

Monitoring training and match load in soccer players: Implications for individual and team performance

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Resumo

Esta dissertação explorou vários aspetos do desempenho no futebol, assim como fatores que contribuem para a fadiga em jogadores de futebol e o desenvolvimento de estratégias para gerir a fadiga para otimizar o desempenho do jogador. Desta forma, investigamos as funções dos padrões de sono e da atividade autonómica cardíaca noturna. Por sua vez, investigamos a variabilidade do sono e da frequência cardíaca (VFC) de jogadores de futebol de elite durante um período de nove semanas na época. Doze participantes do sexo masculino (idade: $24,2 \pm 4,9$ anos) foram incluídos no estudo. Os índices de sono dos participantes foram medidos por meio de actigrafia de pulso, enquanto monitores de frequência cardíaca foram usados para medir a VFC. As cargas de treino e jogo foram caracterizadas por meio da avaliação da percepção subjetiva de esforço de cada sessão (s-RPE), carga do jogador (PL) e da distância total. Jogadores que jogaram mais de 60 minutos tiveram mais oscilações na VFC e nos índices de sono do que aqueles que jogaram menos de 60 minutos. O tempo total de sono ficou abaixo dos níveis recomendados por duas semanas consecutivas para este grupo. Encontramos uma correlação moderada entre s-RPE e duração do sono, indicando que os jogadores que perceberam maior carga de treino/jogo dormiram menos. Houve também uma pequena correlação entre CJ e logaritmo natural de baixa frequência (lnLF), indicando que cargas mais altas dos jogadores estavam associadas a menor VFC. Finalmente, foi encontrada uma correlação moderada entre a eficiência do sono e o logaritmo natural da raiz quadrada da média da soma dos quadrados das diferenças entre intervalos normais a normais sucessivos (lnRMSSD), sugerindo que uma melhor eficiência do sono estava associada a uma maior VFC. Os resultados sugerem que os jogadores tiveram um padrão de sono abaixo do ideal durante este período e a carga de treino/jogo impactou a VFC e os índices de sono. As descobertas ajudam os treinadores no desenvolvimento de programas de treino individualizados para atender às exigências de cada jogador nos treinos e jogos.

Palavras-chave: ACELERÓMETRO; ATIVIDADE AUTONÓMICA CARDÍACA NOTURNA; RECUPERAÇÃO; MONITORAMENTO; DESEMPENHO.

Abstract

This dissertation explored various aspects of soccer performance, as well as factors contributing to fatigue in soccer players and develop strategies for managing fatigue to optimize player performance. In this way, we investigated the roles of sleep patterns and nocturnal cardiac autonomic activity. In turn, we investigated sleep and heart rate variability (HRV) of elite soccer players during a nine-week in-season period. Twelve male participants (age: 24.2 ± 4.9 years) were included in the study. Participants' sleep indices were measured using wrist actigraphy, while heart rate monitors were used to measure HRV. Training and match loads were characterised using the session rating of perceived exertion (s-RPE), player load (PL), and total distance. Players who played more than 60 minutes had more oscillations in HRV and sleep indices than those who played less than 60 minutes. Total sleep time was below the recommended levels for two consecutive weeks for this group. We found a *moderate* correlation between s-RPE and sleep duration, indicating that players who perceived higher training/match load slept less. There was also a *small* correlation between PL and natural logarithm of low frequency (lnLF), indicating that higher player loads were associated with lower HRV. Finally, a *moderate* correlation was found between sleep efficiency and natural logarithm of the square root of the mean of the sum of the squares of differences between successive normal-to-normal intervals (lnRMSSD), suggesting that better sleep efficiency was associated with higher HRV. Findings suggests that players had a suboptimal sleep pattern during this period and training/match load impacted HRV and sleep indices. Findings help coaches in developing individualized training programs to meet the demands of each player in training and match.

Keywords: ACCELEROMETER; NOCTURNAL CARDIAC AUTONOMIC ACTIVITY; RECOVERY; MONITORING; PERFORMANCE.

List of Abbreviations

ANS	Autonomic nervous system
HF	High frequency
HR	Heart rate
HRV	Heart rate variability
LF	Low frequency
Ln	Natural logarithm
LF/HF	Ratio between low frequency and high frequency
PSG	Polysomnography
RPE	Rate of perceived exertion
RMSSD	Square root of the mean of the sum of the squares of differences between successive normal-to-normal intervals
R-R	Normal-to-normal
SE	Sleep efficiency
s-RPE	Session-rate of perceived exertion
SWSE	Slow-wave sleep episode
TST	Total sleep time

Chapter I

Introduction and rationale

1. General introduction

Nowadays, the multiplicity of aspects that contribute to the high performance of soccer teams, leads us to a constant reflection on the range of problems of a methodological nature that training entails. In the sporting context, training load monitoring is crucial in the preparation of soccer players, as it provides coaches with information about individual match and training requirements to optimize physical fitness and reduce the potential risk of injuries (Jaspers et al., 2017). This becomes important during congested schedules, as players are more prone to injuries (Gurau et al., 2023). Thus, it becomes essential to explore sensitive and non-invasive means to control sleep patterns in athletes, with the aim of providing better sleep hygiene and, consequently, better recovery (Silva et al., 2022). In this scenario, it is necessary to optimize recovery to reduce the risk of transitioning to a state of over-fatigue, as well as to reduce the potential risk of injury.

This dissertation aims can contribute to a deeper understanding of various aspects of soccer performance and inform the development of evidence-based strategies for optimizing training and performance in soccer. Concretely, we pretend to examine the factors contributing to fatigue in soccer players, including match frequency and training intensity (though internal and external training and match loads), as well as develop strategies for managing and mitigating fatigue to optimize player performance. For such, it was necessary to evaluate the impact of factors like competition schedules, player fitness levels on training periodization, time of playing per match and develop methods for monitoring and quantifying the training and match load experienced by soccer players.

The article aimed to investigate the impact of training sessions and official matches, over 9 weeks, on cardiac autonomic activity and sleep patterns in elite soccer players, during the competitive season. The study investigated the sensitivity of nocturnal heart rate variability (HRV) to assess the effects of training sessions on cardiac autonomic function in elite soccer players. It should be noted the consideration for the importance of the individualized approach in the analysis of sleep and nocturnal HRV after training and match loads, were carried out in

twelve elite players, focusing specifically on the individual and group analysis of HRV and sleep patterns.

1.1. General characterization of soccer

Collective sports have their roots in ancient societies where different matches were an essential component of each civilization's culture (Weston et al., 2011). Soccer is a team sport with traits that give it a unique identity. Despite appearing straightforward, the core logic of a soccer match comprises a wide variety of physical, tactical-technical, and psychological elements that require adequate development to achieve excellent performance in a competition (Sebastiá-Rico et al., 2023).

Moreover, a player's physical activity fluctuates throughout a soccer match, ranging from a series of sprints to a recuperation phase where the player frequently walks or even stands still (Julian et al., 2021). These rest periods can occur at any time because of a variety of variables that are directly tied to the match's dynamics, execution speed, playing surface or even the tactical mission that each player has during a match. According to Dos'Santos et al. (2022), physical demands may lead to certain positions and tasks to call for physiological characteristics and different training approaches. The anthropometric and physiological profiles of players in various positions serve as evidence for this. However, due to the features of this modality, it is difficult to characterize the physiological makeup of soccer. Scientifically supported interpretations have gradually changed this subjectivity, inherent to soccer's experience and analysis.

1.2. Activity profile and physiological demands of soccer

Elite soccer is characterized to its non-cyclical nature and intensity, which its classified as a high intermittent sport. During a match, players must have both aerobic and anaerobic capacity to cope with the physical demands of the match and to support their technical and tactical skills (Oliva-Lozano et al., 2020). Practitioners must be aware of the physiological profiles of professional soccer players in order to assist them in developing the physiological skills required to enhance performance (Ribeiro et al., 2021).

In order to increase different fitness components and prepare players to meet the demands of a match in all its facets, professional soccer training places a high stress level on the physiological systems (Thorpe et al., 2017). Soccer is an example of an intermittent workout that, in the context of endurance, incorporates periods of high-intensity activity and calls for both aerobic and anaerobic energy systems (Moalla et al., 2016). During soccer matches, aerobic glycolysis serves as the main energy source, with maximum rate of oxygen consumption (VO_{2max}) levels averaging 75–80% (Dolci et al., 2020). The maximum oxygen uptake for current high-level soccer players is thought to range between 60 and 70 ml.kg⁻¹.min⁻¹, with an estimated minimum of 65 ml.kg⁻¹.min⁻¹ for top soccer players (Slimani et al., 2019). However, VO_{2max} values for soccer players were observed to vary greatly. Maximal oxygen consumption varies slightly by position, with midfielders and wider defensive players having greater VO_{2max} values than other outfield players (Dolci et al., 2020).

Most methods used to assess and convert VO_{2max} in soccer are calculated and converted by a relationship between the HR and VO_2 during treadmill running (Djaoui et al., 2017). It is important to note that direct measurements of VO_2 during match play are not accurate. During a match, elite players cover approximately 10-12 km at an average intensity near the anaerobic threshold, with 80–90% of maximal heart rate (Giminiani & Visca, 2017). The variations in work-rate profiles are related to variations in positional responsibilities, environmental factors, playing styles, and player nutrition (O'Donoghue, 2022). Nevertheless, it has repeatedly been noted that midfielders cover more ground than any other player on the squad (Baptista et al., 2018). The relative distance covered by outfield players in various activity patterns was reported outfield players during a match consists of 58.2 - 69.4% walking and jogging, 13.4 - 16.3% low speed running, 12.3 - 17.5% moderate-speed running, 3.9 - 6.1% high-speed running and 2.1–3.7% sprinting (Di Salvo et al., 2007).

Accurate measurement of both internal and external match demands is necessary to successfully evaluate the physical demands placed on soccer players (Gaudino et al., 2015). In order to facilitate this data collection, technology and sport have developed a solid relationship over the past few years.

Technology advancements have allowed researchers and professionals to better understand of the pressures exerted on athletes to enhance their performance, well-being, and ability to avoid injuries during training and competition (Seshadri et al., 2019). As a result, load monitoring may be established with a real-world application to sports performance and has consequently become a crucial component of an athlete management system (Gaudino et al., 2015).

The three most common ways to measure load are from global positioning systems (GPS) (Ravé et al., 2020), session-rate of perceived exertion (s-RPE) (Rodríguez-Marroyo et al., 2012), and heart rate (HR) monitors (Schneider et al., 2018). The player external load is frequently assessed using a GPS device, frequently with an accelerometer included (Strauss et al., 2019). In order to extrapolate training load, s-RPE, a commonly used indicator of internal load that is simply given by the athlete, has been multiplied by the length of the training session (sRPE-TL) (Askow et al., 2021).

During a soccer match, the cardiovascular effort is relatively high and varies amongst players (Hostrup & Bangsbo, 2023). HR measurement is frequently employed as an indirect indicator of exercise intensity. It is a methodological advantage of most available HR monitors which allows one to measure and store the HR values with a high reliability (Dellal et al., 2012). Physiologically, HR measurements present a high relationship with physiological variables such as VO_{2max} in intermittent activities (Macinnis & Gibala, 2017). Current radio telemetry systems allow monitoring HR during short time intervals simultaneously on all team players, improving the planning and control of training (Hostrup et al., 2019).

1.3. Fatigue in soccer

Soccer involves many physically demanding activities, including sprinting, changes in running speed, changes of direction, jumps and tackles, as well as technical actions such as dribbling, passing, and shooting (Brownstein et al., 2017). These activities lead to post-match fatigue, mainly linked to a combination of glycogen depletion, muscle damage, and mental fatigue (Marqués-Jiménez et al., 2017). In elite soccer, recovery strategies are required during off-season and

in-season to alleviate post-match fatigue, regain performance levels, and reduce the potential injury risk (Silva et al., 2018).

The stress associated with training and competition often temporarily impairs players' physical performance. This impairment may be acute, lasting minutes or hours, and may stem from metabolic disturbances and substrate utilization associated with high-intensity exercise (Marqués-Jiménez et al., 2017). Alternatively, exercise-induced muscle injury and delayed-onset muscle soreness that often follow training with a high eccentric component may lead to impairment lasting several days (Peake et al., 2017). Increasing attention in the literature has centred on evaluating the effectiveness of a range of monitoring tools that may serve as valid indicators of recovery status in athletes, including heart-rate derived indices, salivary hormones, neuromuscular indices, and subjective wellness scales (Flatt et al., 2018).

A valid marker of recovery should be sensitive to training and match load variability. Consequently, research has evaluated the sensitivity of monitoring tools in response to changes in training load over extended periods, such as weekly and/or monthly, in sports such as soccer (Brito et al., 2016). In contrast, limited effort has been made to determine the effectiveness of these tools for monitoring recovery in elite team sport players. Team sport athletes compete on a weekly or biweekly basis, so decisions on player wellness and fatigue are frequently required over extended periods (Calleja-González et al., 2021). Under such conditions, monitoring tools more sensitive, such as daily load fluctuations, may be the most effective. Since differences exist in the physiological demands between team sports, it is important to determine which fatigue variables are most sensitive to changes in load associated with specific sports (Coyne et al., 2022).

Furthermore, athletes' training adaptations are typically correlated with the training stimuli experienced during the various training cycles (Skorski et al., 2019). An appropriate training dose at the individual level may allow for the best improvements in fitness and performance, whereas too much or too little training load may result in accumulated fatigue, such as overreaching and/or overtraining (Lorenz & Morrison, 2015). It is, therefore, paramount to monitor athletes' fatigue, fitness and/or performance responses to the various training phases so that

training load and contents can be adjusted and individualized during and between each training cycle (Saw et al., 2015).

Notably, data may be helpful in choosing a team and identifying athletes who are prepared for the demands of competition (Torres-Ronda et al., 2022). Communication and relationship-building with athletes, helpers, and coaches also have several advantages. When athletes are involved in monitoring, it can strengthen their sense of ownership, empower them, and increase their sense of involvement in the training program (Hamlin et al., 2019). The support staff and coaching staff's ability to communicate with one another can be improved with the help of data gathered from training monitoring. Together, these advantages may raise the training program's associated belief and confidence levels. In order to increase their performance, athletes must modify their training loads, particularly by boosting their frequency, duration, and intensity (Skorski et al., 2019). Training loads are altered at various points during the training cycle to either increase or decrease fatigue, depending on the training phase.

The correct titration of fatigue is essential for both training adaptations and competition performance. Modern coaching practice accepts the accumulative fatigue brought on by repeated overload training and/or frequent competition (Lorenz & Morrison, 2015).

