









Sleep, Musculoskeletal Injuries, and Hormonal Status in Different Training Stages in Adolescent **Athletes**

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Abstract

Objective This study aimed to compare sleep, musculoskeletal injuries, and testosterone and cortisol levels in different training stages in adolescent athletes, and to investigate the association between these variables.

Materials and Methods The study has a prospective cohort design, and the sample consisted of 19 adolescent track and field athletes. They were followed for six months, with evaluations done at three different training phases: preparatory, competitive, and post-competitive. In each phase, we evaluated their sleep through actigraphy for 10 days and testosterone and cortisol levels through saliva samples. The incidence of injuries was obtained through the physical therapy department.

Results In the post-competitive phase, the athletes presented a longer total sleep time (TST) than the competitive phase. Moreover, a shorter wakefulness after sleep onset was observed in the post-competitive and competitive phases compared with the preparatory phase. Furthermore, cortisol levels were higher in the preparatory phase than in the post-competition phase. On the other hand, no differences were observed in the incidence of injuries and testosterone, nor was there a correlation between the

Discussion Adolescent track and field athletes had a higher TST in the postcompetitive phase and more fragmented sleep in the pre-competitive and competitive phases. On the other hand, higher cortisol was observed in the preparatory phase, however, there were no correlations between sleep, hormones, and the incidence of injuries.

Keywords

- ► sleep quality
- athletic injuries
- ► testosterone
- cortisol
- ► sports training

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Introduction

Many athletes initiate their participation in competitions and engage in sport-specific training in adolescence. Monitoring training-induced psychobiological adaptations in young athletes is essential to increase longevity in sports participation. Additionally, monitoring training-related variables such as sleep, injuries, internal and external loads, and recovery is essential for adequate training prescription during a competitive season.^{2,3}

The risk of injuries in sports is multifactorial and includes nonlinear relationships between different factors such as athlete biomechanics, training characteristics, recovery, sport specificity, and physiological aspects.⁴ Sleep debt is related to injury risk, changes in mood, and athletes' physiology and performance.⁵ Sleep plays a fundamental role in recovery, cognition, energy restoration, and metabolic functions. Moreover, it has an important role in the development and maturation processes of adolescents.⁶ The American Academy of Sleep Medicine recommends that adolescents should sleep between 8 and 10 hours per night. However, factors such as a delayed circadian rhythm typical of puberty, school activities, use of electronic devices at night, caffeine intake, and training schedules can compromise sleep in adolescent athletes.8

Thus, adolescent track and field athletes, for example, have a total sleep time worse than recommended, 9 and the quantity and quality of sleep have been associated with musculoskeletal injuries. 10 In contrast, sleep complaints have been associated with a history of occurrence, recurrence, and severity of injuries in this population.⁹

In addition to sleep, Foster et al.¹¹ observed an association between excessive training load and the occurrence of injuries. When excessive training demands are associated with insufficient recovery, athletes may suffer preventable injuries or even experience overtraining syndrome. 12 Selfreported scales and hormonal measures can be used to quantify an athlete's response to training. There is evidence that hormonal concentration oscillates concomitantly with changes in training volume and intensity. The measurement of testosterone and cortisol concentrations and the calculation of the testosterone/cortisol (T/C) ratio have been used as endocrine stress biomarkers in the sports medicine context and a marker of the anabolic/catabolic balance. 13 Tyndall et al. observed increased cortisol and reduced testosterone levels in swimmers subjected to intensity and volume variations at different training phases. 14

Training loads are constantly adjusted during a season to promote optimized performance in target competitions.¹⁵ The preparatory phase is associated with the highest loads (i.e., training intensity and volume), whereas the competitive phase is associated with sport-specific stimuli. Thus, the individual monitoring of psychophysiological and performance adaptations at different stages of training can help practitioners implement strategies to prevent injuries, identify flaws in training planning, and take care of the athletes' physical and mental health.

Our objective was to compare the quantity and quality of sleep, musculoskeletal injuries, and the concentrations of testosterone and cortisol (and their ratio) in different training phases in adolescent athletes, and we also investigated whether there was a relationship between injury occurrence, cortisol and testosterone levels, with sleep quantity and quality.

Materials and Methods

Study Design

This was a prospective cohort study. The protocol was approved by the Research Ethics Committee of the authors' institution (64492016.8.0000.5149). After being informed about the study (objectives, procedures, and risks), the athletes were invited to participate, and those who accepted signed a written informed consent form.

