples were collected 5 and 60 min postexercise. Samples were suspended in serum collection tubes (Serum Z/5 ml; Sarstedt, Sarstedt, Germany) for 15 min before being centrifuged for 10 min at 1,500 g. Supernatant was immediately transferred to polypropylene Eppendorf tubes and frozen at −20°C until analysis.

Hormonal analysis. Serum samples were analyzed in duplicate for total testosterone, free testosterone, and DHT concentrations, using commercial ELISA kits (IBL, Hamburg, Germany). All samples for a given participant were analyzed on the same assay plate. Pooled intra-assay sample variance for duplicate samples was 4.5% for total testosterone, 5.6% for free testosterone, and 5.3% for DHT, and interassay sample variance for controls was 6.3% for total testosterone, 6.4% for free testosterone, and 8.9% for DHT.

Statistical Analysis

Demographic comparisons between athlete groups were conducted by one-way ANOVA, and hormonal responses were analyzed using a two-way (group × time) repeated-measures general linear model, with the Greenhouse-Geisser correction applied where assumptions of sphericity were violated. Main effects were isolated by pairwise multiple comparisons using the Bonferroni method. All hormonal data were normalized by natural log transformation before analysis. Hormonal changes from before to after exercise for different androgens were then examined for correlation to each other and to demographic and sprint performance data by Pearson product moment correlation.

All statistical analysis was conducted using IBM SPSS Statistics (Release 20.0.0; IBM, New York, NY), and all data are presented as means ± SD. Statistical significance was set at the level of P < 0.05.

RESULTS

Participant Fitness and Training History

There were no significant differences in age or BM between athlete groups categorized by competition status (Table 1). However, both VO2max (P = 0.011) and Wmax (P = 0.002) calculated during the incremental exercise protocol were significantly higher for competitive compared with recreational athletes. Competitive athletes also reported a significantly higher habitual training volume than recreational athletes (competitive vs. recreational: 10.5 ± 6.8 h/wk vs. 3.7 ± 0.9 h/wk; P = 0.022) but with no difference in years of training history (10.3 ± 5.2 yr vs. 11.4 ± 3.0 yr; P = 0.616).

Sprint Performance

The competitive group completed the sprint task at the target load of 150% of measured work capacity (average power: 149.4 ± 10.1% Wmax). However, the competitive group were less able to sustain the target load for all ten repetitions (average power: 135.3 ± 15.5% Wmax) although the difference between groups did not reach significance (P = 0.066). As a result, competitive athletes performed at a significantly higher average power (competitive vs. recreational: 560.1 ± 62.1 W vs. 404.1 ± 68.4 W; P = 0.001), resulting in a greater work done during sprinting (168.0 ± 18.6 kJ vs. 121.2 ± 20.5 kJ; P = 0.001) compared with recreational athletes.

Hormonal Response to Sprint Exercise

Significant elevations from preexercise to 5 min postexercise occurred in the levels of total testosterone (P = 0.002), free testosterone (P = 0.001), and DHT (P = 0.022), with all three parameters returning to preexercise levels by 60 min postexercise (Fig. 1). Preexercise levels of free testosterone were correlated with levels of total testosterone (r = 0.689; P = 0.006) and DHT (Fig. 2A, r = 0.536; P < 0.05). Changes in DHT levels with exercise (5 min postexercise − preexercise)

Table 1. Participant characteristics (n = 7 competitive, n = 7 recreational)

<table>
<thead>
<tr>
<th></th>
<th>Competitive Athlete</th>
<th>Recreational Athlete</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age, yr</strong></td>
<td>28.5 ± 5.1</td>
<td>28.0 ± 3.1</td>
</tr>
<tr>
<td><strong>Body Mass, kg</strong></td>
<td>74.3 ± 7.3</td>
<td>76.8 ± 10.0</td>
</tr>
<tr>
<td><strong>VO2max, ml·kg·min−1</strong></td>
<td>66.1 ± 4.5</td>
<td>55.9 ± 7.8†</td>
</tr>
<tr>
<td><strong>Wmax, W</strong></td>
<td>375.8 ± 44.5</td>
<td>297.7 ± 26.0†</td>
</tr>
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Values are means ± SD. *P = 0.011, †P = 0.002 vs. competitive.
correlated significantly with changes in levels of total testosterone \( (r = 0.870; P = 0.001) \) and free testosterone (Fig. 2B, \( r = 0.914; P < 0.001 \)).

