Original Research

Intercollegiate Cross Country Competition: Effects of Warm-up and Racing on Salivary Levels of Cortisol and Testosterone

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ABSTRACT

International Journal of Exercise Science 7(4): 318-328, 2014. Team intercollegiate athletic competition is associated with an increase in salivary cortisol (C) and testosterone (T) in men and women. The present study was designed to determine the hormonal effects of warm-up and racing in cross country runners - a sport that has both individual and team components. Members of the Emory University men’s and women’s varsity cross country teams gave saliva samples before warm-up, after warm-up, and immediately after the finish of each of two intercollegiate invitational meets held one year apart in the same setting (2010, N = 10 men, 15 women; 2011, N = 15 men, 20 women). For some racers warm-up was associated with a significant decrease in C (2010 men p = .04; 2011 women, p = .004). With the exception of the 2011 men, warm-up was associated with an increase in T (2010 men, P = .012; 2010 women, p = .006; 2011 women, p = .056). For men and women in both years, racing was related to a substantial increase in both C and T (C: 2010 and 2011 men, p = .001; 2010 women, p = .011; 2011 women, p < .001) (T: 2010 and 2011 men and women, p < .001). Finish time was not related to levels of C or T. Increased hormone levels may result from the psychological effects of competition, physical exertion, or some combination of the two. Competition-related increases in C and T presumably benefit performance in cross country racing and other sports, but the exact character of these benefits remains to be determined.

KEY WORDS: Athletic competition, cross country racing, hormonal correlates

INTRODUCTION

In virtually all sports, athletes warm up in the hour prior to formal competition. Warm-up typically includes multiple forms of aerobic exercise, stretching, and sport-specific drills intended to prevent injury and prepare the body for the physical demands of competition. This is also a time when athletes mentally prepare to meet the demands of the competition to come. Although previous research has established that athletic competition significantly increases levels of cortisol (C) and testosterone (T), across a variety of sporting contexts (2, 16, 17, 21, 23, 40), there is little known about the hormonal responses to the warm-up period. Edwards and Kurlander (16), in what is apparently the only published study of this effect, showed that warm-up for women tennis and volleyball players is associated with a substantial elevation in T (but not C) that, in effect, anticipates the start of the competition. Whether a similar effect would be seen in male athletes is not known. Hormone
changes during warm-up may play an important role in activating various physiological systems or psychological states that are advantageous for sport competition (6, 9, 12).

Cortisol is a glucocorticoid hormone secreted from the adrenal cortex in response to physical and psychological stress (15). Although short-term elevations in C are advantageous in supplying muscle tissue with energy and reducing inflammation (11, 32), sustained high levels of cortisol may be disadvantageously catabolic (42). Testosterone is a steroid hormone secreted from the testes in men and the ovaries in women. Additional amounts of T in both men and women are contributed by the adrenal cortex and by peripheral conversion of precursor hormones (39). T has proven positive effects on muscle mass and strength (3) and levels of this hormone have been shown to increase in response to endurance as well as resistance-based physical exercise (12). Additionally, T levels are thought to increase as a result of a variety of social interactions, primarily those that are competitive in nature (18), and high levels of this hormone appear to be associated with high dominance motivation/implicit power (29, 38). Winners of athletic competitions have been shown to have higher after-competition T levels than losers (25), although there are notable exceptions to this findings (16, 17). Short term elevations in T are likely to be advantageous for mental and physical performance under pressure (12).

Previous studies of the hormonal correlates of athletic competition have focused predominantly on team sports (2, 16, 17, 40) in which athletes play for points, have intermittent physical exertion, and where teammates coordinate their individual efforts towards the shared goal of defeating an opposing team. In endurance sports (e.g., distance running), individual athletes compete for rank among all other competitors with sustained physical exertion. Cross country racing is an endurance sport in which athletes intensely run a distance of between 5-10 kilometers over natural terrain. In the sense that the outcome of the competition for a cross country team (victory/defeat) depends on the summed individual performances of team members, intercollegiate cross country racing has both team and individual components. On the premise that a full understanding of the effects of any hormone on physiology and behavior is informed by an appreciation of the settings and circumstances that affect its fluctuating levels, the present study was designed to determine the effects of warm-up and racing on salivary levels of C and T in men and women cross country runners. To this end, saliva samples were obtained from runners before warm-up, after warm-up, and immediately after race completion and were subsequently assayed for C and T. A secondary purpose of the study was to determine the relationship, if any, between hormone levels and race performance.

