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Title: The neuromuscular, biochemical, endocrine and mood responses to small-sided games training in professional soccer.

Running title: Responses to small-sided games in soccer

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Abstract

The 24h responses to small-sided games (SSG) soccer training were characterized. Professional soccer players (*n*=16) performed SSG's (4vs4 + goalkeepers; 6x7-min, 2-min inter-set recovery) with performance (peak-power output, PPO; jump height, JH), physiological (blood creatine kinase: CK, lactate; salivary testosterone, cortisol), and mood measures collected before (baseline), and after (immediately; 0h, +2h, +24h). For PPO and JH, possibly small-moderate reductions occurred at 0h (-1.1W·kg^{-1}; ±0.9W·kg^{-1}, -3.2cm; ±1.9cm, respectively), before returning to baseline at +2h (trivial) and declining thereafter (small-moderate effect) at +24h (-0.9W·kg^{-1}; ±0.8W·kg^{-1}, -2.5cm; ±1.2cm, respectively). Lactate increased at 0h (likely-large; +1.3mmol·L^{-1}; ±0.5mmol·L^{-1}), reduced at +2h (likely-small; -0.5mmol·L^{-1}; ±0.2mmol·L^{-1}), and returned to baseline at 24h (trivial). A very-likely small increase in CK occurred at 0h (+97u·L^{-1}; ±28u·L^{-1}), persisting for +24h (very-likely small; +94u·L^{-1}; ±49u·L^{-1}). Possibly-small increases in testosterone (+20pg·ml^{-1}; ±29pg·ml^{-1}) occurred at 0h, before likely-moderate declines at +2h (-61pg·ml^{-1}; ±21pg·ml^{-1}) returning to baseline at +24h (trivial). For cortisol, possibly-small decreases occurred at 0h (-0.09ug·dl^{-1}; -±0.16ug·dl^{-1}), before likely-large decreases at +2h (-0.39ug·dl^{-1}; ±0.12ug·dl^{-1}), which persisted for 24h (likely-small; -0.12ug·dl^{-1}; ±0.11ug·dl^{-1}). Mood was disturbed by SSG's at 0h (likely-moderate; +13.6AU, ±5.6AU) and +2h (likely-small; +7.9AU; ±5.0AU), before returning to baseline at +24h (trivial). The movement demands of SSG's result in a bimodal recovery pattern of neuromuscular function and perturbations in physiological responses and mood for up to 24h. Accordingly, when programming soccer training, SSG's should be periodized throughout the competitive week with submaximal technical/tactical activities.

Key Words: Fatigue, recovery, football, muscle damage, monitoring.

INTRODUCTION

Soccer is an intermittent sport which involves periods of high-intensity activity, interspersed with lower intensity actions, as well as technical and tactical components (3). Due to the complex multifaceted game demands, soccer players are required to train multiple physical qualities, including but not limited to: strength, power, speed, agility, aerobic capacity, repeated sprint ability, as well as technical and tactical training. As there is often limited training time between fixtures, a time efficient method of simultaneously developing these physical, technical and tactical qualities is desirable. This usually results in concurrent training methods, with multiple sessions often undertaken on the same day and within 24 hours of one and other. For the players to positively adapt to training, the stimulus should be applied in an order or a spacing that allows recovery to a point where they are able to meet the demands of the following training session (5). Therefore practitioners require an understanding of the physiological and psychological responses to each training stimulus.

Small sided games (SSG) are a popular training method utilized by coaches to optimize training time, as they are thought of as being able to replicate the demands of competition (7, 9, 21). Therefore, SSG's are used extensively to improve and maintain physical fitness, along with technical and tactical performance in professional soccer players. Previous attempts to characterize the internal and external loading of SSG's has been achieved via collection of heart rate, movement demands (i.e., global positioning system; GPS data), blood lactate, and rating of perceived exertion (RPE) responses (21). While studies have shown that manipulating variables such as the playing area, number of players, and the rules of the game can influence the acute physiological response (7, 9, 21), it is not well understood what impact SSG's may have in the hours and days that follow. A greater understanding of this would be of interest to those responsible for the design of soccer training programs, given the possible influence that this may have on additional training sessions performed within the week.