**Hormone Response, Training Status and Sprint Performance**

All hormonal responses were compared in relation to competitive status, and no group effects or group \( \times \) time interactions were found for competitive vs. recreational athletes. However, the sprinting cadence averaged across all ten sprints correlated with changes in free testosterone \( (r = 0.574, P = 0.032) \), and the highest average sprinting cadence across an individual sprint bout was associated with changes in free testosterone \( (r = 0.697, P = 0.006) \), DHT \( (r = 0.625, P = 0.017) \), and total testosterone \( (r = 0.603, P = 0.022) \). Habitual weekly training volume correlated with the change in total testosterone only \( (r = 0.569, P = 0.034) \). Hormonal exercise responses did not vary in relation to work capacity during sprinting or fitness status determined during the incremental exercise trial.

**DISCUSSION**

To our knowledge, this is the first report of the presence of increased circulating levels of the androgen DHT during the period immediately following an exercise stimulus in humans. Elevations in DHT were short lived, returning to baseline levels within 1 h after exercise. This time scale of action is consistent with previous reports for testosterone (13), while adding DHT to the array of androgenic factors elevated in the acute phase response to exercise. The presence of elevated DHT during exercise may be of considerable significance given that DHT has a greater affinity than testosterone at the androgenic receptor and creates a more stable androgen-receptor complex (5, 18). That these elevations were short lived may be of equal importance in determining their functional relevance.

The short-acting effects of androgens during exercise have attracted recent attention (9). Elevations in testosterone during exercise are thought to play a role in supporting the maximal expression of powerful muscular force production, linking motivational behavior with functional output (8), but this relationship appears to depend on training status (10). Whether these modulatory effects of training extend to the activation of 5\( \alpha \)-reductase in exercising skeletal muscle is not yet known. Modulatory increases in skeletal muscle DHT levels in response to exercise training have recently been shown in rodents (1), and chronic upregulation of basal circulating DHT levels with training has also been reported in humans (23). However, acute exercise responses were not reported. In the present study, a significant increase in circulating DHT was observed following sprint exercise in a group of healthy young men with a history of six or more years of exercise training. Neither performance capacity in the tests administered nor the habitual training volume of these athletes discriminated the degree of DHT response. Although these findings are in conflict with previous reports indicating that training exposure can modulate...
the androgen response to exercise (11), this may be unsurprising given the mixed athletic background of this cohort. A similar divergence has been shown in other mixed-ability athletic groups, suggesting that androgen responsiveness may only be pertinent to functional output in a subgroup of athletes (10, 12). Future studies should examine acute DHT responses to exercise in less well trained or sedentary individuals and explore the modulation of these responses and their relationship to functional gain with the imposition of an exercise-training program.

In the trained young men studied here, we found an association between the highest volitional cadence maintained for 30 s during sprint cycling and the degree of responsiveness to the sprint stimulus of all three of the measured androgens, including DHT. Although lacking causation, this correlation points toward the possibility that phenotypical characteristics of athletic ability may play a role in determining the hormonal response to sprinting. In trained cyclists, the proportion of fast-twitch fibers by cross-sectional area of vastus lateralis muscle, an aspect of athletic phenotype known to carry a significant degree of heritability (34), correlates strongly with optimal sprinting cadence and to a lesser degree with maximal sprinting power output (22). This relationship holds with the age-related decline of muscle function in older men (27). At the cellular level, DHT has been shown to increase transcriptional signaling of factors involved in ATP production and calcium cycling in mouse skeletal muscle (41). In the rat, cultured skeletal muscle cells exposed to testosterone or dehydroepiandrosterone exhibit a DHT conversion-dependent enhancement of glucose metabolism by increasing protein expression and translocation of GLUT-4 (33), and administration of DHT to isolated muscle fiber bundles enhances contractile function, but only in fast-twitch fibers (21). It is possible, therefore, that the androgenic response to sprint exercise in men may depend to some degree on the morphology of active muscle. Alternatively, sprint cadence and androgen status may be covariates of an underlying driver related to other characteristics of the exercise response, such as changes in contractile function and cellular metabolism resulting from alterations in anisosmotic compartmental fluid dynamics (7).