1.4. Overtraining and overreaching

Athletes commonly employ the strategy of intensifying their training to improve their performance. However, this can lead to the athlete feeling extremely tired and experiencing a decline in performance after a single intense training session (Gabbet, 2016). Similar or heightened symptoms may persist even after the intensified training period ends, and this state is known as overreaching. Overtraining is believed to occur due to continuous training or non-training stress (Armstrong et al., 2022). The initial stage of overtraining syndrome development is referred to as functional overreaching (Kreher & Schwartz, 2012). Functional overreaching (FOR) is characterized by a decline in performance and emotional state, but these decrements can be mitigated and reversed through sufficient rest within a two-week timeframe (Bellinger, 2020). If an athlete fails to restore their pre-overreached performance within two weeks, they progress to a

phase known as non-functional overreaching (NFO). Continued training beyond the NFO phase leads to further performance deterioration and more severe declines in psychological well-being, immune function, and other manifestations (Kreher, 2016).

Unfortunately, there is limited scientific research on overtraining, and most studies focus on overreaching due to ethical concerns. It is generally believed that overtraining causes more severe symptoms (e.g., performance decline and mood disturbances) than overreaching (Bell et al., 2022). However, there is no scientific evidence to confirm or refute this notion. Currently, there are no objective diagnostic tools to identify an athlete as overtrained. Therefore, diagnosis relies on excluding other potential factors affecting performance and mood. As a result, overtraining can only be diagnosed when no other explanations are found (Carrard et al., 2021). On the other hand, overreaching refers to a short-term decrement in performance capacity, with the physiological and biochemical responses to increased training vary significantly and some measures showing changes and others remaining unchanged (Bellinger, 2020).

The distinction between overtraining and overreaching lies mostly in the time required for performance restoration rather than the type or duration of training stress or the degree of impairment. However, no evidence suggests that the training leading to these conditions is identical in nature and extent (Armstrong et al., 2022). Anecdotal suggestions propose that overreaching may be more common in team and explosive/power sports, while overtraining may be more prevalent in endurance sports (Carrard et al., 2021). Although these definitions are not entirely satisfactory, they are commonly cited in the literature as the most accurate descriptions of the conditions.

Intensified training leading to overreaching and/or overtraining is often regarded as a continuum, where increased stress disrupts homeostasis and temporarily reduces function, which can arise from training, psychological stress, or illness (Bell et al., 2022). If appropriate recovery is allowed, acute fatigue resulting from this disruption can lead to positive adaptations and performance improvements (Greenwood, 2008). This normal training response, characterized by progressively increasing training load followed by sufficient recovery, forms

the basis of effective training programs. However, if the balance between training stress and recovery is disturbed, an abnormal training response may occur, resulting in overreaching (Armstrong et al., 2022).

Therefore, careful monitoring is necessary during training blocks aimed at inducing overreaching to prevent progression to the NFO phase. Given the difficulties in precisely defining these conditions, considering them on a continuum may be more appropriate, with each definition representing different points based on individual physiological and psychological factors (Taylor, 2012).

1.5. Factors influencing training periodization in soccer

It can be challenging to incorporate a periodized training program into team sports because the annual competition training program, which can last for many consecutive weeks, must incorporate a number of different training goals (Lyakh et al., 2016). The need to incorporate technical and tactical training drills include maximum strength, explosive power, metabolic conditioning, hypertrophy, reduction of the potential risk of injury, and recovery strategies (Marín et al., 2017). There is a need for planned variations in the training program to systematically shift in emphasis by promoting these various training effects at different times of the season. These elements suggest that the organization of the exercise prescription should be multidimensional and tailored specifically to soccer (Thorpe et al., 2017).

The micro cycle is organized by the volume, intensity, focus, and methods of the overall training phase, all of which relate to the ultimate performance goal (Mujika et al., 2018). According to the distribution of competitive matches, which is frequently one match every 3–4 days, micro cycles in elite soccer are typically between 3 and 7 days long (Oliveira et al., 2020). Additionally, factors such as travel requirements to domestic and international matches, the head coach's philosophy, individual player requirements, and national team commitments can further influence the weekly training load prescription (Brito et al., 2016). In order to effectively train elite soccer players, a structured approach is needed that allows for variations in training loads over short time frames while also facilitating adaptation (Mujika et al., 2018). This will ensure that the load-recovery cycle is optimally balanced. It is noticeable that the training loads during the weekly micro

cycle phase must be adjusted according to the upcoming fixtures for players to perform at their peak during matches (Lyakh et al., 2016).

Coaches may find it challenging to implement a structured training schedule that aids in post-competition recovery while incorporating team-based fitness training in the middle of the week and a brief pre-match taper between matches (Haugen et al., 2022). Planning the weekly training schedule should consider the high physiological and psychological demands that the numerous competitive matches may place on individual players. Therefore, implementing a suitable periodization strategy in elite soccer poses a significant challenge for both the head coach and the fitness coaching staff (Haugen et al., 2022). As a result, the training methods used by the coach may determine the training loads elicited. This may be one reason why some head coaches may find it challenging to balance the need for players to train and recover in between competitive matches while maintaining everyone's fitness levels (Teixeira et al., 2021).

1.6. Monitoring training and match load

Soccer teams are exposed to greater training volumes and competition demands than ever before (Miguel et al., 2021), which may have implications for overall players' performance. Advances in technology available for monitoring athletes have led to greater opportunities to support players scientifically, and thus, it is possible to manage and monitor the training load so that there is a significant muscular restructuring (Moalla et al., 2016). Therefore, the process of load monitoring should aid coaches in making decisions about players' availability for training and competition, with the primary objectives being to improve performance and reduce the potential risk of injury (Claudino et al., 2019). As a result, there is now a vast array of load measures available that are obtained using telemetry and GPS, as well as other microtechnologies (Strauss et al., 2019).

The quantification of training load is generally based on both external (e.g., distance, power output, number of repetitions) and internal (e.g., oxygen uptake, heart rate, blood lactate, rate of perceived exertion) indicators of effort intensity (Bourdon et al., 2017). External loads are objective measures of the work performed by athletes during training or competition, which are determined by the

organization, quality, and quantity of exercise (Seshadri et al., 2019). The most common external load measures include power output, speed, acceleration, time-motion analysis, and deceleration (Clemente et al., 2019). In contrast, the internal load is defined as the relative biological stressors imposed on athletes during training or competition, reflecting the psychophysiological responses that the body initiates to cope with the requirements elicited by the external load (Miguel et al., 2021). An example is the s-RPE, derived from a perception of effort (Neto et al., 2020). When it comes to monitoring athletes' fatigue and/or performance responses to their training load, there is increasing interest in monitoring the status of the autonomic nervous system (ANS) via measures of HR, including the level and variability of HR at rest (Plews et al., 2013) and following exercise (Williams et al., 2018).

It is widely agreed that an integrated approach, rigorous and consistent, combining internal and external loads, provides more significant information about the stress experienced by soccer players than interpretations based on isolated data. It is also agreed that this information should be simplified, with reporting limited to a few key metrics. In particular, athletes who demonstrate a lower internal load to standardized external load completed under comparable circumstances show increased fitness (Piedra et al., 2021). On the other hand, if the athlete's internal load is raised in this scenario, they may be losing fitness or becoming fatigued (Bourdon et al., 2017). It can be determined which variables to include in an integrated approach by identifying the internal and external load measures employed by investigations that use training or a match as the monitoring environment (Watson et al., 2017).

1.7. Training and match load distribution analysis

The weekly training regimens of soccer players vary based on the phase of the yearly plan, the number of matches during the season, and/or the coach's expertise (Clemente et al., 2019). Training during the pre-season primarily focuses on reestablishing the players' fitness after the off-season. This is in contrast to in-season training, which frequently emphasizes the maintenance of specific abilities acquired during pre-season (Fessi et al., 2016). Typically, professional team players engage in 4-6 training sessions per week during the

competitive season, though this workload can rise to 1-2 sessions, 5 days a week during pre-season (Nobari et al., 2022). These alterations in training patterns substantially elevate the demands placed on players and may result in variations in the physiological stress associated with this work (Hughes et al., 2018).

Limited research has systematically endeavoured to quantify and compare the training loads accumulated by elite professional players over a short duration during the in-season and pre-season periods. This gap may be attributed to the emphasis on developing methodologies for assessing the exertion of professional players, rather than directly comparing the stress associated with different training periods (Haddad et al., 2017). Nonetheless, comprehending the extent of disparities in the physiological stimuli encountered during different training periods could facilitate the development of a model for weekly training loads that could optimize performance and fitness levels at various points in the season (Temm et al., 2022). In addition to quantifying training loads using conventional measures such as average heart rate and RPE, it is essential to consider the specific subcomponents of each training session in terms of training objectives. Subtle variations in the physiological stress associated with specific types of training or discrete sub-components of a training session may potentially explain the anecdotal observations regarding training at specific phases of the yearly plan (Armstrong et al., 2022).

In a systematic review, Teixeira et al. (2021) showed differences in the distribution of loading, especially in the middle of the week (MD-5, MD-4, and MD-3). The type of weekly micro cycle had a significant impact on the distribution of loading. For example, the compensatory session was more intense than the recovery session. The study showed the results of few studies found that total distance and s-RPE decreased in training session three days before match play (MD-3), such as high-intensity movements, such as high-speed running (HSR) and sprinting, were higher in MD-3 and MD-2 compared to MD-1 and demonstrated that MD-4 and MD-3 had the highest intensity and volume within the weekly micro cycle.

1.8. Autonomic nervous system

As its name suggests, the autonomic nervous system (ANS) is subconsciously governed and transmits signals from the central nervous system (CNS) to the periphery. According to Bankenahally and Venkata (2016), the ANS regulates a variety of physiological processes throughout the body, including hepatic gluconeogenesis and glycolysis, smooth muscle contractility, vasodilation and vasoconstriction, lung airway dilation, and endocrine release. According to Lamotte et al. (2021), the ANS can be divided into two distinct branches that differ anatomically and physiologically. The tenth cranial nerve, also known as the Vagus nerve, plays a major role in the parasympathetic nervous system (PNS) branch, which is primarily responsible for the recovery and digestion processes (Breit et al., 2018). While the sympathetic nervous system (SNS) primarily uses the neurotransmitter norepinephrine (NE) in the postganglionic fibers to affect changes at the target, it also plays a role in the classic "flight or fight" response (Daniela et al., 2022).

The action at the specific site results from the predominance of one branch and the relative proportion of receptor types. Most ANS-controlled organs are under the control of both the parasympathetic and sympathetic branches. It has been suggested that exercise-induced bradycardia in athletes is partially caused by an increased vagal tone to the sinus node (Woolf & Butcher, 2011). Vagal tone is the balance between PNS and SNS activity. Sympathetic action will be predominant during exercise, and after exercise has ended, sympathetic activation will gradually decline to make way for more parasympathetic activity (Daniela et al., 2022).

1.9. Nocturnal measurements

Monitoring pulse rate is an essential tool for gathering information about an athlete's physical state. It can be used to measure a variety of situations and analysed in different ways. However, it is important to consider the advantages and disadvantages of each recording condition and analysis method to select the most appropriate monitoring technique for athletes (Breit et al., 2018). Resting HRV is a complex measure that various physiological factors can influence. HRV monitoring is used to detect changes in cardiac autonomic nervous system (ANS)

status due to exercise (Dellal et al., 2012). It is important to standardize recording conditions and isolate training-induced effects on ANS. Night recordings provide the best-standardized recording conditions, but differences in sleep patterns and quality can affect HRV measurements (Singh et al., 2018). HRV data collected during selected slow-wave sleep episodes (SWSE) may provide more accurate measurements. However, night recordings can be difficult to implement daily, limiting their usefulness. Actigraphy is a preferred method for monitoring an athlete's sleep as it is inexpensive and can be administered with minimal impact on their routine.

Sleep monitoring is essential for an athlete's recovery continuum (Povea & Cabrera, 2018). Adequate sleep duration and quality are necessary for optimal health and performance. Athletes usually sleep less than 7 hours, which can impact their psychomotor and technical accuracy and, therefore, their competitive performance (Charest & Grandner, 2020). Subjective tools, such as diaries, are commonly used to monitor athletes' sleep, but they typically correlate poorly with objective data. The "gold standard" for sleep monitoring is polysomnography (PSG), which is useful for investigating sleep pathologies (Buchheit & Simpson, 2016). However, PSG is expensive and impractical for monitoring athletes in the field. Conversely, actigraphy is a cost-effective and minimally disruptive method for objectively monitoring an athlete's sleep during the night (Simim et al., 2020).

Overall, monitoring an athlete's physical state is a complex process that requires careful consideration of various factors. Pulse rate monitoring and HRV monitoring can provide valuable information, but it is important to standardize recording conditions and analysis methods (Ortigosa-Márquez et al., 2017). Sleep monitoring is crucial for an athlete's recovery and performance, and actigraphy is preferred due to its cost-effectiveness and minimal disruption to the athlete's routine (Clemente et al., 2021).

1.10. Autonomic cardiac activity

The cardiovascular system is characterized by a complex interaction of multiple organs and tissues, including the heart and blood vessels (Shen, 2021). The autonomic innervation of the heart includes both the extrinsic and intrinsic cardiac autonomic nervous systems (ANS) (Wake & Brack, 2016). The former

includes the ganglia in the brain or along the spinal cord and their axons on their way to the heart. The latter, in turn, consists of the autonomic ganglia and axons located on the heart itself or along the great vessels in the thorax (Shen, 2021). Numerous structural and functional evidence suggest that the intrinsic cardiac ANS forms a complex neural network composed of ganglionated plexi (GP) concentrated in the epicardial fat pads and the connecting ganglia and axons (Fedele & Brand, 2020). The complex autonomic connections between the internal cardiac ANS and the extrinsic cardiac ANS may be modulated by these GP (Avazzadeh et al., 2020).

Anatomically, the atria and ventricles contain a number of major groups of GP, which are localized at various locations in the ventricular walls of the atria. In particular, the atrioventricular node is innervated by the inferior vena cava-inferior atrial GP, whereas the sinus node is largely innervated by the right atrial GP (at the junction between the inferior vena cava and the left atrium) (Hadaya & Ardell, 2020). In response to varied stimuli and routine activities, the sympathetic and parasympathetic branches of the autonomic nervous system (ANS) work in opposite ways to fine-tune heart tissue (Avazzadeh et al., 2020). However, it has been hypothesized that various cardiovascular illnesses, including arterial hypertension, heart failure, and myocardial infarction, may be caused by an imbalance between sympathetic and parasympathetic drives (Kishi, 2012).