METHODS

Participants
Participants (18-22 years old) for the study were consenting members of the 2010 and 2011 Emory University varsity men’s (n = 13 and n = 15, respectively), and women’s (n = 16 and n = 22, respectively) cross country teams. This research was
approved by Emory’s Institutional Review Board and athletes gave written informed consent prior to participation. As part of the consent procedure women were asked to respond “yes” or “no” to the question “Are you currently using an oral contraceptive?” and to one other: “Are you currently using any injected, implanted, or patch-delivered hormone contraceptive?”

**Protocol**

In 2010 and 2011, saliva samples were obtained at a single NCAA, Division III, intercollegiate cross country meet in Jacksonville, Alabama during the second week in October. Each year the races were on the same course and held under the same weather conditions (sunny, 23-26°C). Each participant gave three saliva samples: before warm-up (baseline), after warm-up, and immediately after crossing the finish line. Men and women warmed-up with light running for a continuous period of 15-20 minutes at a pace of approximately 7-7:30 minutes per mile for men and 8-8:30 minutes per mile for women. After warming-up, participants gave saliva samples a few minutes before heading to the start line for last minute strides at race pace and stretching.

The men’s contest began at 9:15 AM, with 90-95 men from 10-11 different universities/colleges racing over an 8 kilometer course. The women’s contest began at 10:00 AM, with 100-110 women from 12 different universities/colleges racing over a 5 kilometer course. In some instances (2010 race for men and women and 2011 race for men) a small number of individuals warmed-up with their teammates but did not race. During the race, these individuals cheered for their team near the finish line. For these participants, “after competition” samples were obtained at the same time as the after competition samples for the first cohort of race finishers on their team. Because the number of non-racers was so small, their hormone values were not included in any of the statistical analyses conducted for this report.

Participants were provided with a piece of sugar-free gum (Trident®, original flavor) to stimulate saliva production and they gave samples by passive drool exactly according to the protocol used for athletes in other sports (16). Briefly, participants chewed for two minutes before they began to give the saliva sample to be used for assay. Saliva collection was typically completed within five minutes after that. Although the use of chewing gum to stimulate salivation and speed collection of saliva samples is common, this practice has been recently questioned in reports that chewing gum may distort salivary T levels relative to unstimulated samples (43). In the present study, all participants chewed Trident® original flavor gum. Provided individuals chew for more than one minute before they begin to deliver the sample, this particular gum does not appear to affect salivary T level relative to what is assayed from unstimulated samples (14, 22). Dehydration may reduce saliva flow. Although this may affect the concentration of larger molecules such as DHEAs and total protein concentration, flow rate does not affect the concentration of small molecules such as C and T that undergo passive diffusion into the saliva compartment (7, 44, 45).
Samples were stored at -80°C and were later assayed in duplicate for C and T by the Biomarkers Core laboratory of the Yerkes Primate Center in Atlanta, Georgia using competitive enzyme immunoassay kits from Salimetrics (State College, PA). The average intra assay coefficients of variation for the 2010 and 2011 samples were 6.13% and 3.15% for C and 6.37% and 5.46% for T, respectively. Because the molecular size of hormone binding proteins in blood is too large to allow entry into salivary compartments, it is commonly believed (34) that hormone levels measured in saliva represent the un-bound (bioactive) fraction of hormones in blood (37).

Statistical Analysis
The SPSS statistical package was used for calculation of independent t-tests (two-tailed), Pearson correlation coefficients, and repeated-measures ANOVA with within subjects post-hoc contrasts for the changes in hormone levels across warm-up and racing. In all cases, $p \leq 0.05$ was required for statistical significance.

RESULTS
Levels of C and T for both men and women in this study were within the ranges described as being typical (Salimetrics.com) for saliva samples from adult men and women assayed with the Salimetrics enzyme immunoassay (EIA) kits. Salivary levels of C for men and women were similar and means were not significantly different at any of the times sampled. Baseline T was substantially higher for men than women (2010, $t(27) = 7.80$, $p < .001$; 2011, $t (35) = 10.17$, $p < .001$) undoubtedly owing to the substantial hormonal contribution of the testes in men.