Participants

The sample was obtained by convenience and 30 track and field athletes of both sexes, aged between 12 and 21 years considered adolescents according to the American Academy of Pediatrics¹⁶-were recruited at the Sports Training Center of the authors' institution. They were runners (i.e., 100, 200, and 400 m) and participated in competitions at local, regional, and/or national levels at least in the six months before data collection.

Procedures

This was a study with a 6-month follow-up. Initially, anthropometric data were collected. In female athletes, information about the date of last menstruation and whether they were using birth control pills were also collected. The athletes were followed through 6 months, and we monitored the occurrence of musculoskeletal injuries resulting from their sports practice. Injury data were collected from the Physical Therapy Department of the Sports Training Center. Athletes were evaluated by physical therapists and a questionnaire to describe the characteristics of these injuries was provided to them. Sleep and hormone concentrations were monitored at three specific time points (using the same procedures) that corresponded to different training phases, as described below:

Preparatory phase: from August to early October and corresponded to the preparation period for the competitions. According to the coaches, the training sessions had a high volume and intensity.

Competitive phase: from October to December. According to the coaches, the principle of volume versus intensity interdependence was emphasized, and the training volume was reduced while the intensity was kept high.

Post-Competitive phase: from December to January. According to the coaches, both the training volume and intensity were reduced, with sessions being characterized by regenerative and more recreational activities.

Actigraphy

Actigraphy has been considered the gold standard method to evaluate the sleep-wake cycle in the general population. 17 It is the less invasive way to objectively measure sleep quality and quantity, with minimal impact on athletes' sleep and training habits. The participants were instructed to wear an actigraph (Philips Respironics®, Andover, MA, USA) on the non-dominant wrist and to use it for 10 days, maintaining their usual sleep habits during this period. They received the actigraph on a Thursday to englobe two weekends. Simultaneously, the athletes filled out a sleep diary to facilitate the data analysis. The data was analyzed using the Action-W version 02 software (Ambulatory Monitoring Inc®, Andover, MA, USA). We extracted the following variables: time awake (TA), total sleep time (TST), sleep onset latency (SOL), sleep efficiency (SE), and wakefulness after sleep onset (WASO).

Hormonal Collection and Analysis

Hormonal collection for analysis of free testosterone, cortisol, and testosterone/cortisol (T/C) ratio was done through saliva samples collected during five consecutive days of training at 3:00 PM, 30 minutes before each training session. Samples were always collected at the same time of day in the three training phases to avoid circadian rhythm interference. The athletes were instructed not to feed or ingest fluids except water for 30 minutes and to remain at rest for one hour before collection. They were also instructed to avoid brushing their teeth at least two hours before collection and to perform gentle mouthwash immediately before collection. Salivette ® tubes with a synthetic swab for cortisol determination and a cotton swab for testosterone analysis were used. The athletes were instructed to remain for at least 60 seconds with the swab in their mouth stimulating salivation before returning it to the tube. 18 The collection of saliva samples in women was performed between the first to the 14th day of the menstrual cycle (follicular phase) following the same pattern of collection days and times for males.

After collection, the Salivette® tubes were centrifuged (3600 rpm) for 20 minutes at 4°C. ¹⁹ The salivary content was then removed, stored in sterile falcon tubes, and frozen at -20° C until posterior analysis. After the end of the three training phases, the samples were thawed and centrifuged again to remove any impurities that could be present. Free testosterone concentration was analyzed using the enzyme-linked immunosorbent assay (ELISA), with a detection limit ranging from 7.00 to 1500.00 ng/dL (0.24 to 52.05 nmol/L). Cortisol concentration analysis was performed using the electrochemiluminescence method, with a detection limit ranging from 1.5 nmol/L to 1750 nmol/L (0.054 to 63.4 μ g/dL). ²⁰

Musculoskeletal Injury Monitoring

To record musculoskeletal injuries, we used an adapted version of the questionnaire developed by Fuller et al., which was used in a consensus manuscript by the Fédération Internationale de Football Association (FIFA).²¹ The following information was recorded during the 6 months of monitoring: date, classification, location (i.e., body part), diagnosis, recurrences, mechanism, withdrawal duration from training and competition, physiotherapy treatment duration, and when the injury occurred (i.e., training or competition). We followed the injury definition by Fuller et al.: "any

physical complaint reported by an athlete that resulted from a training or competition, regardless of the need for medical attention or withdrawal from activities"²¹.

We asked the athletes to complete a form daily during the six months to record all the variables mentioned above. However, when the athletes reported an injury, they always underwent a physiotherapy evaluation in the Physical Therapy Department to exclude complaints that were not classified as injuries according to our definition (e.g., delayed onset muscle soreness).