In regards to elite soccer, players exhibit remarkable anaerobic fitness capabilities that allow them to handle different physical demands experienced during matches (Hostrup & Bangsbo, 2023). An example is the activity of the ANS evaluated through the analysis of HRV (Arakaki et al., 2023; Bush et al., 2016; Mejía-Mejía et al., 2020). HRV has become one of the practical and popular methods to monitor positive and negative aspects for training adaptations in athletes, in particular cardiac autonomic activity (parasympathetic modulation) (Nakamura et al., 2016). This is a physiological marker of balance between the sympathetic and parasympathetic nervous systems and is sensitive to fatigue, allowing the assessment of individual training responses in soccer players (Morales et al., 2019). Monitoring training-related cardiac autonomic responses

has been facilitated by using after-waking ultra-short-term HRV measurements in athletes (Nakamura et al., 2016).

HRV analysis can be performed using linear or non-linear methods, with linear methods being divided into the time domain and the frequency domain (Shaffer et al., 2020). The indices obtained from the time domain are derived from the measurement of intervals between successive normal beats (R-R) and can be evaluated by statistical or geometric patterns, including RMSSD, SDNN, NN50, and pNN50 (Shaffer & Ginsberg, 2017). On the other hand, the linear method in the frequency domain decomposes the total variability of the signal into specific components that operate in different frequency bands, allowing identification. Furthermore, to reduce any potential non-uniformity or skewness in HRV, data can be log-transformed by taking the natural logarithm before conducting any statistical analyses (Estévez et al., 2015).

One of the methods chosen to assess HRV in studies was short-term windowed spectral analysis of R-R intervals (SWSE) (Estévez et al., 2015). This method considers the first 10 minutes of the first low and regular HR episode lasting at least 15 minutes, the lowest standard deviation of R-R intervals (SDNN) throughout the period of interest and a low inter-beat autocorrelation between successive R-R intervals (Claiborne et al., 2021). Using SWSE, a single session of supramaximal intermittent exercise was found to cause a reduction in vagally-mediated HRV indices among young non-athletes during night sleep following exercise (Mario Estévez et al., 2015). However, the SWSE requires the analysis of "only" 10 minutes out of many hours of sleep. In contrast, an "hour-by-hour" approach using all the R-R intervals recorded throughout the sleep period (J. Costa et al., 2018). In addition to being beneficial in assessing individual training-induced stress and allostasis disruption, HRV is sensitive to fatigue brought on by higher training loads (Corrigan et al., 2021).

Night recordings may provide better differentiation of the changes in autonomic nervous system homeostasis since parasympathetic activity is elevated during sleep at night (Zoccoli & Amici, 2020). In general, sympathetic influence was observed 1h after exercise cessation, and pre-exercise levels appeared to be restored as early as 24h after exercise (Dellal et al., 2012;

Matsumura et al., 2021). On the other hand, the exercises performed in the study above differ from those routinely performed by athletes in their training programs because of their length and environmental conditions (Cayres et al., 2015). Periods of intensified training loads have been shown to increase the level of disturbance in sleep and HRV, and it is well-known that changes in nocturnal HRV after exercise may last up to 24h (Nummela et al., 2016).

Another important variable that can determine differential autonomic responses during recovery from an intense training bout are circadian rhythms (Thomas et al., 2020). These rhythms influence our daily behaviour. People typically exhibit activity preferences at different times of the day based on their innate circadian rhythms. The literature has shown differences between clonotypes for several physiological variables, involving sleep patterns, hormone secretion, physical performance, HRV (Montaruli et al., 2021). Bonato et al. (2017) demonstrated that the circadian typology of an individual determines varied autonomic responses during the recuperation period after intense morning-only training. Nevertheless, alterations HRV that may transpire during post-exercise sleep could be influenced by the athletes' exposure to either chronic or acute late-stage exercise.

1.11. Sleep patterns

Sleep has essential biological functions concerning physiological processes, cognition, memory and learning (Simon et al., 2022). A minimum of 7 to 9 hours of total sleep time per night is advised to support health and cognitive function (Fry & Rehman, 2022). Although sleep's function is not fully understood, it is generally accepted that it permits recovery. It is considered a fundamental requirement for maintaining human health and essential to athletes' muscle and mental restoration (Halsen, 2014). Several crucial psychological and physiological processes during sleep may be critical to healing, creating a neurologically dynamic behaviour with a rapid reversibility and elevated arousal threshold (Joiner, 2018).

According to Fry and Rehman (2022), 85% sleep efficiency is usually recommended as a relevant indicator of healthy sleep quality, whereas 74% indicates poor sleep quality for young and older people. A variety of factors may

influence sleep quality and duration. In this regard, bedtime, sleep latency (i.e., the period from bedtime to falling asleep), number and time of awakenings, late morning awakenings and waking times, number and duration of naps, and sleep quality as determined by the feeling of being well rested are all considered (Schwab, 2020). Despite the complexity surrounding sleep's necessity, justification, and effects, the fact that it has persisted throughout human evolution suggests that it must be essential for us (Simon et al., 2022). During sleep, five distinct stages occur, and metabolic activity is at its lowest point (i.e., slow breathing, low heart rate, and low cerebral blood flow). Thus, sleep does not represent a single "state of being" (Assefa et al., 2015). Rapid Eye Movement (REM) and non-REM sleep (NREM) are two fundamentally different "states of being" that constitute sleep cycles.

The latter is further subdivided into "deep" (slow wave) and "light" stages (Stages 1 and 2) (Tarun et al., 2021). The biological, neurological, and physical manifestations of these various stages of sleep differ based on specific differences in brain activity. Stage 3 and 2 slow cortical wave activity oscillations occur during NREM sleep (Stevner et al., 2019). In contrast, during REM sleep, the electroencephalogram (EEG) patterns of the brain resemble those in waking with associated eye movement in the phasic REM stage and without eye movement in restorative REM sleep (Deliens et al., 2013).

The circadian process is essential in sleep regulation and the "two-process model" for alertness determination (Borbély et al., 2016). Circadian regulation is demonstrated by maintaining a 24-hour rhythm cycle in sleep propensity independent of prior sleep. The brain's natural cycle is slightly longer than 24 hours, but blue light resynchronises it daily. Thus, a healthy circadian rhythm ensures that people are awake during the day and sleepy at night. One key mechanism guaranteeing the operation of the "biologic clock" is the brain's pineal gland secreting the hormone melatonin at night (Potter et al., 2016).

Moreover, although the suprachiasmatic nucleus of the hypothalamus controls circadian rhythms, the suprachiasmatic nucleus is unable to compensate for the fact that humans are susceptible to variations in their natural environment (Fullagar et al., 2015), most notably through the light-dark cycle (Skeldon et al.,

2017). Endogenous circadian rhythms and regular sleep-wake cycles in athletes can become out of sync when they experience disruptions to their environments (Nobari et al., 2023). These changes in sleep patterns may lead to increased homeostatic pressure and changes in core temperature, melatonin levels in the blood, and emotional control.

In turn, the increase in sleep propensity during waking hours and its decrease during sleep are indicators of homeostatic regulation. The balance between alertness and sleepiness is determined by the interaction of circadian sleep-alertness cycles and homeostasis. In this sense, the achievement of appropriate sleep rates must be analysed (Hauswirth et al., 2014). Available evidence suggests that sleep optimization can influence the performance of various physical activities: improvements in sleep duration and quality seem to improve reaction time, psychomotor achievement, technical accuracy and endurance performance, with probable positive effects on competitive performance (Doherty et al., 2021).

Despite the importance of sleep in restoring physiological and cognitive functions, athletes often sacrifice hours due to overloaded schedules (Costa et al., 2021). The effects of sleep loss on physiological responses to exercise can harm muscle recovery and lead to a reduction in immune defence, resulting in impairments in most aspects of cognitive function and mood stability, outcomes that can potentially tarnish the neurocognitive components of many sports (Fullagar et al., 2015; Khan et al., 2018). Rupp et al. (2008) recruited 24 healthy young adults who completed seven nights of sleep restriction (3h time in bed) followed by five recovery nights (8h time in bed). In both the usual and extended sleep groups, within subjects' performance decreased rapidly across days of 3h time in bed. However, between groups' performance was significantly less impaired in the extended sleep group than in the usual sleep group. Impressively, the differences among groups persisted even following five recovery nights of 8h time in bed.

Soccer players maintain to strict match and training schedules during currently competitive periods, which can shorten sleep time (Sargent, Lastella, et al., 2014) and prevent recovery processes (Fullagar, 2016), particularly when

practice sessions and/or matches are occurring close to bedtime (Vitale et al., 2019). If the recovery rate could be enhanced, higher training loads would be appropriate, and a higher performance plateau could develop (Hughes et al., 2018). Finding non-invasive recuperation techniques is crucial for human health and performance predictors. The state of an athlete's fatigue has been studied using monitoring technologies utilized by high-performance programs, such as heart rate recovery and sleep (Nässi et al., 2017).

Sleep practices and exercise and sports training have a complex and reciprocal interaction (Sawczuk et al., 2021). As a result, it is necessary to investigate the relationships between training and match loads and both sleep quantity and quality (Oliveira et al., 2023).

Chapter II

Aims and structure

2. Aims

This dissertation aims can contribute to a deeper understanding of various aspects of soccer performance and inform the development of evidence-based strategies for optimizing training and performance in soccer. Concretely, we pretend to examine the factors contributing to fatigue in soccer players, including match frequency and training intensity (though internal and external training and match loads), as well as develop strategies for managing and mitigating fatigue to optimize player performance. For such, it was necessary to evaluate the impact of factors like competition schedules, player fitness levels on training periodization, time of playing per match and develop methods for monitoring and quantifying the training and match load experienced by soccer players.

As recovery process, we choose to investigate the architectures and the roles of autonomic nervous system and sleep in regulating physiological responses during soccer activities. For that, we examined the use of nocturnal measurements (e.g., sleep quality, sleep duration and $\ln\text{RMSSD}$) as indicators of player readiness and recovery in soccer and the impact of stress and fatigue on autonomic cardiac function and sleep. Furthermore, we explored the relationship between autonomic cardiac function and sleep patterns in soccer players and explored how interventions targeting autonomic function and sleep can improve recovery.

Subsequently, this research investigates an examination of the methodological protocols meticulously devised for the acquisition of data pertaining to sleep patterns, heart rate variability (HRV), and training loads (TLs). The overarching objective of this investigation is to comprehensively delineate the patterns of inter-individual variability, focusing on differences among individual subjects, in the domain of sleep patterns, nocturnal HRV and training loads. Additionally, it explored the intra-individual associations of training/match load with sleep and HRV measures in elite soccer players during a nine-week competitive period.

3. Dissertation structure

The present dissertation is characterized by basic research, of a descriptive nature, with an analytical quantitative approach (Eyisi, 2016). Thus, this thesis is structured according to the following order:

1. The first chapter consisted of the general introduction, with a succinct general review of the literature, referring to its relevance, and where we defined the problem and objectives of the thesis.
2. In the second chapter, the one study carried out in the form of scientific article was included.
3. In the third, the general discussion appeared, presenting the innovation of the study, the main conclusions regarding the correlations between training loads and sleep variables and HRV, as well as intra and inter-subjects.
4. The fourth chapter presented the conclusions, the practical applicability of the work and its limitations.
5. In the fifth chapter, bibliographical references were placed.
6. Finally, in the sixth and final chapter, appendixes.

Chapter III

The effect of accumulated training and match load in sleep indices
and nocturnal heart rate variability of elite soccer players

Title: The effect of accumulated training and match load in sleep indices and nocturnal heart rate variability of elite soccer players

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Abstract: The aim of this study was to investigate the sleep patterns and nocturnal cardiac autonomic activity (CAA) of elite soccer players during a nine-week in-season period. Twelve male participants (age: 24.2 ± 4.9 years) were included in the study. Participants' sleep indices were measured using wrist actigraphy, while heart rate monitors were used to measure CAA. The training and match loads were characterised using the session rating of perceived exertion (s-RPE), player load (PL), and total distance. Players who played more than 60 minutes had more oscillations in heart rate variability (HRV) and sleep indices than those who played less than 60 minutes. Furthermore, the total sleep time was

below the recommended levels for two consecutive weeks for this group. The study found a *moderate* correlation between s-RPE and sleep duration, indicating that players who perceived higher training/match load slept less. There was also a *small* correlation between PL and natural logarithm of low frequency (lnLF), indicating that higher player loads were associated with lower HRV. Finally, a *moderate* correlation was found between sleep efficiency and natural logarithm of the square root of the mean of the sum of the squares of differences between successive normal-to-normal intervals (lnRMSSD), suggesting that better sleep efficiency was associated with higher HRV. Findings suggests that players had a suboptimal sleep pattern during the in-season nine-week period and training/match load impacted HRV and sleep indices. This may help coaches in developing individualized training programs to meet the demands of each player in training and match.

Keywords: accelerometer; nocturnal cardiac autonomic activity; recovery; monitoring; performance.

Introduction

Monitoring and managing training loads (TL) has been a topic of substantial interest in sports, providing coaches and athletes with information about training requirements (Jaspers et al., 2017). This becomes even more important during congested periods when providing fatigue data and adjusting training according to players' adaptation would enhance the optimization of training programs and reduce the potential risk of injury (Gurau et al., 2023).

Furthermore, intensive training sessions and matches may cause extreme fatigue as a result of the central nervous system's continued, extraordinary effort, making the muscle incapable of responding as effectively as it does when rested (Sargent et al., 2014). Recovery techniques, therefore, become an important mechanism for managing fatigue (Cullen et al., 2021; Rattray et al., 2015). Athletes must employ efficient techniques to enhance the recovery process, which is a crucial component of the adaptive training process and prevent the risk of injuries (Bowen et al., 2017). Researchers have attempted to determine the effect of training load on the risk of sports injuries and other sports-related health problems. The training load is an amount of physical activity the athlete has been exposed to and, in so doing, consists of exposure itself as well as physiology and psychology pressures that are being put on him or her by those exposures (Meeuwisse et al., 2007; Renshaw & Goodwin, 2016). The relationship between risk factors and sports injuries are often complex, as the impact of these risk may depend of the current condition of the athlete (Griffin et al., 2020). If the recovery rate could be enhanced, higher training loads would be appropriate, and a higher performance plateau could develop (Hughes et al., 2018).

Therefore, it is important to find non-invasive recovery strategies for human health and predictors of performance. Research has investigated monitoring tools used by high-

performance programs to reveal the state of fatigue of an athlete, including heart rate recovery and sleep (Halson, 2014). Recently, the use of heart rate variability (HRV) indices during sleep has been implemented to assess exercise-induced disturbances as well as recovery from workloads and other sources of stress (Mishica et al., 2021). Overnight sleep measurements on consecutive days are considered the most appropriate recording condition to collect HRV after high-intensity exercise (Costa et al., 2019).