Ten women on the 2010 team and 14 women on the 2011 team reported using oral contraceptives; one woman on the 2011 team reported using some other form of hormone contraception. For the women who raced, baseline salivary C levels were similar and not significantly different between hormone contraception users and non-users in either 2010 or 2011 ($t(13) = 1.42$, $p = .179$; $t(18) = .214$, $p = .833$, respectively). Women using hormone contraceptives had lower baseline T on average than women not using hormone contraceptives, but the difference between baseline levels for users and non-users was not statistically significant for either the 2010 or 2011 women ($t(13) = 1.12$, $p = .282$; $t(18) = 1.53$, $p = .146$, respectively). Absent a significant difference between hormone levels for hormone contraception users and non-users, hormone values for users and non-users were combined for purposes of analysis and presentation.

![Figure 1. Before warm-up, after warm-up, and after competition salivary levels of cortisol and testosterone for the 2010 men. Group means are shown for men who raced and men who did not race. Error Bars = ±1 SEM. Statistically significant differences between before and after warm-up and between after warm-up and after competition means are noted. * $P < .05$, **$P \leq .001$.](http://www.intjexersci.com)
Salivary C and T means are shown in Figures 1 and 2 for men and Figures 3 and 4 for women. The number of individuals who warmed-up but did not race in any given year for men or women was too small to make meaningful statistical comparisons, but hormone values for these athletes are included in the figures as useful reference points.

Repeated-measures ANOVA revealed a significant main effect of the time of hormone sampling on C levels in men (2010: $F(2, 18) = 21.61, p < .001$; 2011: $F(2, 28) = 13.96, p < .001$). Within subjects contrasts showed that in 2010, but not 2011, C levels were significantly decreased from before to after warm-up (2010: $F(1,9) = 5.75, p = .040, r = .62$; 2011: $F(1,14) = 3.77, p = .073, r = .46$). In both 2010 and 2011, contrasts revealed that for men who raced, C levels were significantly increased from before to after competition (2010: $F(1,9) = 24.92, p = .001, r = .86$; 2011: $F(1,14) = 16.16, p = .001, r = .73$).
For women, repeated-measures ANOVA also revealed a significant main effect of the time of hormone sampling on C levels (2010: F(2, 28) = 7.29, p = .003; 2011: F(2, 38) = 23.78, p < .001). Within subjects contrasts showed that in 2011, but not 2010, C levels in women who would later go onto race were significantly decreased from before to after warm-up (F(1,14) = 1.29, p = .276, r = .29; F(1,19) = 10.42, p = .004, r = .60). In both 2010 and 2011, contrasts revealed that for women who raced, C levels were significantly increased from before to after competition (2010: F(1,14) = 8.66, p = .011, r = .62; 2011: F(1,19) = 27.27, p < .001, r = .77).

Repeated-measures ANOVA revealed a significant main effect of the time of hormone sampling on T levels in men (2010: F(2, 18) = 38.81, p < .001; 2011: F(2, 28) = 47.52, p < .001). Within subjects contrasts showed that in 2010, but not 2011, T levels were significantly increased from before to after warm-up (F(1,9) = 9.75, p = .012, r = .72; 2011: F(1,14) = .31, p = .596, r = .15). In both 2010 and 2011, contrasts revealed that for men who raced, T levels were significantly increased from before to after competition (2010: F(1,9) = 40.09, p < .001, r = .90; 2011: F(1,14) = 98.99, p < .001, r = .94).

For women, repeated-measures ANOVA also revealed a significant main effect of the time of hormone sampling on T levels (2010: F(2, 28) = 24.87, p < .001; 2011: F(2, 38) = 25.65, p < .001). Within subjects contrasts showed that in 2010, T levels in women who would go onto race showed an increase from before to after competition, but this effect only approached significance, F(1,14) = 10.22, p = .006, r = .65. In 2011, women who would go onto race showed an increase in T levels from before to after competition (2010: F(1,14) = 10.22, p = .006, r = .65). Within subjects contrasts showed that in 2010, T levels in women who would later go onto race were significantly increased from before to after warm-up, F(1,14) = 10.22, p = .006, r = .65. In 2011, women who would go onto race showed an increase in T levels from before to after competition, but this effect only approached significance, F(1,19) = 4.16, p = .056, r = .42). In both 2010 and 2011, contrasts revealed that for women who raced, T levels were significantly increased from before to after competition (2010: F(1,14) = 28.43, p < .001, r = .82; 2011: F(1,19) = 27.13, p < .001, r = .77).