Subjective Rating of Perceived Exertion (RPE)

RPE was obtained daily, 30 minutes after each training session of each trained day. Athletes indicated the level of difficulty experienced during the session, ranging from 0 (rest condition) to 10 (greatest physical effort). We calculated the internal training load by multiplying the RPE score (intensity) by the session duration expressed in minutes (volume) of every day, and the results were expressed in arbitrary units.

Total Recovery Quality (TQR) Scale

Before each training session, the athletes answered the question "How do you feel about your recovery?" to verify their perceived recovery related to the previous training session. The answer is based on a scale ranging from 6 (nothing recovered) to 20 (fully recovered).²²

Statistical Analysis

The Shapiro-Wilk test was performed to verify the data normality. Means and SDs were used to describe sleep characteristics. One-way analyses of variance (ANOVAs) with repeated measures were used to compare the variables between training phases. When statistically significant differences were found, Fisher's Least Significant Difference (LSD) post hoc test was performed to compare pairs of means. For data that did not present normal distribution, Friedman's ANOVAs on Ranks with repeated measures were performed, using Tukey's post hoc tests whenever applicable. The Spearman test was used to analyze the correlation between sleep, injury, and hormone levels. The significance level was set at $\alpha < 0.05$. The SPSS software (version 20.0, Chicago, Illinois, US) was used.

Results

Of the 30 athletes recruited, 19 (13 male and 6 female) were included in our sample. Eleven athletes who did not attend all three evaluations were excluded from our sample. The characteristics of our sample are detailed in **Table 1**. There were statistically significant differences in the body mass and height between boys and girls (p = 0.001 and p = 0.038, respectively), though no differences were observed for age, BMI, and training frequency. None of the female athletes used birth control pills during the study.

All the athletes combined presented 25 injuries and attended 55 physiotherapy sessions during the 6-month follow-up, where fourteen athletes suffered injuries and

Table 1 Sample characteristics

	Age (years)	Weight (kg)	Height (m)	BMI (kg/m²)	Weekly training frequency (days)
Total (n = 19)	16.8 ± 2.7	62.3 ± 8.3	1.72 ± 0.11	21.1 ± 1.4	4.6 ± 0.7
Males (n = 13)	17.0 ± 2.9	65.4 ± 8.3	1.75 ± 0.11	21.2 ± 1.6	4.4 ± 0.8
Females $(n=6)$	16.6 ± 2.2	55.6 ± 2.1	1.64 ± 0.05	20.6 ± 0.8	5.0 ± 0.0

Abbreviation: BMI, body mass index. Values are presented as means \pm SD.

five of them had no injuries. **Supplementary Table S1** (online only) describes individual data for injury variables. During the preparatory phase, the athletes presented the highest incidence of injuries, ¹² of which resulted in 41 physiotherapy sessions. In the competitive phase, 8 injuries and 8 sessions were recorded, whereas, during the post-competitive phase, the athletes suffered 5 injuries and attended 6 physiotherapy sessions. **Table 2** shows the characteristics of injuries, such as the need to withdraw from sports practice, the body part and side affected, and the mechanism of injuries (overuse or trauma). It is noteworthy that most of the athletes (92%) did not need to withdraw

from training due to injury. The most injured body regions were the thigh (28%) and ankle (28%), and the most common injury mechanism was muscle overuse (92%).

Table 3 shows the comparison of sleep variables, perceived recovery and exertion, and injuries between the three training phases. In the post-competitive phase, the TST $(7.5\pm0.9\,\mathrm{hour})$ was statistically higher than in the competitive phase $(6.9\pm0.7\,\mathrm{hour})$ (p=0.01); and TA was statistically lower in the competitive (15.9 ± 1.1) and post-competitive (15.4 ± 1.0) phases (p=0.004). In addition, the athletes presented lower WASO values in the competitive $(29.2\pm9.9\,\mathrm{minute})$ and post-competitive $(36.1\pm15.2\,\mathrm{minute})$ phases

Table 2 Injury characteristics during the whole period evaluated and according to the training phase

Injury characteristics	Whole period	Preparatory Phase	Competitive Phase	Post-Competitive
Withdrawal from sports practice				
Yes	1	1	0	0
Partial	1	1	0	0
No (only physical therapy treatment)	23	10	8	5
Body part				
Lumbar/sacrum/pelvis	4	2	2	0
Shoulder/collarbone	2	0	2	0
Handle	1	0	0	1
Hip/groin	1	1	0	0
Thigh	7	4	2	1
Knee	1	1	0	0
Achilles tendon/ Calf	7	3	2	2
Ankle	2	1	0	1
Body side				
Dominant	10	6	1	3
Non-dominant	3	1	1	1
Bilateral	7	3	3	1
Does not apply	5	2	3	0
Injury mechanism				
Stress fracture	1	1	0	0
Ligament sprain/injury	1	0	0	1
Muscle strain/tension/injury/cramp	22	10	8	4
Tendon injury/rupture or tendinosis	1	1	0	0
Over-use	23	11	8	4
Trauma	2	1	0	1

Values are presented as absolute frequency.