Previous research has examined the acute post exercise responses induced by strength (6, 19), speed (24), and endurance (15, 34) training. It is well known that any repeated eccentric or stretch shortening cycle actions, such as those used in soccer, are likely to induce muscle damage (16), muscle soreness (8) and reduce neuromuscular performance (33). Therefore, measures of neurmuscular function and markers of muscle damage are often used to assess fatigue and recovery from soccer specific exercise (31). In addition, the hormones testosterone and cortisol have previously been shown to respond to metabolic stress associated with these types of exercise (40, 42). More specifically, testosterone and cortisol have been shown to respond in opposite directions in response to metabolic stress, and the ratio between the two hormones has been reported as a balance of anabolic/ catabolic activy. Despite some authors suggesting these hormonal changes can effect acute performance, protein signalling and muscle glycogen synthesis (13, 18), the endocrine response to SSG activity has not been previously reported. In addition to objective markers, subjective cognitive measures such as athlete mood, subjective muscle soreness, stress and motivation are also widely used to assess fatigue and recovery in sports (26). The brief assessment of mood states questionnaire has been shown to be a reliable, valid and simple method of examining the dose-response relationship between exercise and fatigue (26, 39).

To date, there are no data on the magnitude of fatigue and the recovery time-course of any variable from SSG training sessions in soccer. Given the popularity of SSG's and that multiple training sessions are often programmed on consecutive days in soccer, a greater understanding of the response to SSG's may be of interest to those responsible for designing soccer training programs. Therefore, the aim of this study was to characterize the neuromuscular, endocrine, metabolic and mood response to a SSG session over 24 hours.

METHODS

Experimental Approach to the Problem

This observational study assessed the neuromuscular, endocrine, biochemical and mood responses to a SSG training session. The study took place at the end of the 2015 – 2016 competitive season with players being given two complete rest days before test involvement. Players were instructed to refrain from physical activity in the rest days and in their time away from the training ground. Countermovement jumps (CMJ; peak power output, PPO, and jump height, JH), bloods (creatine kinase; CK, and lactate concentrations), saliva (testosterone and cortisol concentrations), and a brief assessment of mood (BAM+) were collected before (baseline), and after (immediately; 0h, 2 hours; +2h, 24 hours; +24h) the session. Objective training loads from the SSG's were assessed using 10 Hz GPS devices and subjective RPE's were collected using Borg's CR10 scale.

Subjects

Data are presented from 16 male professional soccer players (age: 21 ± 2 years, mass: 74.8 ± 5 kg, height: 1.81 ± 0.06 m) who represent a Premier League under-23 soccer team. Despite the involvement of goalkeepers in the SSG protocol, only outfield players were included in the current study and they represented a range of playing positions. All players were considered healthy and injury-free at the time of the study and were in full-time training. Players were in the maintenance phase of their training season, undertaking resistance training programs, team-based conditioning sessions, and technical and tactical training. On a typical microcycle which consisted of 1 game per week, players were completing five on-field training sessions and two resistance training sessions. Ethical approval was granted by the ethics advisory board of Swansea University. Players were also informed of the risks and benefits and provided written informed consent prior to participation in the study.

Main Trial Procedures

On arrival at the training ground and before breakfast (~08:45 h), baseline salivary samples and BAM+ mood questionnaire scores were obtained. Players were then instructed to follow their normal breakfast routines and eat the food and drink prepared for them at the training facilities. After breakfast (~09:30 h), a capillary blood sample was taken and CMJ's were performed on a portable force platform. Prior to CMJ testing, players completed a 5-minute standardized warm up consisting of jogging and dynamic stretching. The SSG training session began at 10:30 h and individual player workload was monitored using GPS and RPE. Follow up measures (saliva, BAM+, blood & CMJ's) were collected at 0h, +2h and +24h post-training. Players consumed a nutritionally balanced lunch and drank water as normally provided at the training ground.