The mechanisms of regulation of HRV find their origin in the autonomic nervous system (ANS), parasympathetic and sympathetic, and HRV has been suggested as a quantitative marker of ANS (Pham et al., 2021; Yugar et al., 2023). Additionally, HRV has been useful in evaluating individual training-induced stress since it is sensitive to fatigue due to increased TL (Flatt & Esco, 2015). Moreover, considering that parasympathetic activity is high during night sleep, night recordings may allow better discrimination of the changes in autonomic nervous systems equilibrium (Zambotti et al., 2018). Heart rate (HR) represents a simple, accessible, and non-invasive tool to investigate variability in the short, medium or long term, which allows adjusting physiological needs to environmental constraints (Tiwari et al., 2021).

In turn, sleep induces good performance in athletes due to its vital function, it allows the heart to rest, as well as to repair cells and tissues, and it is widely regarded as critical to cognitive and physiological function (Silva et al., 2022). A minimum of 7 to 9 hours of total sleep time per night is advised to support health and cognitive function (Fry & Rehman, 2022), although the sleep duration of athletes is frequently less than 7 h (Sargent et al., 2014). Since sleep is essential to an athlete's performance, its potential should be maximized (Roberts et al., 2019).

Nowadays, overnight sleep measurements over several days and weeks are applicable in the recovery of HRV following exercise (Costa et al., 2021), which depends on multiple factors, namely the time of day, training and match loads, and chronotype (Nedelec et al., 2018). Despite this approach helping to provide fundamental insight into sleep and HRV indices in athletes, it lacks details of how these indicators may vary across weeks. For instance, between-night individual variability in sleeping time might indicate the need for individualized sleep education strategies and interventions to promote appropriate sleep (Nedelec et al., 2018). Currently, there is a need for individualized strategies and interventions in HRV and sleep over time, as individual variability can describe differences within individuals (Costa et al., 2021).

During congested competitive periods, soccer players follow strict match and training schedules, increasing sleep disturbance and HRV, which impairs recovery processes (Fullagar, 2016). Therefore, it is fundamental to use valid load metrics to properly contextualize the impact of training and/or match workload on sleep habits and nocturnal HRV responses (Costa et al., 2018), and in the s-RPE (Halsen, 2014). The effects of HRV and sleep loss on athletes performance have been studied in the last years. Costa et al. (2021) found negative correlations between s-RPE and sleep duration and efficiency, respectively ($p < 0.001$; and $p = 0.02$). Costa et al. (2019) observed small individual fluctuations in nocturnal cardiac autonomic activity during a female soccer international tournament. However, due the complexity of privacy of HRV and sleep function, the variability in the individual requirement for sleep and the limited availability of athletes to participate in sleep and HRV studies (Peacock et al., 2019) reveals the need to carry out further studies.

As a result, the aims of this study are twofold: (1) to describe the inter-individual (i.e., variability between subjects) patterns of sleep, nocturnal HRV and training load, and (2) to explore the intra-individual associations of training/match load with sleep and HRV measures in elite soccer players during a 9-week competitive period. We hypothesized that athletes would present a high inter and intra-variability in sleep and HRV measures. Additionally, we expected a negative within-subject association of sleep and HRV with training/match load.

Methods

1. Study design and setting

A within-subject design was used to longitudinally evaluate the association between the weekly average internal and external load, the regulation of HRV and sleep indices in an elite male soccer team. This study followed the STROBE guidelines for reporting descriptive and observational designs (von Elm et al., 2008), comprising a data collection process carried out over 9 weeks (Figure 1). External TL and match load (ML) were collected over the sessions, while internal TL and ML were collected 15 to 30 minutes after sessions (Askow et al., 2021). Nocturnal HRV and sleep indices data were collected at players' home overnight. Timeline was design to collect data in the match-day (MD), 2 days before MD (MD-2) and 3 days after the MD (MD+3).

Prior to data collection, athletes were familiarized with the devices used during the investigation. The study protocol was approved by the Ethics Committee of the Faculty of Sport of the University of Porto, with the following approval code assigned CEFADÉ 14 2020.

Figure 1 - Data collection during a week of elite male soccer players (n = 12)

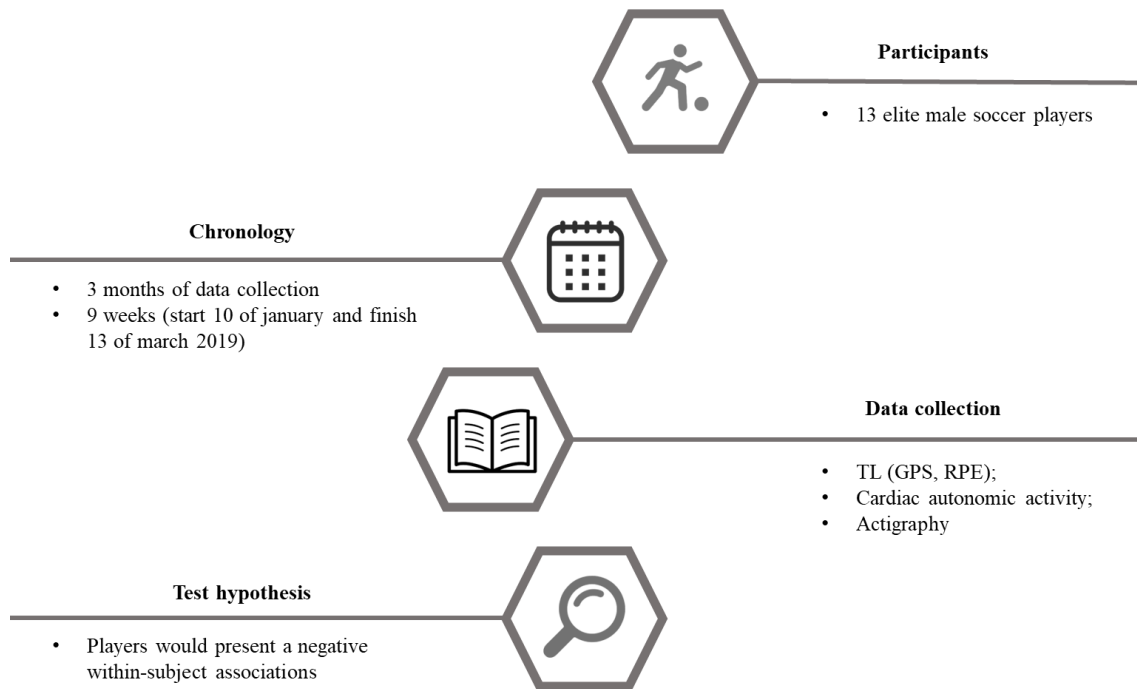
MD -2	MD -1	MD	MD +1	MD +2	MD +3
TL (GPS, RPE); Cardiac autonomic activity; Actigraphy	TL (GPS, RPE); Cardiac autonomic activity; Actigraphy	TL (GPS, RPE); Cardiac autonomic activity; Actigraphy	TL (GPS, RPE); Cardiac autonomic activity; Actigraphy	TL (GPS, RPE); Cardiac autonomic activity; Actigraphy	TL (GPS, RPE); Cardiac autonomic activity; Actigraphy
  	  	  	  	  	  

Training load (TL), heart rate variability (HRV) and session-rating of perceived exertion (s-RPE) included training-days and match-days. Training load (TL) was assessed using Global Positioning System (GPS) and session-rating of perceived exertion (s-RPE). Sleep and nocturnal heart rate variability indices were assessed

2. Participants

A convenience sample of 12 elite male soccer players (age: 24.2 ± 4.9 years; height: 179.0 ± 5.5 cm; body mass: 74.4 ± 5.6 kg), competing in the Portuguese top league was recruited to participate in the study during 9 weeks. The data collection process involved GPS and accelerometers data, and the use of effort perception questionnaires as illustrated in Figure 2. The following exclusion criteria were defined: a) age < 18; b) tobacco use; c) and use of medications and medical conditions contraindicating physical exercise. According to the 64th WMA Assembly, Fortaleza, Brazil (2013), all general research ethics principles were respected, following the standards suggested by the Helsinki Declaration. Therefore, written informed consents were obtained and the participants informed about the study procedures and the possibility to withdraw from the study at any moment, without sanction.

Figure 2 - Flowchart of methodology of the study. Participants, chronology, data collection and test hypothesis are illustrated.



3. Variables

Nocturnal heart rate variability monitoring

The Bodyguard 2 heart rate monitor set (Firstbeat, Finland) was used to record RR intervals. This tracking system demonstrated validity and acceptable levels of accuracy (TEM <0,05%) in the study of Parak and Korhonen (2017). The raw data were converted to ASCII format in the form of RR intervals in milliseconds and exported to Kubios HRV TM version 2.0 (Department of Physics, University of Kuopio, Kuopio, Finland. Although a previous study (Perrotta et al., 2017) showed an unsatisfactory level of agreement between Kubios HRV 2.2 and Elite HRV, the relationship seen as expressed through the Pearson's product-moment correlation was nearly perfect ($r > 0,90$, CI = 95%). Non-parametric method, namely the Fast Fourier Transform, was used to obtain measurements of HRV in the frequency domain. Information about the total power and

the frequency was obtained by default: very low frequency (VLF: 0-0.04 Hz), low frequency (LF: 0.04 -0.15 Hz) and high frequency (HF: 0.15-0.4 Hz). The normalized LF and HF units were calculated. The LF/HF ratio was calculated as it can express the balance between the sympathetic and parasympathetic nervous systems (Kim et al., 2023).

Heart rate data were recorded at 5 second intervals by short-range radio telemetry (Firstbeat Sports, Jyväskylä, Finland) during training and matches. A previous study (Parak & Korhonen, 2014) showed the validity and provided acceptable heart rate accuracy ($r = 0,86-0,87\%$) and reliability scores for $<5\%$. Subjects wore an adjustable elastic chest strap where the devices were attached. Data were transferred to a computer using Firstbeat Sports Server corporate software version 4.7.3.1. From this software, the training impulse (TRIMP) was calculated as proposed by Bannister (1991), later applied by Stagno et al. (2007). The HR peak presented in the match was used to calculate different HR zones: 60-69%, 70-79%, 80-89% and 90-100%.

Sleep monitoring

For the quantitative measurement of sleep were used accelerometers (Actigraph GT3-x). The validity and acceptable levels of accuracy ($ICC = 0,99$) of this tracking system have previously been presented (Anderson et al., 2019). Participants were instructed to strap on the accelerometer every night as they prepared to go to bed. When participants turned on the lights in the morning, or when they got up, the accelerometer was removed. Measures of sleep latency and efficiency, total sleep time and wake time after sleep onset were taken. In the morning, before training and matches, the Hooper index was used to quantify subjective ratings of sleep quality, fatigue, stress and muscle pain (Rabbani et al., 2019).

Internal training and match load

To quantify internal training and match loads, it was used session rating of perceived exertion-derived training load (sRPE-TL) calculation suggested and validated by Foster et al. (2001). This method quantifies internal TL by multiplying the training session RPE with the duration of the training session, in minutes (Askow et al., 2021). The perceived exertion collection was performed approximately 15 to 30 minutes after the end of each training session and matches to ensure that the perceived exertion refers to the entire session and not to the intensity of the most recent exercise (Haddad et al., 2017). All players were familiarized with this scale for at least before the start of the study and standardized instructions for collecting perceived exertion were followed.

External training and match load

GPS technology was used to quantify external training load (ETL). OtimEye X4 from Catapult Sports TM (Australia) with a sampling frequency of 10Hz for the time motion analysis and a sampling frequency of 100Hz for the inertial sensors was used. The validity and acceptable levels of accuracy (bias <10%) of this tracking system have previously been presented (Johnston et al., 2013). The variables selected to quantify the ETL were player load (PL), total distance (TD), sprint distance (SpD, above 25.5 km/h; m), number of accelerations (ACC, $>2\text{m/s}^2$) and number of decelerations (DCC, $<-2\text{m/s}^2$). Players wore specific vests where the devices were placed on the upper back, between the shoulder blades (Buchheit & Simpson, 2016).

4. Statistical analysis

Based on the type of the data, the sample distribution was tested using the Shapiro-Wilk test for sleep and HRV indices, training, and match loads for each day over these three months. An exploratory analysis of the data was carried out to verify the normality of the distribution corresponding to each of the study variables. Descriptive statistics included the mean and standard deviation (SD). The coefficient of variation ($CV = [\text{standard deviation}/\text{mean}] \times 100$) was calculated for the entire group and individually for sleep and HRV indices over the nine weeks to analyse the variability.

A linear mixed model analysis was performed in TL, HRV and sleep indices across the nine weeks. The days with training sessions and matches were included as a fixed effect and player identity (subject ID) as the random effect. The variance–covariance structures were selected according to the smallest Akaike information criterion. Pairwise comparisons (Bonferroni) were used to show the week-to-week mean differences for training and match loads, sleep and HRV indices. It should be noted that, during data analysis, the reference value of significance was ≤ 0.05 . The CV was calculated for the whole group and individually for sleep duration, sleep efficiency, HRV indices ($\ln RMSSD$, $\ln LF$ and $\ln HF$) and load parameters (TD, PL and s-RPE).

Within-subjects correlations were calculated between sleep duration, sleep efficiency), HRV indices ($\ln RMSSD$, $\ln LF$ and $\ln HF$) and load parameters (TD, PL and s-RPE). Correlation magnitudes were interpreted qualitatively using the following criteria: very small ($r = 0.1$), small ($r = 0.1–0.3$), moderate ($r = 0.3–0.5$), large ($r = 0.5–0.7$), very large ($r = 0.7–0.9$), and nearly perfect ($r = 0.9$) (Hopkins, W.G., et al, 2009).

Results

Actigraphy sleep and nocturnal HRV indices, training and matches load parameters and perceived ratings of wellbeing per week are summarized in Tables 1, 2 and 3. Descriptive group data ($n = 12$) responses for HRV (ie, $\ln\text{meanRR}$, mean HR, $\ln\text{RMSSD}$, $\ln\text{LF}$, $\ln\text{HF}$ and $\ln\text{LF/HF}$), sleep patterns (sleep latency, efficiency, duration and WASO) and training and match load parameters (i.e., s-RPE, total distance, player load, sprint distance, accelerations and decelerations) show the significant differences between weeks for the same variable.