Table 3 Comparison of sleep variables, perceived recovery and exertion, and injury incidence during ten days of actigraph use in three different training phases (n = 19)

	Preparatory Phase	Competitive Phase	Post-Competitive	<i>P</i> -value
TA (h)	15.9 ± 1.1	16.3 ± 1.1	$15.4 \pm 1.0^*$	0.004
TST (h)	7.2 ± 0.7	6.9 ± 0.7	$7.5 \pm 0.9^*$	0.012
SOL (min)	20.8 ± 10.4	24.1 ± 13.5	18.4 ± 13.0	0.201
WASO (min)	46.0 ± 12.4	29.2 ± 9.9 #	36.1 ± 15.2 #	0.001
SE (%)	82.5 ± 3.6	83.2 ± 6.5	84.2 ± 4.9	0.447
TRQ	5.55 ± 0.77	5.36 ± 0.63	5.66 ± 0.64	0.360
Internal Load	492.4 ± 88.8	447.6 ± 61.5	495.7 ± 148.3	0.343

Abbreviations: h, hours; min, minutes; SE, sleep efficiency; SOL, sleep onset latency; TA, time awake; TRQ, total recovery quality; TST, total sleep time; WASO, wakefulness after sleep onset.

compared with the preparatory ($46.0 \pm 12.4 \, \text{minute}$) phase (p = 0.00). There were no significant differences in the other variables analyzed.

► Table 4 shows the comparison of hormone levels between the three training phases. We only found statistically significant differences in cortisol levels. There was a difference without stratification by sex (p = 0.01) and among male athletes (p = 0.04). Post hoc analyses showed that cortisol levels were higher in the preparatory phase than in the postcompetitive phase (p = 0.02, all samples; p = 0.03, males). No differences were found for testosterone levels and T/C ratio.

No significant correlation was found between sleep, hormone levels, and incidence of injuries in the different phases of training (p > 0.05).

Discussion

This study aimed to compare the quantity and quality of sleep, the incidence of musculoskeletal injuries, and the

concentrations of testosterone and cortisol in different training stages in adolescent track and field athletes. The results showed that the mean TST and SE were below the recommended (i.e., < 8h and 85% respectively) in all phases, and SOL and WASO were within the recommended $(\geq 30 \text{ minutes and } 20 \text{ minutes respectively}).^{23} \text{ Moreover, the}$ athletes presented higher TST and lower WASO in the postcompetitive phase, compared with the competitive and preparatory phases, respectively. These findings corroborate previous studies showing that athletes, on average, sleep less than the recommended.²⁴ We found a mean TST of 7h. Previous studies have shown that athletes who experience sleep restriction may exhibit motor and cognitive deficits, altered mood, increased reaction time, and fatigue.²⁵

The athletes' sleep quality can be influenced by sports schedules. Previous studies have found a poorer sleep quality in periods close to competition, ²⁶ corroborating our findings. In the present study, a higher TST was observed in the post-competition compared with the competitive phase, in

Table 4 Comparison of hormonal variables between the three training phases in boys and girls

	Preparatory Phase	Competitive Phase	Post-Competitive Phase	Р		
Cortisol (nmol/L)						
All (n = 19)	1.99 ± 0,61	2.42 ± 1,12	2.81 ± 1.60	0.01*		
Males (n = 13)	1.98 ± 0.59	2.36 ± 1.18	2.81 ± 1.39	0.04*		
Females (n = 6)	2.01 ± 0.61	2.53 ± 1.08	2.79 ± 2.15	0.38		
Testosterone (pg/mL)						
All (n = 19)	67.25 ± 39.85	57.94 ± 31.68	56.86 ± 30,53	0.30		
Males (n = 13)	87.19 ± 31.34	69.86 ± 31.64	72.08 ± 23.27	0.19		
Females (n = 6)	24.05 ± 9.21	32.12 ± 6.53	23.89 ± 12.25	0.32		
Testosterone/Cortisol Ratio						
All (n = 19)	0.10 ± 0,06	$0.08 \pm 0,05$	0.08 ± 0.06	0.09		
Males (n = 13)	0.13 ± 0.05	0.10 ± 0.05	0.10 ± 0.05	0.09		
Females (n = 6)	0.04 ± 0.02	0.04 ± 0.01	0.03 ± 0.02	0.76		

Values are presented as means $\pm\,\mathrm{SD}.$

Values are presented as means \pm SD.