Small-Sided Games (SSG)

After a five-minute warm-up, which consisted of dynamic stretching and short sprints, players were split into four teams of five by coaching staff. The teams were organized such that playing positions were balanced within each team (e.g., one goalkeeper, one defender, one winger, one midfielder, and one striker) and teams were perceived to be of equal standard. The sport surface was a modern third generation artificial grass pitch and players wore their normal soccer boots during the SSG's. Players were instructed to play against another team for seven blocks of six minutes (overall work = 42 minutes) with two minutes between each game being allowed to drink water and passively rest before the next repetition.

Pitch size was 24 x 29 meters (width x length) and full-sized goals with goalkeepers were used. Further, players were allowed unlimited touches of the ball and the aim was to score as many goals as possible. This SSG format complemented the player's training regimes and was similar to previous literature (11, 27). The total time the participants were on the field, from the beginning of the warm-up to the end of the SSG's, was 59 minutes.

Countermovement Jump (CMJ) Testing

A portable force platform (Type 92866AA, Kistler) was used to measure performance of the lower body. This required CMJ's to be performed at maximum effort, with arms akimbo to isolate the lower body musculature. Two CMJ's were completed after a standardized warm-up at each time-point. The vertical ground reaction forces from the jumps were used to assess PPO from previously reported methods (32). This data was converted into relative peak power ($W \cdot kg^{-1}$) by dividing PPO by the player's body weight in kilograms. Additionally, JH was calculated by multiplying the velocity at each sampling point by the time (0.005 s). It was then defined as the difference between vertical displacement at take-off and maximal vertical displacement. Test-retest reliability (intraclass correlation coefficient) for PPO, and JH were 0.89 and 0.84, respectively. The coefficient of variation (CV) for PPO and JH were 2.3% and 3.2%, respectively.

Salivary Testosterone (T) and Cortisol (C) Assessments

At all time-points, 2 ml of saliva was collected by passive drool into sterile containers. Saliva samples were stored at -20 °C for seven days until assay. After thawing and centrifugation (2000 rpm x 10 minutes), the saliva samples were analyzed in duplicate for testosterone and cortisol concentrations using commercial kits (Salimetrics LLC, USA). The minimum detection limit for the testosterone assay was 6.1 pg.ml with an inter-assay CV of 5.8%. The cortisol assay had a detection limit of 0.12 ng.ml with inter-assay CV of 5.5%

Blood Creatine Kinase (CK) and Lactate Testing

After immersing the subjects hand in warm water, whole blood was collected via fingertip puncture using a spring-loaded disposable lancet (Safe-T-Pro Plus, Accu-Chek, Roche Diagnostics GmBH, Germany). First, a 5-µL sample of whole blood was taken for the immediate determination of lactate (Lactate Pro, Arkray, Japan). Next, a 300-µL sample was collected in a capillary tube and immediately centrifuged (Labofuge 400R, Kendro Laboratories, Germany) at 3000 revolutions$\cdot$min^{-1} for 10 min for the extraction of plasma, which was subsequently stored at -20 °C.

The plasma samples were left to thaw before 6-µL was used in the analysis of CK using a semi-automated analyser (ABX Pentra 400; ABX Diagnostics, Northampton, UK). Sample testing was carried out in duplicate and the mean CV for CK assays was 1.6%.

Mood Assessment

Mood state was assessed using a modified version of the brief assessment of mood questionnaire (BAM+). This 10-item questionnaire is based on the Profile of Mood State assessment, and consists of a scale where players mark on a 100-millimetre scale how they feel at that moment in time. Scale anchors ranged from 'not at all' to 'extremely'. The questions assess the following mood adjectives: anger, confusion, depression, fatigue, tension, alertness, confidence, muscle soreness, motivation and sleep quality. Players completed the questionnaires in isolation of teammates and it took approximately 2 minutes complete. The BAM+ questionnaire has been shown to be an effective tool for monitoring the fatigue and recovery cycles in elite athletes (39). Scores range from 0 – 100, with 0 indicating the best mood and 100 indicating the worst.

Ratings of Perceived Exertion (RPE)

Using Borg's CR10 scale, players were asked to give an RPE on a scale of 1 – 10. This question was asked verbally and in isolation from other team mates. These measures were obtained 10 minutes after the end of the SSG training session. RPE has been shown to have high correlations (r = 0.75–0.90) with heart rate based methods of training load (12), with this association being shown across various team sports (1, 11).