As a group, sleep duration ranged between 423 (399; 447) to 479 minutes (467; 492) during all 9 weeks of competition. In this way, weeks 1 and 9 had the lower value compared week 4, respectively ($p = 0.032$ and $p = 0.018$). Furthermore, the players who played more than 60 minutes presented 2 weeks above than recommended (week 8, 409 minutes; week 9, 403 minutes) compared the rest of weeks of competitions. Sleep efficiency ranged between 85% (84; 86) and 91% (90; 91). Weeks 7 and 8 was lower compared the other weeks, especially with week 2, respectively ($p < 0.001$ and $p = 0.042$). Also, week 7 was lower compared week 4 ($p = 0.002$). In addition, a lower SE was found in week 4, especially when compared the same week for players who played less than 60 minutes (-7%). All the sleep-related variables are presented in Table 2.

Individually, some players slept less than recommended (<7 hours; 420 min) over the 9 weeks at least one week: week 2 ($n = 1$; player 12), week 4 ($n = 1$; player 2), week 6 ($n = 2$; player 3 and 12), week 8 ($n = 1$; player 7) and week 9 ($n = 3$; player 4, 7 and 10). Sleep duration CV ranged between 13 and 23%. Just one player presented bad sleep quality in weeks 8 and 9, respectively (73% and 70%). The rest of players had a good

sleep quality (i.e., sleep efficiency > 74%; individual range between 78–94%) across all weeks of the competitions (Figure 5).

Table 1 - Players nocturnal heart rate variability indices during the 9 weeks in competitive phase (n = 12).

	Ln Mean R- R (ms)	Mean HR	ln RMSSD (ms)	Ln LF (ms2)	Ln HF (ms2)	Ln (LF/HF)
Week 1	7.21 (7.15; 7.28)	44.80 (41.97; 47.63)	4.66 (4.54; 4.78)	8.12 $\Pi, \Psi, \Gamma, \Sigma, \Theta$ (7.87; 8.37)	7.96 (7.66; 8.25)	1.02 (1; 1.05)
Week 2	7.26 (7.23; 7.28)	43.08 (41.98; 44.18)	4.58 (4.51; 4.64)	7.99 α (7.91; 8.07)	7.71 (7.58; 7.85)	1.02 (1; 1.05)
Week 3						
Week 4	7.20 (7.17; 7.24)	45.18 (43.68; 46.69)	4.58 (4.49; 4.68)	8.05 α (7.96; 8.15)	7.79 (7.58; 7.99)	1.05 (1.03; 1.07)
Week 5	7.14 (7.09; 7.18)	48.26 (45.98; 50.55)	4.59 (4.38; 4.8)	8.18 α (7.91; 8.44)	7.98 (7.59; 8.37)	1.03 (1.01; 1.06)
Week 6	7.25 (7.22; 7.29)	43.36 (41.82; 44.9)	4.55 (4.48; 4.61)	8.16 (8.11; 8.22)	7.61 (7.49; 7.73)	1.04 (1; 1.07)
Week 7	7.29 (7.23; 7.35)	41.87 (39.29; 44.45)	4.66 (4.51; 4.82)	8.15 (7.99; 8.3)	7.88 (7.61; 8.16)	1.04 (1.02; 1.06)
Week 8	7.19 (7.16; 7.23)	46.20 (44.51; 47.9)	4.62 (4.53; 4.72)	8.31 α (8.19; 8.43)	7.88 (7.69; 8.06)	1.07 (1.05; 1.08)
Week 9	7.15 (7.11; 7.19)	48.32 (46.38; 50.26)	4.30 (4.21; 4.4)	7.75 α (7.66; 7.85)	7.24 (7.07; 7.41)	1.08 (1.06; 1.09)

Values are group mean and 95% confidence interval estimates. Ω Significantly different from week 1. Π Significantly different from week 2. β Significantly different from week 3. Ψ Significantly different from week 4. Γ Significantly different from week 5. μ Significantly different from week 6. Σ Significantly different from week 7. Σ Significantly different from week 8. Θ Significantly different from week 9.

Regarding HRV indexes, ln LF ranged between 7.75 ms (7.66; 7.85) and 8.31 ms (8.19; 8.43). Week 1 was the week with most impact in other weeks, especially weeks 2, 4, 5, 8 and 9, respectively ($p = 0.004$, $p = 0.032$, $p = 0.002$, $p = 0.046$ and $p = 0.002$). Overnight CV of ln RMSSD, ln LF and ln HF ranged between 7% - 14%, 6% - 10% and 9% - 15%, respectively. All the HRV-related variables are presented in Table 1.

Individually, player 4 appeared to present higher lnRMSSD CV (11%) over 9 weeks of the competition period, particularly in week 9 (CV = 19%, average = 3.80 ms). This occurred simultaneously with a reduced lnRMSSD average for the (3.98 ms), throughout the competition period, in contrast to the remaining team (individual lnRMSSD ranging

between 3.81 and 5.16 ms, and lnRMSSD CV ranging between 1 and 7%) (Figure 1). Individual lnLF and lnHF CV ranged between 1–11%, 1–13%, and 2–15%, respectively (Figure 6).

Table 2 - Players actigraphy sleep indices during the 9 weeks in competitive phase (n = 12)

	Sleep latency (min)	Sleep efficiency (%)	Sleep duration (min)	Sleep WASO (min)
Week 1	5 (3; 8)	86 (84; 88)	423 [∇] (399; 447)	65 (52; 77)
Week 2	6.88 (6; 8)	90 ^{£, ж} (89; 91)	456 (446; 466)	44 ^{£, ж} (40; 48)
Week 3				
Week 4	5 ^º (4; 6)	90 [£] (89; 91)	479 ^{Ω, º} (467; 492)	50 [£] (44; 56)
Week 5	11 (8; 13)	87 (83; 90)	464 (424; 504)	65 (48; 82)
Week 6	6 (5; 6)	91 (90; 91)	465 (453; 477)	40 (38; 43)
Week 7	8 (7; 9)	85 ^{Π, ∇} (84; 86)	451 (435; 467)	69 ^{Π, ∇} (64; 75)
Week 8	5 (5; 6)	87 ^Π (86; 88)	448 (434; 461)	61 ^Π (55; 67)
Week 9	11 [∇] (8; 14)	89 (88; 90)	425 [∇] (411; 439)	43 (39; 47)

Values are group mean and 95% confidence interval estimates. ^Ω Significantly different from week 1. ^Π Significantly different from week 2. ^β Significantly different from week 3. [∇] Significantly different from week 4. [‡] Significantly different from week 5. ^μ Significantly different from week 6. [£] Significantly different from week 7. ^ж Significantly different from week 8. ^Θ Significantly different from week 9.

With reference to training loads, player load ranged between 560 (528; 592) and 747 (701; 793). Week 7 was the week lower load in comparison with other weeks, notably weeks 2, 5 and 6, respectively ($p = 0.002$, $p < 0.001$ and $p = 0.001$). In turn, total distance ranged between 5635 m (5196; 6075) and 7532 (6885; 8179). Week 7 was the week lower load in comparison with other weeks, notably weeks 1, 5 and 6, respectively ($p = 0.016$, $p = 0.002$ and $p = 0.003$). Lastly, s-RPE ranged between 155 A.U. (131; 179) and 425 A.U. (384; 466). Weeks 1 and 2 were the weeks with lower load in comparison with other weeks, notably weeks 3, 5, 6, 7, 8 and 9, respectively ($p = 0.003$ and the remaining $p <$

0.001). The average of s-RPE, total distance and player load of players who played more than 60 minutes per match ranged 207 A.U. and 513 A.U., 5996 m and 8887 m and 612 and 836. At the other side, the average of s-RPE, total distance and player load of players who played less than 60 minutes per match ranged 138 A.U. and 395 A.U., 5218 m and 6975 m and 532 and 705. In this way, all of results from each week were bigger players who played more than 60 minutes per match in comparison to players who played less than 60 minutes per match. All the training loads-related variables are presented in Table 3.

Table 3 - Players training and match loads during the 9 weeks in competitive phase (n = 12).

	s-RPE (A.U.)	TD (m)	Player Load
Week 1	189 ^{β, ̂, μ, £, ж, Θ} (166; 212)	7141 [£] (6737; 7546)	691 (656; 727)
Week 2	155 ^{β, ̂, μ, £, ж, Θ} (131; 179)	6769 (6084; 7455)	736 (668; 804)
Week 3	322 ^Π (285; 359)	5877 (5429; 6325)	617 (573; 661)
Week 4	249 [̂] (217; 281)	6460 (5876; 7044)	654 (598; 710)
Week 5	425 ^{Ω Π ̂} (384; 466)	7171 [£] (6773; 7568)	747 ^{£, ж} (701; 793)
Week 6	351 ^{Ω Π} (319; 383)	7152 [£] (6650; 7654)	712 [£] (665; 758)
Week 7	344 ^{Ω Π} (318; 370)	5635 ^{Ω, ̂, μ} (5196; 6075)	560 ^{Π, ̂, μ} (528; 592)
Week 8	338 ^{Ω Π} (300; 376)	6640 (6247; 7032)	631 [̂] (587; 675)
Week 9	341 ^{Ω Π} (303; 379)	7532 (6885; 8179)	728 (665; 792)

Values are group mean and 95% confidence interval estimates. Ω Significantly different from week 1. Π Significantly different from week 2. β Significantly different from week 3. ̂ Significantly different from week 4. ̂ Significantly different from week 5. μ Significantly different from week 6. £ Significantly different from week 7. ж Significantly different from week 8. Θ Significantly different from week 9.

The within-subject correlations of sleep and nocturnal HRV indices with training and match load parameters during the nine-week analysis are presented in Tables 4 and 5. A *moderate* positive correlation [$r = 0.4$ (0.2; 0.59); ($p = 0.003$)] was found between s-RPE and sleep duration, as well as found a *small* negative correlation [$r = -0.23$ (-0.43; -0.01); ($p = 0.04$)] between PL and lnLF. A *moderate* positive correlation [$r = 0.27$ (0.11; 0.41); ($p = 0.001$)] was found between sleep efficiency and lnRMSSD.

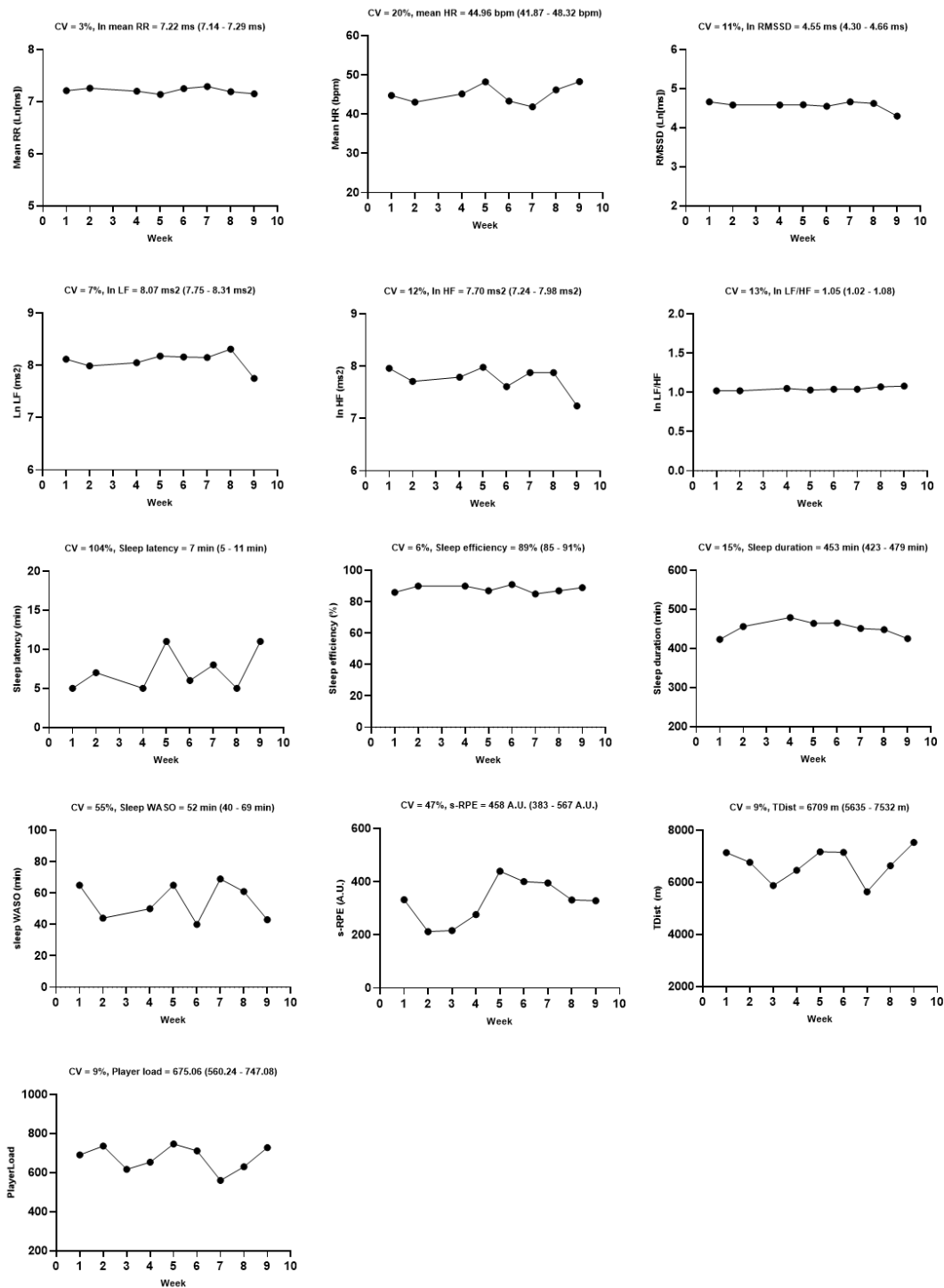


Figure 3 - Descriptive group data (n = 12) responsiveness for sleep and nocturnal HRV indices, training and match load parameters, and perceived ratings of well-being during 9 weeks in competitive phase in high-level male soccer players. Horizontal black lines show group mean $\pm$ 95% confidence interval for each week. Coefficient of variation (CV), averages, maximum, and minimum values are also presented.

Discussion

This observational study describes habitual sleep and nocturnal cardiac autonomic activity, and their relationship with training and/or match load in soccer players during 9-week competitive period. We found that most players slept less than the recommended 420 min *per* night (7h) (Fry & Rehman, 2022), especially during weeks 8 and 9 (Figure 4). Another key finding was the association between training and/or match load (s-RPE) and sleep duration. Our findings support the need to develop and implement individual sleep strategies, due the known negative consequences of reduced sleep time or sleep deprivation on athletes' health and performance (Dolezal et al., 2017). Finally, the training and match loads affect players' nocturnal cardiac autonomic activity, demonstrated negative *moderate* correlation.

Therefore, based on our data showing an increase in variability of sleep and HRV measures by soccer players for a 9-week competitive period, we confirm one hypothesis, which we expected a negative within-subject association of sleep and HRV with training/match load. The other hypothesis was validated, since sleep duration has a positive relationship with s-RPE and PL as negative associated with lnLF.