For the men and women who raced, individual performance was measured by finish time. Men completed their 8k race in 26-32 minutes and women completed their 5k race in 18-23 minutes. Correlation analysis showed that there were no consistent significant relationships between either C or T and finish time at any point in which these hormones were measured (Table 1). T levels for women in 2010 showed significant positive correlations

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**Table 1.** Correlation coefficients (in parentheses) for the relationship between hormone levels and their percent change and finish time. *P < .05.

<table>
<thead>
<tr>
<th></th>
<th>Before Warm-up</th>
<th>After Warm-up</th>
<th>After Competition</th>
<th>Warm-up % Change</th>
<th>Competition % Change</th>
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<tbody>
<tr>
<td>Men 2010</td>
<td>T(-.03)</td>
<td>T(.13)</td>
<td>T(-.18)</td>
<td>T(.35)</td>
<td>T(-.37)</td>
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<tr>
<td></td>
<td>C(-.09)</td>
<td>C(-.57)</td>
<td>C(-.08)</td>
<td>C(-.29)</td>
<td>C(.55)</td>
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<tr>
<td>Men 2011</td>
<td>T(-.17)</td>
<td>T(-.02)</td>
<td>T(38)</td>
<td>T(-.36)</td>
<td>T(-.47)</td>
</tr>
<tr>
<td></td>
<td>C(-.03)</td>
<td>C(-.42)</td>
<td>C(.02)</td>
<td>C(-.35)</td>
<td>C(.52*)</td>
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<tr>
<td>Women 2010</td>
<td>T(.75*)</td>
<td>T(.64*)</td>
<td>T(.73*)</td>
<td>T(.08)</td>
<td>T(.13)</td>
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<tr>
<td></td>
<td>C(-.27)</td>
<td>C(-.04)</td>
<td>C(.30)</td>
<td>C(.26)</td>
<td>C(.41)</td>
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<tr>
<td>Women 2011</td>
<td>T(-.02)</td>
<td>T(-.18)</td>
<td>T(-.26)</td>
<td>T(-.27)</td>
<td>T(-.04)</td>
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<td></td>
<td>C(.19)</td>
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with finish time. However, this relationship was carried by one individual who had the highest T (more than two standard deviations above the mean) and the slowest finish time by over a minute.

DISCUSSION

Warming up is a common practice in competitive sports, with clear physical advantages for performance and injury prevention (36). Cortisol is a hormone that increases in response to both physical and psychological stress, two elements that would seem to be a natural part of warming up for a strenuous athletic competition. However, there was a decrease in mean level of C associated with warm-up in all cases and this decrease was significant in the 2010 men and 2011 women. Although high intensity exercise over 20 minutes increases C levels, low to moderate exercise characteristic of a typical cross country warm-up, does not (24). The decrease in C associated with warm-up in the present study may reflect the well-established circadian morning decline in C (19). Additionally, increased blood flow resulting from warming up may increase the metabolic clearance of high morning levels of C accumulated during rest.

For the 2010 men, warm-up was also associated with a statistically significant increase in salivary T levels (Fig. 1). This is, we believe, the first demonstration of such an effect in men’s athletic competition. We did not see a similar effect for the 2011 men, so some caution about the reliability of this effect for men is warranted. For the 2010 and 2011 women, warm-up was associated with a statistically significant or nearly significant increase in salivary T levels (Figs. 3 and 4). These results complement similar results seen in intercollegiate women volleyball (16) and soccer (7) players. Taken together, they indicate that an increase in T is a common part of the warm-up experience in women athletes competing in a variety of sports. This increase may reflect the psychological anticipation of the competition to come and/or may be a consequence of the running/stretching associated with the warm-up routine. Despite the fact that athletes in virtually all sports warm-up before competition, the warm-up period has been essentially ignored in studies of the hormonal correlates of athletic competition. The results of the present study highlight the need for a more detailed study of the biological and psychological/cognitive correlates of the warm-up period in men and women athletes.