Time-motion Analysis

Time-motion analysis data was collected via 10 Hz GPS units embedded with 100 Hz tri-axial accelerometers (OptimEye X4, Catapult Innovations, Melbourne, Australia), which have shown to hold an acceptable level of reliability and validity when tracking player movements (25). Each unit was attached to the upper back of players using a specifically designed vest garment. The data was downloaded and processed automatically using Catapult Sports software (Openfield, Catapult Innovations, Melbourne, Australia). The high speed running threshold was defined as the total distance (m) covered at a velocity >5.5 $m \cdot s^{-1}$, and was set in line with previous work in soccer time-motion analysis (38, 41). Player load [Playerload^{TM}] is defined as the sum of gravitational forces on the accelerometer in each individual axial plane (anteroposterior, mediolateral and vertical), and has been shown to predict changes in CMJ performance and hormones following elite soccer match play (37).

Statistical Analysis

Data are reported as mean ± SD. Visual inspection of the residual plots revealed no clear evidence of heteroscedasticity, therefore we performed all analyses on the raw untransformed data. Separate mixed linear mixed models (SPSS v.21, Armonk, NY: IBM Corp) were used to examine the effect of SSG on our physical variables (total distance, high-speed running, and player load) and also on our fatigue marker responses (mood score, creatine kinase, peak power output, jump height, testosterone, cortisol, and blood lactate). For these models, SSG (1-6) and time point (baseline, 0, +2, and +24 hours), respectively were entered as the fixed effect. In both models, players were included as a random effect with random intercept to account for the hierarchical nature of our design (e.g. repeated measurements from the same players). Following this, a custom-made spreadsheet (22) was used to determine magnitude based inferences for all differences, with inferences based on standardized thresholds for small, moderate and large differences of 0.2, 0.6 and 1.2 of the pooled between-subject standard deviations (23). The chance of the difference being substantial or trivial was interpreted using the following scale: 25–75%, possibly; 75–95%, likely; 95–99.5%, very likely; >99.5%, most likely (4). The uncertainty in our estimates is expressed as 90% confidence limits (CL). We classified the magnitude of effects mechanistically, whereby if the 90% confidence limits overlapped the thresholds for the smallest worthwhile positive and negative effects the effect was deemed unclear (23).

RESULTS

Physical demands of SSG's

The GPS data for each SSG repetition, the difference between repetitions and the sum of all repetitions are presented in Table 1. The mean total distance covered during the SSG's (excluding rest periods) was 4388 ± 231 m. There were moderate or large reductions in total distance in all SSG's in comparison to SSG 1. All other changes in total distance between SSG's were small or trivial. The total high speed running distance accumulated during the SSG's was 41 ± 30 m. Similar to total distance, there were moderate or large reductions in high speed running in all SSG's in comparison to SSG 1. All other changes in high speed running between SSG's were small or trivial.

The total player load [Playerload™] accumulated over the SSG's was 483 ± 38 AU. Whilst no large between-SSG differences in Playerload™ were observed, there were moderate reductions in all SSG's in comparison to SSG 1. All other changes in Playerload™ between SSG's were small or trivial. The mean RPE reported for the 42 minutes of SSG's was 7.1 ± 1.3 arbitrary units (AU), which is classified as 'very hard' on the scale used.

********** INSERT TABLE 1 NEAR HERE **********

Impact of SSG's on Fatigue Markers

Mood Questionnaires

The absolute changes in mood scores across each time-point are presented in Table 2. Relative to baseline, there was an immediate disturbance in mood at 0h (likely moderate increase; +47.2%) which persisted at +2h (likely small; +27.4%) but not +24h where mood had returned to near baseline-values (trivial; +8.7%).

Biochemical Response

The time-course changes in blood lactate and CK concentrations are presented in Table 2. There was an immediate increase in lactate concentrations at 0h (likely large; +100.2%). In comparison to baseline a decrease was observed at +2h (likely small; -34.2%). Values were similar to baseline at +24h (trivial; +5.9%). There was an immediate elevation in CK at 0h (very likely small; +40.6%), which persisted at +2h (possibly moderate; +49.2%), and at +24h (very likely small; +39.2%).