According to Hausswirth et al. (2014), training-related stress may potentially be a significant influence affecting athletes' sleep patterns. For instance, it has been reported that exposure to intense training loads in athletic populations may increase the chance of sleep disturbance and shorten sleep lengths, as well as numerous psychological factors which potentially influence an athlete's ability to perform at their peak level such as mood, anxiety, confidence, fatigue, levels of energy and motivation (Cook & Charest, 2023; Khan et al., 2018). Some athletes report having trouble falling asleep, being restless while they sleep, and having heavy legs while they sleep during periods of high training loads (Lastella et al., 2015).

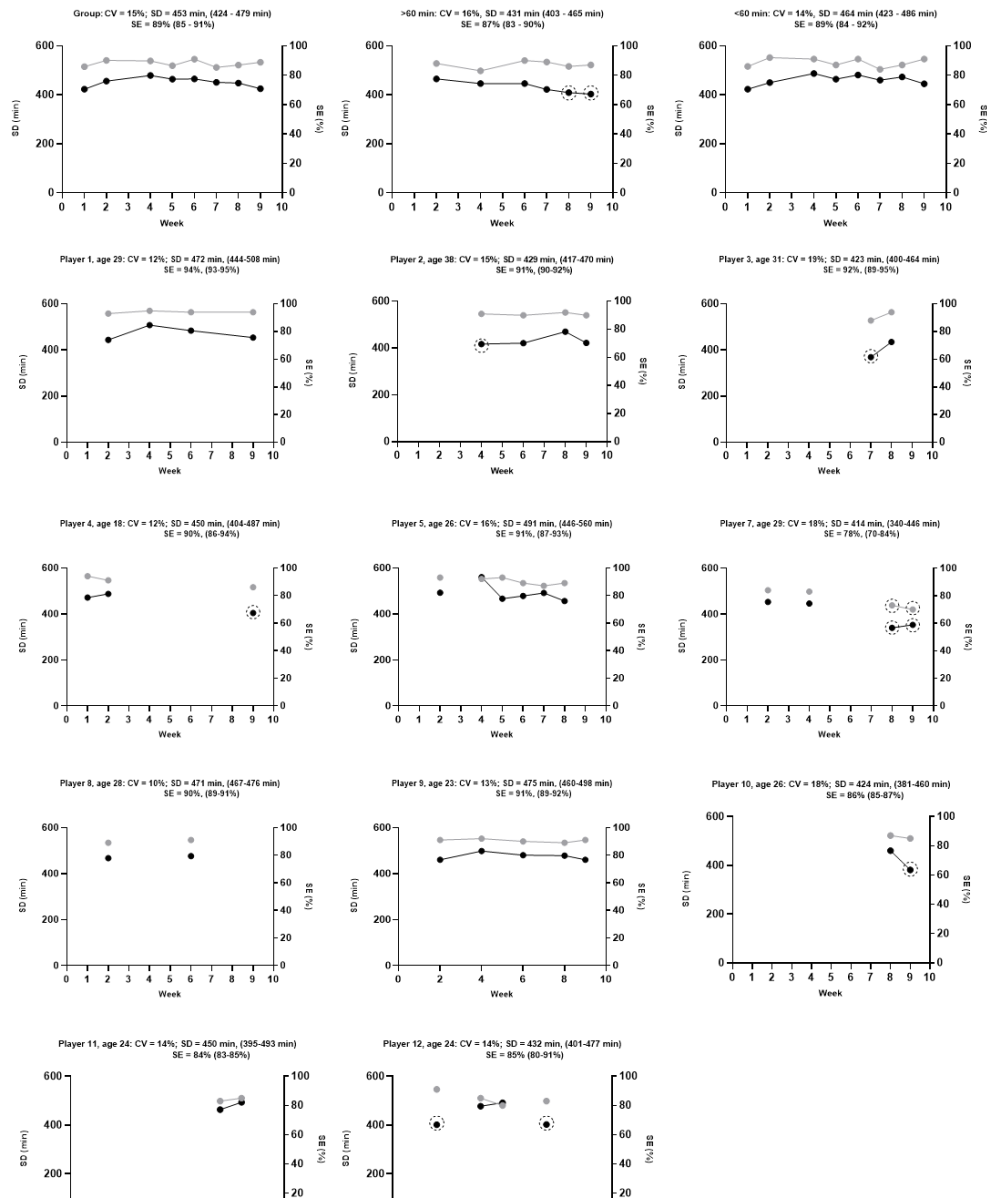


Figure 4 - Descriptive group data and individual (n = 12) responsiveness for sleep efficiency and sleep duration during 9 weeks in competitive phase in high-level soccer players. Horizontal black lines show group mean $\pm$ 95% confidence interval for each week. Coefficient of variation (CV), averages, maximum, and minimum values are also presented. The small circles represent the weeks where sleep duration and sleep efficiency were lower than the recommended amounts (i.e., sleep duration < 7 h and sleep efficiency < 75%), respectively. The black points represent sleep efficiency and grey points represent sleep duration.

Female youth soccer players' sleep quantity and quality have been found to be negatively correlated with increases in training load (Watson et al., 2017), as well occurs in this study. Several innate and adaptive immune functions are suppressed with increases in training load, including noticeably decreased neutrophil function, lymphocyte proliferation, and the quantity of circulating T cells (Assefa et al., 2015; Watson, 2015).

On the other hand, heart rate variability indexes, which represent parasympathetic or mixed sympathetic/parasympathetic modulation of heart rate, were not significantly affected by increasing fitness level (Singh et al., 2018). Physical exercise is known to significantly decrease HRV despite the long-term effects of endurance training. The autonomic nervous system, which controls the body's homeostatic processes, rapidly changes its output when the body moves from exertion to recovery (Nummela et al., 2016). It is well known that when exercise is stopped, vagal activity increases and sympathetic drive decreases (Berger et al., 2019).

It is therefore suggested that strategies for extending sleep duration in order to reach the recommended level may contribute to improving performance, mood and stress levels (Ohayon et al., 2017).

In addition to this negative relationship between PL and ln LF, it is also possible to do an individual analysis, where two players showed higher lnHF CV (players 3 and 4) and one player (player 4) show lnRMSSD CV and lnLF CV higher than the other players (>10%) (Flatt et al., 2017). To understand the importance of HRV indices, the lnRMSSD CV has been assessed in studies involving highly-trained athletes as a marker of variation in daily assessed lnRMSSD (Flatt et al., 2018). In one study of Flatt et al. (2017), the authors suggested that a high diurnal lnRMSSD CV (>10%) was positively associated with perceived fatigue and negatively associated with the physical fitness of

female soccer players. Moreover, another study found that diurnal lnRMSSD CV measured in swimmers can increase to values $>10\%$ during overload periods. This results from reduced vagal activity after intense training, followed by a parasympathetic rebound 48 hours later, which underscores lnRMSSD CV's value in assessing training adaptation. Elevated lnRMSSD CV could signify initial physiological stress, while minimal change indicates well-tolerated training. Athletes with higher fitness levels typically have quicker parasympathetic recovery and smaller lnRMSSD CV, which suggests severe physiological disturbance, potentially requiring intervention. In that study, a subject's baseline lnRMSSD CV influenced their response to overload ($>10\%$) exhibited the least favorable response to the overload phase and required more rest and reduced training.

At the other side, (F. Nakamura et al., 2016) concluded that players displaying higher values of lnRMSSD during each week of training presented lower perturbation of the cardiac autonomic system. Maintenance of high and relatively unchanged daily HRV values have been shown to facilitate greater cardiorespiratory fitness improvement in response to training than standardized training which was not guided by the HRV responses (Boullosa et al., 2012; Thorpe et al., 2017).

In the present study, as a group, lnRMSSD derived from the SWSE method displayed high average CV (11%). Moreover, individually, only one player (player 4) presented higher lnRMSSD CV and reduced lnRMSSD, contrasting with the remaining players. This player shows an increased lnRMSSD CV (19%) in week 9, where the TD and PL were higher than another weeks. Furthermore, it could be speculated that higher lnRMSSD CV linked with reduced average lnRMSSD during training and matches may be interpreted as a sign of overreaching (Flatt et al., 2016).

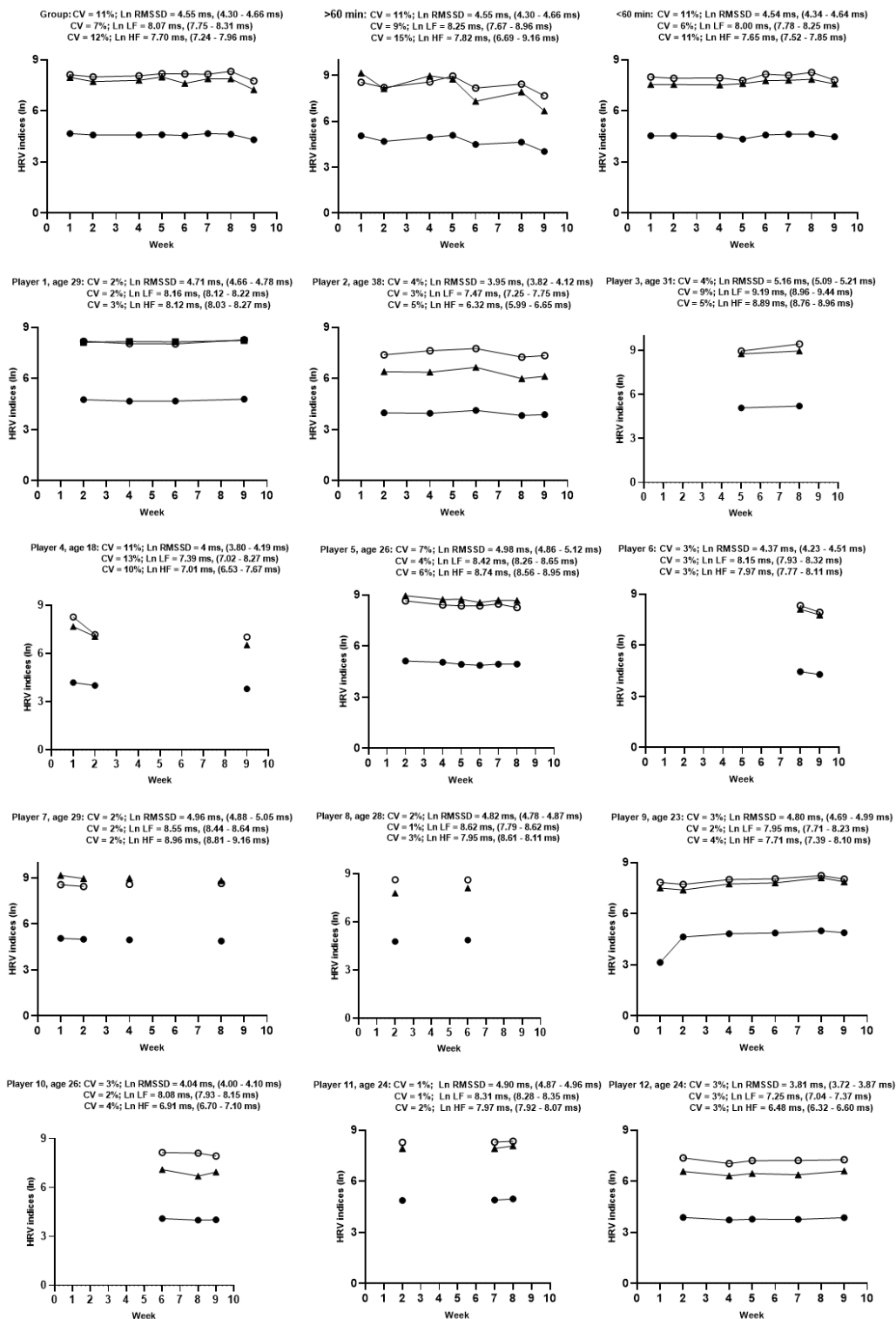


Figure 5 - Descriptive group data and individual ($n = 12$) responsiveness for lnRMSSD, lnLF and lnHF during 9 weeks in competitive phase in high-level male soccer players. Horizontal black lines show group mean $\pm$ 95% confidence interval for each week. Coefficient of variation (CV), averages, maximum, and minimum values are also presented. The black points represent lnRMSSD, the triangles represent lnLF and the white circles represent lnHF.

Throughout the 9-week period, the group's sleep duration remained steady, but when we specifically examined players participating over 60 minutes (in weeks 8 and 9), their sleep duration was lower than recommended (420 min). Notably, week 9 displayed the highest combination of match and training loads, which could be linked to our hypothesis. This finding aligns with earlier research suggesting that sleep duration might be impacted by the type of training day in the schedule (Robey et al., 2014; Silva, 2022) (Robey et al., 2014; Silva, 2022) as well as congested calendars. Consequently, this promote an accumulation of training and match loads that impairs recovery (Oliveira et al., 2023).

This link between type of day and sleep duration may be related to the preparation strategies for the match, as players may recognize the benefits of longer sleep time for recovery and performance, as well as the influence of the match on match-day (Costa et al., 2021). This finding is also corroborated by the observed *moderate* correlation between sleep duration and s-RPE, justifying periods of intensified training loads have been associated with higher disturbance levels in sleep (Silva, 2022).

This occurrence may be a result of overreaching, increased levels in muscular soreness, and pro-inflammatory responses (Halsen, 2014). Similarly, high training loads are also likely to induce similar physiological responses. Further research is required to determine the physiological effects of the observed relationship and how it may impact recovery and performance (Gurau et al., 2023).

Although sleep efficiency seemed less affected by training/match load, we found a *moderate* positive within-subject correlation between sleep efficiency and lnRMSSD. Just one player shows a lower sleep efficiency during two consecutive weeks (player 7).

Despite being a player who did not play many minutes in this week's match, sleep duration was also below recommendations for athletes (<420 min) (Fry & Rehman, 2022).