^{*}Significantly differs from the Competitive Phase.

^{*}Significantly differs from the Preparatory Phase.

^{* =} statistically significant.

addition to higher WASO in the preparatory phase. However, the increase in TST in the post-competition phase still does not correspond to what is recommended for adolescents.²³ The reduction in sleep quantity and quality in periods of preparation and competition can be explained by factors such as anxiety related to competitions, long travel routines, and training schedules.²⁷ It is common for athletes to present sleep restriction conditions, mainly due to the higher intensity and volume of training.²⁸ Silva et al.²⁹ evaluated 146 Olympic athletes from the Brazilian national team, during the preparatory phase for the RIO 2016 Games and recorded a total of 250 sleep complaints. Furthermore, the athlete and adolescent population is subject to specific demands that combine school and sports calendars, which can have an impact on sleep variables.³⁰ It was also observed by Oliveira et al. that the practice of physical exercise in adolescents and a physically active life may have positive effects on sleep patterns, corroborating the findings of this study.³¹

Regarding the internal training load, no statistically significant differences were observed between the training phases. This may be related to why there were no significant variations in testosterone and T/C ratio. Furthermore, the lack of hormonal variation may indicate that athletes did not have poor adaptation to training loads during the season, or that the training load and volume imposed on athletes were not high enough to generate variations in these variables.^{32,33}

Training loads, depending on the dose, can disrupt muscle homeostasis and cause a series of physiological responses generating strength and function. During strenuous exercises, there is a great caloric expenditure, with high production of ATP through metabolic pathways, where glycogen depletion has been related to muscle fatigue during exercise.³⁴ In addition, muscle microlesions may occur from the high demand of training and repeated muscle contractions and mechanical loads on the myofibrils during exercise.³⁵ In the present study, injuries occurred in all three evaluation phases; however, we did not find statistically significant differences between the three phases. The most injured sites were the thighs and ankles, and most injuries were due to overuse. These findings corroborate Edouard et al. 36 who found a higher injury incidence in thighs and feet during an international athletics championship.³⁶

Salivary cortisol is recommended and widely used as a training stress index. 13 The cortisol levels in our sample were within the range expected for athletes (1.8-19.9 nmol. L-1), but it increased as the season progressed and the stress and chronic load of training and competitions. The increase in cortisol levels induced by training stress was shown to be lower in trained individuals than in untrained individuals,³⁷ which may explain the little hormonal variation in the present sample between training phases.

Testosterone is an anabolic hormone that has functions such as tissue repair, and muscle tissue growth and is related to athletes' motivation.¹³ We did not find differences in testosterone concentration between training phases. It was expected that testosterone levels would be higher in the competitive period, given that adequate hormonal signaling is essential for physical adaptations, and low testosterone levels are associated with decreased performance, energy, and strength. 13 Thus, as there were no significant changes, there was also no variation in the T/C ratio and this is a positive point, since its prolonged decrease in the T/C ratio may be associated with losses in sports performance due to proteolysis and a decrease in protein synthesis.¹³

Some limitations of the present study should be mentioned. We had a relatively small sample size and a large sample loss, reflecting the difficulties in objectively monitoring sleep in adolescent elite athletes. Also, we did not control the training schedule and parameters in the three different phases; however, we tried to mitigate it by evaluating the internal load in each phase. Thus, more studies with larger sample sizes and control of training parameters should be performed.

We conclude that adolescent athletes presented a higher TST and lower WASO in the post-competitive phase compared with the competitive and preparatory phases, respectively. However, cortisol was higher in the preparatory phase. Despite this, no differences were observed in the other sleep variables, testosterone, and incidence of musculoskeletal injuries, nor were the variables correlated.

The present study suggests that there is a need to encourage better sleep habits, especially in the pre-competitive and competitive phases in adolescent athletes. In addition, it seems that in adolescent track and field athletes, monitoring with biomarkers was not correlated with the incidence of injuries, and, therefore, we suggest that new methods be used to seek to investigate the relationship of injuries with other variables related to training in this population.

Data Availability Statement

The datasets generated during and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Ethics Approval Statement

The study was approved by the Research Ethics Committee of the Federal University of Minas Gerais (n° 27518619.4.0000.5149).

Patient Consent Statement

All participants signed a Consent Form.

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Conflict of Interest Disclosure

The authors certify no conflict of interest.

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