Racing was associated with a substantial increase in salivary levels of C and T and this effect was seen for both men and women in two nearly identical competitions one year apart. This effect is in accordance with the demonstration of competition-related increases in salivary C and T levels in men and women athletes competing in a variety of team sports including soccer, doubles tennis, volleyball, and rugby (1, 2, 16, 17). Increases in C and/or T have also been noted for men and women competing in direct “one on one” athletic contests including judo, wrestling, and singles tennis (8, 21, 23, 40). Although previous reports of the hormonal effects of marathon racing are mixed (10, 27, 35), the present report provides unequivocal evidence to the effect that cross country racing, at a shorter distance, is associated
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with a substantial elevation in levels of C and T in men and women racers.

Race related elevations in C and T could be due to the psychological effects of competition, a response to physical exertion, or some combination of the two. Total and free T in plasma increase during aerobic exercise in proportion to the duration of the workout (13, 41). High levels of physical exertion could also increase salivary levels of C and T through decreased metabolic clearance rate (4) and/or increased hemoconcentration (26).

A rise in C and/or T during athletic competition may have physiological and/or psychological benefits. Short-term increases in C have been shown to be adaptive in increasing glucose levels in blood (thereby supplying muscle tissue with energy) and in reducing inflammation (11). Exogenously administrated C has been shown to increase mood as well as decrease anger, anxiety, and fatigue following a cognitive task (33).

There are few studies of the connection between transient changes in endogenous T and human performance. Placing men and women in “high power” physical poses increases T (and decreases C) and increases subjective feelings of power and tolerance for risk (5). In men, increased T in response to watching a four minute aggressive or training motivational video was positively related to heavier lifting during a resistance training exercise (9). Short-term increases in T have also been shown to have rapid and non-genomic effects on neural functioning in some species (20), and peripherally and neuronally-produced T may play a role in exercise-related neurogenesis in the hippocampus (31). Exogenous administration of T appears to have beneficial effects on cognitive function (i.e. memory and learning) and the electrophysiological and contractile properties of skeletal muscle that positively affect muscle performance (12). Given the established relationship between T and status seeking behaviors (28, 38) it is possible that transiently elevated levels of T during racing and other kinds of athletic competition elevate dominance motivation, and high levels of T may increase willingness to compete again after a defeat (6, 30).

We found no clear evidence of a relationship between salivary C or T and finish time. It is possible that transient changes in levels of C and/or T have an effect on athletic performance, but other factors including individual differences in fitness, nutrition, training load, sleep, stress, pain tolerance, and an athlete’s “mental toughness” may mask effects due to individual differences in hormone levels.

In any cross country race, there is only one individual winner – the one who crosses the finish line first. By this strict criterion, all the Emory runners lost their races. However, cross country racers more often assess performance relative to personal best times and expectations of placement based on the other individuals in the competition. By these more realistic standards, Emory racers, as is expected for any given race, had some relatively “good” and relatively “bad” performances. Despite this, virtually all racers showed an increase in T from before to after competition and these increases were not related to finish time.
As for other intercollegiate sports (16, 17), when saliva samples are obtained immediately after the finish of the contest, similar competition-related increases in C and T are seen in both winners and losers.

For intercollegiate cross country racers, the warm-up period for some athletes was associated with an increase in T (but not C) that, in effect, anticipated the competition to come. Further, this study demonstrates a clear increase in C and T associated with cross country racing, sport in which physical exertion is sustained without pause and competition is for finish place among many, rather than a coordinated team victory or defeat. As in the present study, endocrine effects of athletic competition have been most often demonstrated with elite athletes. The extent to which these effects may also be present in more purely recreational sporting contests remains to be determined. Sports-related elevations in C and T have been most commonly associated with competitions that require considerable physical exertion. There are, however, formal athletic competitions that involve relatively little physical exertion. The study of these sports would appear to be critical for understanding the relative contributions of physical and psychological elements to hormone changes associated with athletic competition.

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