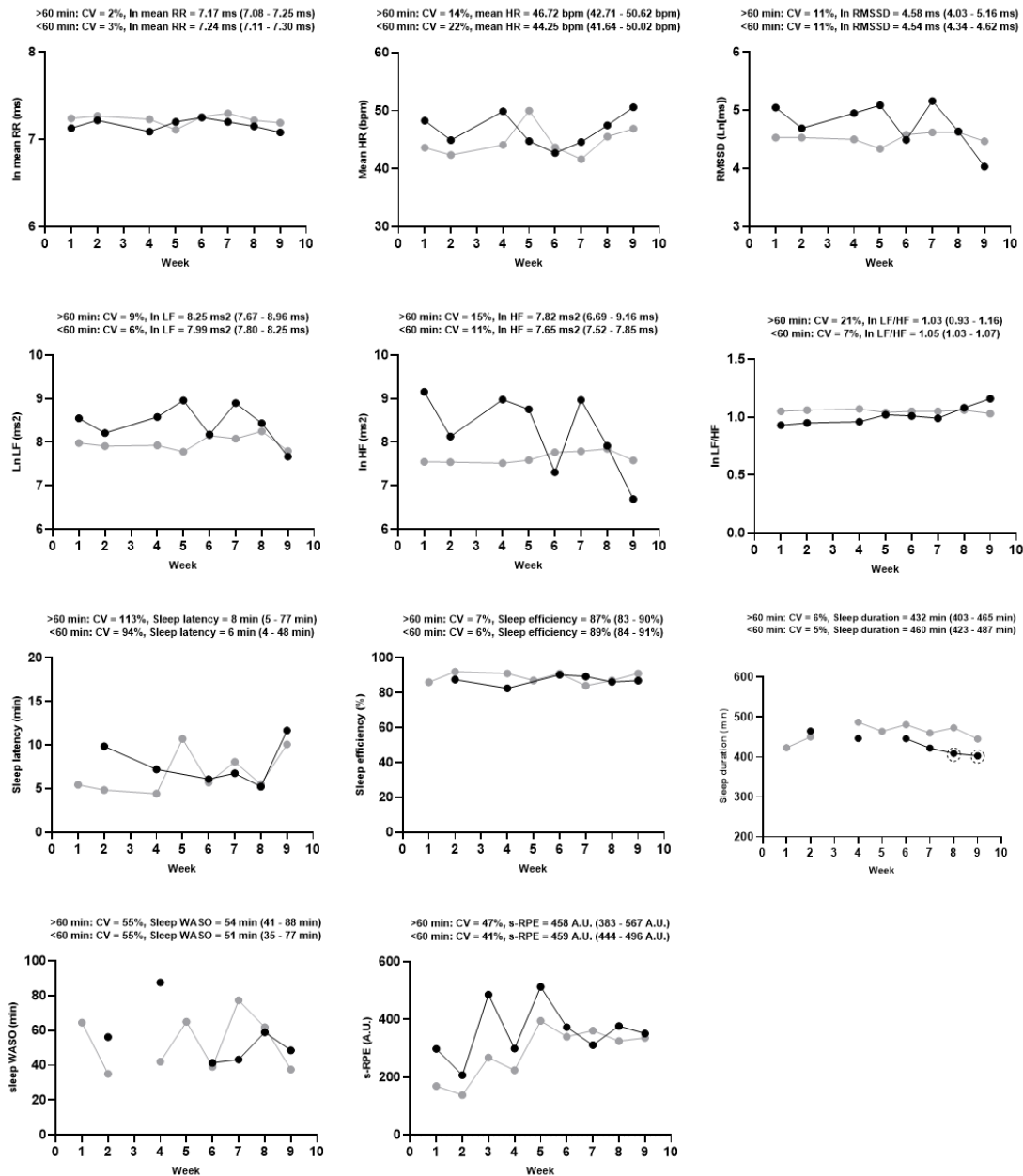


Figure 6 - Descriptive group data (n = 12) responsiveness for sleep and nocturnal HRV indices, training and match load parameters, and perceived ratings of wellbeing during 9 weeks in competitive phase in high-level male soccer players who played more of 60 minutes in matches over 9 weeks. Horizontal black lines show group mean $\pm$ 95% confidence interval for each week. Coefficient of variation (CV), averages, maximum, and minimum values are also presented. The small circles represent the weeks where sleep duration and sleep efficiency were lower than the recommended amounts (i.e., sleep duration < 7 h and sleep efficiency < 74%), respectively. The black points represent players which play more than 60 minutes in the match week and grey points represent players which play less than 60 minutes in the match week.

Moreover, the decrease in sleep duration and sleep efficiency in some weeks for each player can be related to the training or match schedule. Indeed, the impact of night schedule time on subsequent sleep was confirmed elsewhere (Hamlin et al., 2021). Costa et al. (2019) observed later sleep initiation and reduced sleep duration after evening matches compared to day matches, as well as that sleep onset took longer after evening matches than after-day matches. In other study, Costa et al. (2021) observed negative correlations between s-RPE and sleep duration ($r = -0.43$; $p < 0.001$) and sleep efficiency ($r = -0.25$; $p < 0.001$), as well as presented negative correlations between training impulse (TRIMP) and sleep duration ($r = -0.17$; $p = 0.02$) and sleep efficiency ($r = -0.20$; $p = 0.004$). In this study, the trainings usually start at the morning and most of the matches start after 18h. In contrast of the late schedules of matches, (Sargent, Halson, et al., 2014) shows a certain type of training schedule has a negative impact on sleep. On training days, the swimmers' first session started three hours before nine o'clock, and they slept 1.7 less hours than on rest days. This can disrupt the sleep indices once training days have different schedules of matches schedules. Besides, matches away from home can affect sleep is affected by many factors, e.g., training, competition, travel, sleeping environment (e.g. airplane, bus), regularity of sleep-wake schedules (social jet lag), light exposure and wake-length prior to sleep (Nedelec et al., 2018).

Based on the actigraphy findings to date, sleep efficiency is recommended for monitoring sleep quality (Claudino et al., 2019). In present study, the sleep efficiency CV was low ($CV < 10\%$), while the sleep duration CV oscillated between 10% and 20% for the group and for each player.

Table 4 – Within-subject correlation between sleep and nocturnal heart rate variability indices with session-rating of perceived exertion (s-RPE) and Player Load (PL) during 9 consecutive weeks of in-season competitive phase (n=12).

	s-RPE			Player Load		
	r (95% CI)	p	Description	r (95% CI)	p	Description
Sleep efficiency	0.01 (-0.19; 0.21)	0.91	Unclear	-0.01 (-0.25; 0.22)	0.92	Unclear
Sleep duration	0.4 (0.2; 0.59)	0.003	Moderate	-0.10 (-0.33; 0.14)	0.41	Unclear
lnRMSSD	0.13 (-0.06; 0.30)	0.18	Unclear	0.04 (-0.18; 0.26)	0.72	Unclear
lnLF	-0.13 (-0.31; 0.05)	0.16	Unclear	-0.23 (-0.43; -0.01)	0.04	Small
lnHF	-0.03 (-0.21; 0.15)	0.74	Unclear	-0.04 (-0.25; 0.19)	0.75	Unclear

Table 5 – Within-subject correlation between nocturnal heart rate variability indices with sleep efficiency and sleep duration during 9 consecutive weeks of in-season competitive phase (n=12).

	Sleep efficiency			Sleep duration		
	r (95% CI)	p	Description	r (95% CI)	p	Description
lnRMSSD	0.27 (0.11; 0.41)	0.001	Moderate	0.03 (-0.13; 0.19)	0.72	Unclear
lnLF	0.01 (-0.15; 0.18)	0.87	Unclear	0.10 (-0.07; 0.26)	0.24	Unclear
lnHF	-0.1 (-0.26; 0.06)	0.22	Unclear	0.08 (-0.09; 0.24)	0.36	Unclear

Furthermore, a *small* correlation was observed between PL and ln LF. This demonstrates the impact that training and match loads can have on HRV (Flatt & Esco, 2015). Although there is no correlation between HRV indexes and training and match loads, it was found that player 10 presents an *almost perfect* negative correlation between PL and lnLF for the week 9. In this case, player 10 showed in that week an increased TD and PL compared to other weeks. In the same week, it was also observed that this player slept 381 min, less than recommended (>420 min). The same episode occurs with players 3, 4, 5, 6, 8, 9 and 12, who showed a decrease in sleep duration values or HRV indices in the week where the TD and PL were highest. Although not all show correlations between sleep and HRV indices and training and match loads, this leads us to conclude that as highest is training and match load, higher could be the impact on sleep and/or HRV indices. Other study also reported higher internal training loads were positively correlated

with lnRMSSD CV in an individual perspective. In this way, it is intended a greater exercise tolerance had lower weekly allostatic perturbation, leading to smaller weekly changes in lnRMSSD, and in turn, lower CV values, as well as reduced perceived fatigue (Nakamura et al., 2023).

Conclusions

The present longitudinal study revealed a significant impact of training and match loads on sleep patterns and HRV measures. It was observed some players consistently experienced inadequate sleep durations on specific weeks, with one player enduring poor sleep quality for two consecutive weeks. Additionally, variations were noted in the nocturnal cardiac autonomic activity (lnRMSSD, lnLF, and lnHF) among some players. Notably, players who participated for more than 60 minutes in matches displayed heightened fluctuations in several metrics, including lnRMSSD, lnLF, lnHF, and s-RPE.

Moreover, this group experienced prolonged periods of inadequate sleep for two consecutive weeks, potentially contributing to the accumulation of physical and mental fatigue. In fact, the study emphasized the substantial individual differences in sleep patterns and HRV measures. This underscores the importance of adopting a personalized approach to monitor sleep, training, match loads, and recovery, allowing for a more nuanced understanding of how players navigate the challenges of high-intensity competitions.

Limitations

Considering the observational nature of this study, conclusions must be considered within its limitations. Factors like hormonal changes, pre-bed food consumption, light exposure, and electronic device use could have influenced sleep. Technical issues and missing data due to player non-compliance led to the exclusion of sleep diary data. Interpretation of CV values for sleep and HRV should be prudent due to missing data. The absence of a true baseline for sleep and HRV comparisons across the nine-week period is a limitation.

The results were grouped into weeks, with some days having different results for each variable (e.g., results on a match day to a player who play every match will be different for results on the recovery day). This average can remove some limitations in the interpretation of the results.

Practical applications

This longitudinal study of the sleep and nocturnal HRV responses of elite soccer players is intended to assist coaches and trainers in identifying sleep and HRV disturbances during official competitions, especially during periods of heavy congestion. In fact, identifying the optimal amount of sleep depends individually, but at a minimum, adult athletes who demonstrate an average sleep of less than 7 hours, likely warrant additional evaluation to identify their specific sleep barriers (Doherty et al., 2021).

Consequently, it is recommended that people who have experienced adverse effects due to inadequate sleep duration and quality of sleep should be offered the use of sleep hygiene techniques with the aim of improving their energy levels during the day and alertness at night (Suni & Rehman, 2023).

Longitudinal monitoring of training and competition intensity, stress, mood, sleep and fatigue improves the identification of those risks as well as helps track improvements in sleep health and performance in demanding competitions (Jones et al., 2017). Therefore, it is important for coaches to constantly encourage and discuss with players the importance of monitoring loads and health parameters in areas such as recovery periods during different matches and training restrictions if we are to avoid a high level of data loss.

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Chapter IV

General discussion and conclusions

4. General discussion

This observational study investigates the association between habitual sleep, nocturnal cardiac autonomic activity and training and match load in soccer players throughout a nine-week competitive season. We discovered that the majority of players slept for less than the suggested 420 minutes per night (7 hours) (Fry & Rehman, 2022), especially during weeks 8 and 9. Another key finding was the association between training and match load (s-RPE) and sleep duration. Due to the reported adverse impacts of shorter sleep duration or sleep deprivation on athletes' health and performance, our findings reinforce the necessity to create and apply personalized sleep strategies. (Dolezal et al., 2017). Finally, the training and match loads affect players' nocturnal cardiac autonomic activity, demonstrated negative *moderate* correlation.

Therefore, we predicted to be a negative within-subject relationship of sleep and HRV with training/match load, based on our data indicating an increase in variability of sleep and HRV measurements by soccer players throughout a nine-week competitive period. Since sleep duration had a positive correlation with s-RPE and PL as well as a negative relationship with lnLF.

According to Hausswirth et al. (2014), athletes' sleep habits may be significantly affected by training-related stress. For instance, it has been reported that exposure to intense training loads in athletes may increase the possibility of sleep disturbance and shorten sleep duration. Also, it can affect a few psychological factors and, consequently, impact an athlete's capacity to perform at their highest level, such as mood, anxiety, confidence, fatigue, levels of energy, and motivation (Cook & Charest, 2023; Khan et al., 2018). During periods of intense training loads, some athletes report having problems falling asleep, being restless while they sleep, and feeling heavy legs while they sleep (Lastella et al., 2015). (Watson et al., 2017) showed a negative association between sleep quantity and quality and training load in female youth soccer players, as well occurs in this study. Several innate and adaptive immune functions are suppressed with increases in training load, including noticeably decreased

neutrophil function, lymphocyte proliferation, and the quantity of circulating T cells (Assefa et al., 2015; Watson, 2015).

However, the degree of fitness did not substantially impact heart rate variability indices, which indicate parasympathetic or mixed sympathetic/parasympathetic regulation of heart rate (Pham et al., 2021). Despite the long-term effects of endurance training, physical exercise has been shown to significantly reduce HRV. When the body moves from exercise to recovery, an autonomic nervous system that controls its homeostatic processes makes rapid changes in output (Hynynen et al., 2010). It's common knowledge that vagal activity rises and sympathetic response decreases when exercising is interrupted (Berger et al., 2019). Accordingly, it is suggested that strategies for increasing the duration of sleep may contribute to improve performance, mood and tension levels in order to attain a level deemed appropriate (Ohayon et al., 2017).

In addition to this negative relationship between PL and ln LF, it is also possible to do an individual analysis, where two players showed higher lnHF CV (players 3 and 4) and one player (player 4) show lnRMSSD CV and lnLF CV higher than the other players (>10%) (Flatt et al., 2017). To understand the importance of HRV indices, the lnRMSSD CV has been assessed in studies involving highly-trained athletes as a marker of variation in daily assessed lnRMSSD (Flatt et al., 2018). In one study of Flatt et al. (2017), the authors suggested that a high diurnal lnRMSSD CV (>10%) was positively associated with perceived tiredness and negatively related to the physical fitness of female soccer players. In addition, another study has shown that in swimmers diurnal lnRMSSD CV values can increase >10% within periods of overload. This result demonstrates the value of ln RMSSD CVs for evaluation of training adjustment, which shows a reduction in vagal activity after intense training and a parasympathetic rebound within 48 hours. Elevated lnRMSSD CV could signify initial physiological stress, while minimal change indicates well-tolerated training. The faster parasympathetic recovery and smaller lnRMSSD CV, which suggests a serious physiological disturbance and may require intervention, are typically seen in athletes with higher fitness levels. In that study, a subject's baseline lnRMSSD CV influenced their response to overload (>10%) exhibited the least

favorable response to the overload phase and required more rest and reduced training.