Neuromuscular Function

Average force platform data for PPO and JH are presented in Table 2. We observed a bimodal recovery pattern for both PPO and JH. There was an immediate decrease in PPO at 0h (possibly small; -2.1%), which returned to near baseline values at +2h (trivial; +1.3%), before further impairment at +24h (possibly small; -1.7%). Similarly, JH was decreased at 0h (possibly moderate; -8.6%), which returned to near baseline values at +2h (trivial; +0.2%), before further impairment at +24h (likely small; -6.8%).

Hormonal Response

The average time-course changes in testosterone and cortisol are presented in Table 2. Testosterone was increased immediately at 0h (possibly small; +11.1%), before a reduction at +2h (likely moderate; -33.9%) and returning to near baseline at +24h (trivial; +1.2%). Cortisol was decreased at 0h (possibly small; 16.5%), with a further reduction at +2h (likely large; -71.8%), which remained below baseline at +24h (likely small; -21.3%).

********** INSERT TABLE 2 NEAR HERE **********

DISCUSSION

The primary aim of this study was to characterize the neuromuscular, biochemical, endocrine and mood response of professional soccer players following a SSG training session. Immediate disturbances in mood, JH, PPO and CK occurred following 42 min of SSG's, which in the case of JH and PPO had returned to pre-exercise values following a 2-hour passive recovery period. On the following morning (+24h), there was a secondary impairment in CMJ performance (PPO & JH), whilst disturbances in CK persisted but mood scores had returned to baseline values. This is the first study that profiles the 24h response to SSG training; findings that will be of interest to those responsible for designing and monitoring soccer specific training, especially given the possible influence that such acute changes have on subsequent training design and recovery strategies used throughout the training week.

The demands of the SSG training session were designed to replicate the workload players are exposed to during a typical training session. The mean total distance players completed over the six SSG's was 4388 ± 231 m, at an average intensity of 104 ± 5 $m \cdot min^{-1}$. This playing intensity is similar most other previous studies (1), despite the total distance being greater, which likely reflects the longer amount of time on the field (1). These demands resulted in the players subjectively rating the session as 'very hard' (RPE 7.1 ± 1.3 AU). Although a likely large increase in blood lactate immediately after completion of the SSG's was observed (Table 2), the magnitude of the lactate increases observed here are low in comparison to other SSG specific studies (11, 27). This difference occurred despite pitch size and game rules being similar (i.e 4 vs 4 plus goalkeepers), however it is hard to compare the external load of the present study to the previous studies mentioned, as they occurred before the introduction of GPS technology. This could be a result of differences in session volume and intensity, player training status or skill level as we present data from professional in-season soccer players who are more accustomed to this type of training. Notably, previous studies have reported data from younger elite players (<18 years old) and recreational athletes.

Whilst PPO (possibly small; -2.1%) and JH (possibly moderate; -8.6%) were immediately impaired, these markers had returned to baseline values after 2 hours of passive recovery. Mood scores in the current study presented a similar pattern, however were still higher (likely small; +27.4%) than baseline values at +2h. This would suggest that if multiple sessions are programmed in the same day (e.g., resistance training and SSG's) as is often the case in professional soccer, then they should be separated with at least 2 hours recovery time if additive effects of depressed mood and fatigue wish to be ameliorated. Furthermore, the likely large reduction in cortisol at +2h may be most noteworthy here given its modulating effect on testosterone (14). Whilst large impairments in CMJ performance have been consistently reported for more than 48 hours post soccer matches (31), the responses to SSG's in the current study saw small and moderate decreases in PPO and JH respectively. This may highlight the greater detrimental effect that volume of work has in comparison to intensity on neuromuscular function; SSG playing volume was 42 min vs match-play (> 90 min). Despite recovery of these variables at +2h, there was another impairment in PPO (possibly small; -1.7%) and JH (likely small; -6.8%) at +24h; perhaps suggesting that stretch-shortening cycle derived fatigue follows a bimodal recovery pattern as described by previous authors (16, 24). A likely explanation for the initial impairment in PPO and JH at 0h is a reduced functioning of the muscle fibre contractile mechanisms in the presence of metabolites (hydrogen ions, adenosine diphosphate, inorganic phosphate) accumulated during exercise (24). More specifically, this theory proposes that there is a decreased calcium ion release from the sarcoplasmic reticulum, resulting in less calcium ion binding to troponin and a negative influence on actin-myosin interactions during cross-bridge cycling (24).