The tight coupling of exercise-induced increases in total and free testosterone and DHT in the present study (Fig. 2) is suggestive of a common pathway of androgenic promotion during repeat sprint exercise, with skeletal muscle a likely candidate in driving this response. In support of this view, preliminary findings from our group indicate that elevations in DHT can also be detected in human muscle dialysate during high-intensity exercise (unpublished observations). In rodents, exercise has been shown to promote active bioconversion of androgens by skeletal muscle, resulting in temporal elevations in both DHT and testosterone of a similar magnitude and time frame to those seen here (2). However, it is presently not known whether human skeletal muscle is capable of local androgenic bioconversion in the face of an exercise stimulus. Until recently, it was widely thought that human skeletal muscle lacked expression of the 5α-reductase responsible for conversion of testosterone to DHT (36, 37). Empirical data directly supporting these claims in normal, healthy humans is scarce in the available literature, with inferences regarding 5α-reductase inactivity largely dependent on observations of normal muscular development in patients with congenital 5α-reductase deficiency (17, 31), and with evidence of sustained muscle hypertrophy during exogenous testosterone administration in the presence of 5α-reductase inhibition in testosterone-deficient men (26). Crucially, the provenance of tissue samples from the available studies directly examining 5α-reductase in healthy humans is unclear, leaving open the prospect that expression of 5α-reductase is actively modulated in skeletal muscle by factors such as phenotype, age, and exercise training. This view is strengthened by recent reports of a third isoenzyme of 5α-reductase that shows an expression profile in human skeletal muscle (16, 39). Further studies are required to explicitly address this question in humans by conducting muscle biopsies following an exercise stimulus to determine both the changes in local androgen metabolism and the fiber type specificity of this response.

Much debate surrounds the functional role of androgens in the anabolic response to exercise training (29, 32). Although beyond the scope of this study, future investigations should consider the potential relevance of DHT to exercise-induced anabolism because administration of DHT to isolated intact muscle fiber bundles from the rat promotes protein accretion (20), with effects mediated by mitogen-activated protein kinase, and activates a transcriptome signature highly supportive of anabolic pathways in an equivalent mouse model (41). If DHT is found to be of relevance to anabolic signaling, then consideration should also be given to the resilience of the exercising DHT response under chronic training load. Depletion of circulating androgen levels is a feature of the continued high-level stimulation of an intense conditioning exercise program (19). Because 5α-reduction of testosterone to DHT is an irreversible step in androgen metabolism, necessitating the inactivation of DHT following a stimulus (28), the repeated elevation of DHT in response to the day-to-day demands of an intense conditioning exercise program may place a strain on androgen metabolism with potential implications for maintenance of future training load and subsequent adaptive response. That the habitual training volume of participants in the present study was related to the total testosterone response to repeat sprint exercise could be indicative of an adaptation designed to sustain circulating androgen levels under training load.

This study represents a small staging post in the examination of the role of DHT in exercising muscle function. Extension of the findings presented here is limited by the small cohort studied and the extensive training history of participants. Whether the relationships reported would hold for more diverse groups encompassing sedentary or less well trained individuals and highly trained elite athletes, or for different modes or durations of exercise, was beyond the scope of the present study. A further note of caution is required in comparing the acute responses of this group since there may have been variable adherence to advice regarding the control of training load in the days before the sprint trials took place. It remains possible that residual training fatigue differentially influenced both the ability of participants to perform optimally and the extent of their hormonal response.

Conclusion

Repeat sprint exercise is capable of inducing an increase in circulating DHT in healthy active young men, particularly when sprinting cadence is high. It would be of significant value
to determine whether DHT is converted locally from other androgens by active muscle during exercise and what factors might determine the extent of the exercising DHT response, such as age, sex, competitive athletic status, exercise training history, and characteristics of the exercise stimulus. The role of DHT in modulating muscle function during exercise training and sports performance warrants further investigation.

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GRANTS

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS


REFERENCES


39. Yamana K, Labrie F, Luu-The V. Human type 3 5α-reductase is expressed in peripheral tissues at higher levels than types 1 and 2 and its activity is potently inhibited by finasteride and dutasteride. *Horm Mol Biol Clin Invest* 2: 293–299, 2010.