At the other side, (F. Nakamura et al., 2016) concluded that the cardiac autonomic system has been less disturbed by players with higher values for lnSDRMS during each week of training. Maintenance of high and relatively stable daily HRV levels is shown to facilitate higher cardiorespiratory fitness enhancement in response to the training than standard exercises, which are not guided by an HSV response. (Boullosa et al., 2012; Thorpe et al., 2017).

In the present study, as a group, lnRMSSD derived from the SWSE method displayed high average CV (11%). Moreover, individually, only one player (player 4) presented higher lnRMSSD CV and reduced lnRMSSD, contrasting with the remaining players. This player shows an increased lnRMSSD CV (19%) in week 9, where the TD and PL were higher than the other weeks. Furthermore, it could be speculated that higher lnRMSSD CV linked with reduced average lnRMSSD during training and matches may be interpreted as a sign of overreaching (Flatt et al., 2016).

The sleep duration remained constant throughout the nine weeks for the group. However, players who played for over 60 minutes during Weeks 8 and 9 showed that their sleep time was shorter than recommended at 420 min per hour. In particular, there was a significantly higher combination of match and training activity at week 9 that may be related to our hypothesis. This finding is consistent with a prior study which suggested that the type of training day may have an impact on sleep duration (Robey et al., 2014; Silva, 2022), as well as congested calendars. Consequently, this promote an accumulation of training and match loads that impairs recovery (Oliveira et al., 2023).

The fact that players are able to recognise the benefit of longer sleep periods for recovery and performance, as well as having an impact on match-day can also be associated with this connection between type of day and period of sleep, which may relate to match preparation strategies (Costa et al., 2021). This finding is also corroborated by the observed *moderate* correlation between sleep duration and s-RPE, justifying periods of intensified training loads have been associated with higher disturbance levels in sleep (Silva, 2022).

This occurrence may be a result of overreaching, increased levels in muscular soreness, and pro-inflammatory responses (Halsen, 2014). Similarly, high training loads are also likely to induce similar physiological responses. Further studies are needed in order to establish the physiologic effects of the relationship and how it may affect recovery and performance (Gurau et al., 2023).

Although sleep efficiency seemed less affected by training/match load, we found a *moderate* positive within-subject correlation between sleep efficiency and lnRMSSD. Just one player shows a lower sleep efficiency during two consecutive weeks (player 7). Despite being a player who did not play many minutes in this week's match, sleep duration was also below recommendations for athletes (<420 min) (Fry & Rehman, 2022). In this way, a previous study found the sleep quality of female nurses was poor and this affected their autonomic nervous system (Hsu et al., 2021). Negative correlations were observed between sleep quality and low frequency and the low frequency/high frequency ratio ($r = -0.269$, $p < 0.05$; $r = -0.266$, $p < 0.05$).

Furthermore, a schedule of training or match may be related to the decrease in sleep time and sleeping effectiveness for each player over several weeks. In fact, elsewhere the effects of night time schedules on subsequent sleep have been confirmed (Hamlin et al., 2021). Costa et al. (2019) observed later sleep initiation and reduced sleep duration after evening matches compared to day matches, as well as that sleep onset took longer after evening matches than after-day matches. In other study, Costa et al. (2021) observed negative correlations between s-RPE and sleep duration ($r = -0.43$; $p < 0.001$) and sleep efficiency ($r = -0.25$; $p < 0.001$), as well as presented negative correlations between training impulse (TRIMP) and sleep duration ($r = -0.17$; $p = 0.02$) and sleep efficiency ($r = -0.20$; $p = 0.004$). In this study, the trainings usually start at the morning and most of the matches start after 5 p.m. Heishman et al. (2017) conclude the performance is suppressed with morning training and is associated with a decrease in self-reported quantity of sleep. However, this relationship needs to be analysed at future research.

In contrast of the late schedules of matches, other study shows a certain type of training schedule has a negative impact on sleep. On training days, the

swimmers' first session started three hours before nine o'clock, and they slept 1.7 less hours than on rest days (Sargent et al., 2014). This can disrupt the sleep indices once training days have different schedules of matches schedules. Besides, matches away from home can affect sleep is affected by many factors, e.g., training, competition, travel, sleeping environment (e.g. airplane, bus), regularity of sleep-wake schedules (social jet lag), light exposure and wake-length prior to sleep (Nedelec et al., 2018). Furthermore, night matches may be affecting sleep. Costa et al. (2019) found significantly decreased durations of sleep duration and efficiency after evening matches compared with day matches during an international tournament. Sargent and Roach (2016) looked at how elite Australian soccer players slept on the night immediately after a day match or the night after an evening match. The players started sleeping 2.5 hours after the evening match and got 2.1 hour less sleep than they did the night after the day match. In our study, athletes had matches starting, at least, at 5:30 pm, that is, the beginning of the night period. From the point of view of sports performance, it is a consensus that most physical performance rhythms reach a plateau between 3:00 pm and 9:00 pm (Chtourou & Souissi, 2012). Although it was not evaluated in this study, it is assumed that it is another reason that explains our results.

Based on the actigraphy findings to date, sleep efficiency is recommended for monitoring sleep quality (Claudino et al., 2019). In present study, the sleep efficiency CV was low ($CV < 10\%$), while the sleep duration CV oscillated between 10% and 20% for the group and for each player.

Furthermore, a *small* correlation was observed between PL and ln LF. This demonstrates the impact that training and match loads can have on HRV (Flatt & Esco, 2015). Although there is no correlation between HRV indexes and training and match loads, it was found that player 10 presents an *almost perfect* negative correlation between PL and lnLF for the week 9. In this case, player 10 showed in that week an increased TD and PL compared to other weeks. In the same week, it was also observed that this player slept 381 min, less than recommended (>420 min). The same episode occurs with players 3, 4, 5, 6, 8, 9 and 12, who showed a decrease in sleep duration values or HRV indices in the week where

the TD and PL were highest. Although not all show correlations between sleep and HRV indices and training and match loads, this leads us to conclude that as highest is training and match load, higher could be the impact on sleep and/or HRV indices. Other studies have also reported that in an individual context, higher internal training workload was associated with a positive correlation of InRMSSD CV. In that way, a greater tolerance to exercise is supposed to result in lower weekly allostatic fluctuations leading to less frequent InRMSSD changes and CV reductions as well as reduced perceived fatigue (Nakamura et al., 2023).

In conclusion, to gain insight into the unique responses of athletes and to enhance the timing of training and recovery protocols, it is imperative to conduct a thorough assessment of the athlete's performance, physical condition, and fatigue levels. Furthermore, adopting an individualized approach to athlete monitoring can facilitate more precise prescription of training regimens, effective fatigue management, and the provision of pertinent post-match recovery strategies, including interventions to optimize sleep hygiene.

4.1. Limitations of the study

Given applied research nature of this thesis several limitations can be pointed out:

- Despite affording players a period of acclimatization to the equipment designed to prevent disruptions in their training, match activities, or sleep patterns, some players did express instances of discomfort during the data collection process. Regrettably, such constraints inherent to the utilization of heart rate monitors and wristwatches for data collection were unavoidable;
- Although the existence of a relatively standardized recommended sleep duration of approximately 7-9 hours per day for the majority of the population, it is worth noting that many individuals exhibit varying sleep needs per night (Estevan et al., 2021). These individuals are typically categorized as either short or long sleepers (Emami, 2021). Short sleepers may experience a satisfactory perception of sleep quality and recovery

status with only a few hours of sleep, whereas long sleepers require nine or more hours of sleep to achieve a sense of full recovery and restfulness. However, it is important to acknowledge that this categorization was not undertaken, thus representing a notable limitation of the current thesis;

- As this study was conducted in a real-world setting, various uncontrolled variables beyond match and training schedules and loads may have influenced the athletes' sleep patterns. These factors include the consumption of caffeine, engagement with social media after bedtime, use of electronic devices, exposure to light during the daytime, room arrangements, room temperature, and individual perceptions of stress and well-being. In fact, it is essential to incorporate additional metrics, such as well-being assessments, to provide context and enhance the interpretation of HRV changes (Bellenger et al., 2016; Jones et al., 2018). Regrettably, due to constraints related to time and technology, data on well-being, fitness levels, and perceived stress were not collected. We acknowledge this as a significant limitation of the present study, as it hinders a more comprehensive understanding of HRV values for each player during the nine-week period;
- As a result of missing data on certain days, it is important to exercise caution when interpreting the CV values pertaining to sleep and HRV. Moreover, it is imperative to acknowledge that this study had limitations, as it did not establish a definitive "true baseline" to facilitate potential comparisons of sleep and nocturnal HRV across the nine-week tournament period. Lastly, while the s-RPE method is known for its simplicity, validity, and reliability, it is worth noting that this study did not incorporate other forms of internal training and match load monitoring (e.g., heart rate monitors), which could have offered objective data for a more comprehensive interpretation of players' physiological responses;
- The results were grouped into weeks, with some days having different results for each variable (e.g., results on a match day to a player who play

every match will be different for results on the recovery day). This average can remove some limitations in the interpretation of the results.

- The sample of the total group presented 757 results. However, when we separate the group for players who play more than 60 minutes (group 1) and players who play less than 60 minutes (group 2), it was observed a huge difference between the groups (group 1 with 208 results and group 2 with 549 results). In this way, some results may not be adjusted to the number of samples and may lead to less specific conclusions.
- Lastly, it is essential to underscore that due to the potential impact of individual traits, such as aerobic fitness, on the correlation between RPE and HR, our findings should not be extrapolated to other samples of soccer players.

4.2. Future research

The results of the current thesis have also suggested a few matters for future study, including:

- It is recommended to investigate sleep patterns and (nocturnal) HRV together rather than independently, particularly when training and competitions take place close to bedtime. To further understand fatigue and recovery in soccer players, it is advised to look at the relationships (such as correlations) between recovery status variables (such as sleep pattern and nocturnal HRV) and internal and external load.
- Future research should explicitly analyse sleep patterns and HRV responses concerning playing time, given the association between exercise duration and corresponding load.
- This thesis only included observational and descriptive investigations. Therefore, it may be crucial that future studies evaluate various sleep hygiene practices (such as napping) in conjunction with performance factors in professional soccer players, particularly when practice and game days are near tonight.

- The article presented players who training at the morning and usually play at night. This relationship is unknown and would be interesting to find some results to monitoring training of elite soccer players.
- For identifying changes in performance in soccer, integration of tactical and technical indicators with recovery status data is crucial. True soccer performance needs to be considered as a construct, even though as sports scientists we may tend to focus on the physical and physiological components of sport. A coach can be interested in a change in technical or tactical factors that, in their opinion, might have more impact on the result of a match than physical and physiological factors.

4.3. Practical applications

The primary purpose of this thesis was to expand the current scientific knowledge in elite soccer, especially in areas related to autonomic cardiac function and sleep patterns. The following practical applications should be considered by practitioners in the field:

- A notable of prior research has been dedicated to this area of study, demanding distinctive attention. The meticulous tracking of sleep patterns and recovery metrics throughout the duration of a competitive season has the potential to offer valuable support to coaches and sports scientists in their efforts to make well-informed decisions concerning training and recovery strategies tailored to elite soccer athletes.
- Recent research has underscored the significance of the ANS status in shaping training responses. This is primarily because the assessment of ANS function effectively encapsulates crucial insights into the acute and chronic physiological processes that transpire before, during, and after exercise training sessions. Our study findings unequivocally affirm that the monitoring of HRV during nighttime can serve as a straightforward and non-invasive tool for evaluating, on an individualized basis. In this way, the effects of training and match loads can be performed at different times of the day and in different environments.

- Soccer players follow strict match and practice schedules during intense competitive periods, which can cause them to get less sleep overall. The recovery process can then be affected by this reduction, especially if training and competitions occur right before bed. It's remarkable that reports from Portugal have regularly indicated that soccer matches are frequently scheduled for after 5 p.m. Given this situation, it is suggested for players and coaches to carefully plan training sessions in order to improve sleep patterns, thus maximizing the recovery process, in addition to coordinating the training program with match schedules.
- The inclusion of actigraphic variables and HRV indices contributes to the creation of an extensive dataset that includes cardiac autonomic function and sleep patterns. We now have a more complete understanding of an athlete's recovery status due to the combination of these data. Additionally, both sets of indices can be used to improve the accuracy of tracking training and match loads. These evaluations can be easily carried out at locations where athletes train and compete or inside their homes. It is significant that these metrics can be incorporated into a team's regular practices, simplifying practical adoption in the setting of professional sports.
- The significant individual variation in sleep and HRV points to the need for load monitoring, recovery treatments, and individualized sleep strategies in team sports. Team rotation occurs frequently when matches are played successively in a competitive environment. Some players, though, will frequently be required to participate in many matches. To make sure that every player is at their peak performance for every match in these circumstances, individualized recovery and monitoring strategies may be necessary.
- Longitudinal monitoring of training and competition intensity, sleep, fatigue (e.g., via HRV), stress and mood not only help identify those at risk but can also help track improvements in sleep, health and performance during demanding competitions.

5. Conclusions

Based on the conclusions of each of the studies included in this dissertation, the following major conclusions can be outlined:

- During nine weeks of in-season, some elite soccer players presented less sleep than recommended in some weeks. Moreover, one player had a bad sleep quality for two consecutive weeks. Additionally, most players seemed to present small fluctuations in nocturnal cardiac autonomic activity.
- Overall, elite soccer players from the first division soccer of Portugal seemed to be highly resilient to training and match loads as a group. However, this study highlighted the substantial individual variability in sleep and HRV measures, suggesting the adoption of an individual approach to monitor sleep, training and match loads, and recovery, to better understand how players cope with highly demanding competitions.
- As a group, players who played more than 60 minutes presented more oscillations compared to players who played less than 60 minutes (i.e., lnRMSSD, lnLF, lnHF, s-RPE). Furthermore, players who played more than 60 minutes showed a total time of sleep below than recommended for two consecutive weeks.
- The time of day for soccer practice and match location may cause disruption in sleep patterns.
- Players can disrupt sleep and HRV measures, even if they play in match week. This can explain the load accumulations and special individual attention through fatigue markers.
- This study concluded training and match loads impact sleep and HRV measures, as well as sleep efficiency impact in lnRMSSD, for a nine-week in-season period.

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Appendixes

Ethical approval



Comissão de Ética

Declaração

Para os devidos efeitos declara-se que o projeto CEFAD 14 2020 intitulado “Heart Rate Variability, Sleep and Wellness responses to training and match load in an Elite and sub-Elite soccer team”, submetido à Comissão de Ética da Faculdade de Desporto da Universidade do Porto por Miguel José Ferreira Andrade Cid, foi aprovado por ter em conta os requisitos éticos recomendados.

Porto, 30 de setembro de 2020.

A Presidente da Comissão de Ética

Zélia Maria Matos de Almeida Roque Pinto