It seems curious that PPO and JH had recovered at +2h, whilst CK and mood scores were still above baseline values. It may be that the recovery observed at +2h may have occurred before the inflammatory process had started, and was likely due the removal of the metabolites that were initially present. Taking this time-frame into account, it is hypothesized that the recovery in PPO and JH observed at +2h occurred prior to the initiation of the inflammatory response, and was most likely due to the removal of the metabolic by-products that had initially built up immediately after the SSG's (16). Addtionally, the secondary drop in PPO and JH observed at +24h may be related to the inflammatory process which is likely to be in process at this time point; supported by previous literature in soccer that suggests CK peaks between 24 – 48 h post match play (31).

The declines in PPO and JH at +24h may also have implications for training design. The current study supports previous research which has shown both jump and sprint performance to be depressed when muscle damage and soreness has been induced by training 24 hours prior (20). Given this, it may be advised to place explosive/maximal effort training relatively close together and practitioners may consider programming their training in an order that takes advantage of maintained neuromuscular performance. However, as there is no data on the implications of multiple training sessions performed on the same day in soccer players, further research is required into the effect of performing additional training in this window on muscle damage, neuromuscular fatigue, mood and recovery time. It is also suggested that performance in submaximal activities would appear to be unaffected at +24 h. Therefore, a strategy of alternating high intensity explosive training days containing multiple sessions with days emphasising submaximal technical/ tactical activities may take advantage of the observed pattern of neuromuscular performance.

The SSG's used in the current study may have resulted in possible small increases in testosterone and decreases in cortisol at 0h. Whilst this is the first study to report endocrine responses to SSG training, the lack of immediate response we present at 0h contrasts previous work in sprinting (35) and resistance training (10). As previous work has highlighted that metabolic accumulation is linked to post-exercise elevations of testosterone (29, 42) and cortisol (40), it may be that the comparitable lower lactate levels immediately post the training protocol in the current study may explain this. While testosterone and cortisol were both found to be likely largely reduced from baseline values when measured at +2h, these depressions are similar to the normal circadian variations previously reported in the literature (28). The hormonal changes observed in the current study may be explained by natural changes in the player's circadian rhythm, where testosterone and cortisol in men has been shown to peak in the early morning followed by progressive reduction (30-40 %) throughout the day (30). Therefore, it seems unlikely that these declines were a direct response to the training stimulus. However, the lack of non-exercise control data in the current study means that this cannot be confirmed.

Although we aknowledge that the current findings may reflect the charicteristics of the SSG format used, this is the first study to report the responses to this type of training over a 24 hour period. Additionally there are a number of limitations within this study, which should be noted. Firstly, the natural day to day variation in the fatigue markers we have used was not measured prior to conducting our study. Therefore it cannot be ruled out that some of the changes in markers were driven by this natural variation, as opposed to the SSG's. In addition, no heart rate data was collected during the SSG's to give a marker of internal training load, in combination with the external load (GPS variables). This data would also have been interesting to compare to previous research. Finally, it would have been interesting to compare the responses to this specific type of SSG format (4 vs 4 + goalkeepers) to other formats (i.e 2 vs 2, 6 vs 6, 8 vs 8 etc), and also to manipulate the playing area size. This could be an area for future research.

PRACTICAL APPLICATIONS

This study shows that 42 minutes of SSG's resulted in immediate small to moderate disturbances in muscle damage, neuromuscular performance, and mood. As soccer players are often required to concurrently train multiple physical qualities in the same day (i.e. strength and soccer), coaches and sports scientists should try to allow adequate recovery (> 2 hours) between physically demanding sessions. Additionally, consideration of the 24-hour fatigue response accumulated from SSG's should be considered when programming into the training week. It is suggested that performance in submaximal activities would appear to be unaffected at 24 hours post. Therefore, a strategy of alternating high intensity explosive training days containing multiple sessions with days emphasising submaximal technical/ tactical activities may be beneficial. In addition, it is advised that those responsible for the design of soccer training programs should allow adequate recovery time (> 24 hours) between SSG's and competitive matches.

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Table 1. Mean (± SD) physical variables across each SSG during the training session, along with qualitative infer between game number comparisons.

Variable	Game Number 1	2	3	4	5	6	Total	Qualitative inferences for effe
Total Distance (m)	797 ± 36	736 ± 48	714 ± 42	705 ± 73	730 ± 60	704 ± 48	4388 ± 231	***Large***: 1v4**(-92; ±29), 1v6** (- ***Moderate***: 1v5***(-67; ±31), 1v2 ***Small***: 2v4** (-31; ±37), 2v6** (±34), 2v3* (-22; ±30), 3v5* (+16 ***Trivial:*** 2v5 (-6; ±38), 3v4 (+9; ±
High Speed Running (m)	15 ± 10	4 ± 4	5 ± 6	7 ± 9	6 ± 9	4 ± 5	41 ± 30	***Large***: 1v2* (-11; ±5), 1v6* (-11; ***Moderate***: 1v3** (-10; ±6), 1v4* ***Small***: 2v4** (+3; ±4), 2v3* (+1; ±5), 4v6* (-3; ±5), 5v6* (-2; ±4) ***Trivial:*** 2v6 (0; ±5), 3v5 (+1; ±5)
Player Load (AU)	86 ± 6	81 ± 9	80 ± 6	79 ± 8	80 ± 10	77 ± 7	483 ± 38	***Moderate***: 1v6*** (-9; ±3), 1v4* (-6; ±4), ***Small***: 2v6** (-4; ±5), 3v6** (-3; ±4), 5v6* (-3; ±4) ***Trivial:*** 2v3 (-1; ±5), 2v5 (-1; ±5)

SD, standard deviation; SSG, small-sided game; CL, 90% confidence limits; AU, arbitrary units;

*25-75 %, possibly; **75-95 %, likely; ***95-99.5 %, very likely.

Table 2. Mean (± SD) fatigue marker responses across each time-point, along with mean differences and quali
magnitude for differences from baseline values.

	Time-point						
Variable	Baseline	0h	Mean difference from baseline; ±90% CL	*QI*	+2h	Mean difference from baseline; ±90% CL	*QI*
Mood Score (AU)	28.8 ± 14	42.4 ± 16.5	+13.6; ±5.6	*M***	36.7 ± 12.8	+7.9; ±5.0	*S***
Creatine Kinase ($\mu \cdot L^{-1}$)	239 ± 174	336 ± 214	+97; ±28	*S****	357 ± 195	+118; ±24	*M**
Peak Power Output ($W \cdot kg^{-1}$)	53.1 ± 4.8	52.0 ± 4.2	-1.1; ±0.9	*S**	53.8 ± 4.9	+0.7; ±0.8	*T*
Jump Height (cm)	37.1 ± 4.4	33.9 ± 6.5	-3.2; ±1.9	*M**	37.2 ± 4.6	+0.1; ±0.9	*T*
Testosterone ($pg \cdot ml^{-1}$)	181 ± 64	201 ± 71	+20; ±29	*S**	119 ± 41	-61; ±21	*M***
Cortisol ($\mu g \cdot dl^{-1}$)	0.54 ± 0.28	0.45 ± 0.29	-0.09; ±0.16	*S**	0.15 ± 0.06	-0.39; ±0.12	*L***
Blood Lactate ($mmol \cdot L^{-1}$)	1.3 ± 0.5	2.6 ± 1.1	+1.3; ±0.5	*L***	0.8 ± 0.2	-0.5; ±0.2	*S***

SD, standard deviation; CL, 90% confidence limits; AU, arbitrary units.

Qualitative Inferences *(QI)*: Trivial *(T)*; Small *(S)*; Moderate *(M)*; Large, *(L)*.

*25-75 %, possibly; **75-95 %, likely; ***95-99.5 %, very likely.