



ESCUELA INTERNACIONAL DE DOCTORADO

PROGRAMA DE DOCTORADO EN BIOTECNOLOGÍA, MEDICINA Y CIENCIAS BIOSANITARIAS

UNIVERSIDAD FRANCISCO DE VITORIA

**EFFECTOS DEL FENOTIPO CIRCADIANO, MOMENTO DEL DÍA Y EL
SEXO COMO FACTORES MODULADORES DEL RENDIMIENTO
NEUROMUSCULAR EN JUGADORES DE VOLEIBOL**

TESIS DOCTORAL

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2026, Madrid



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TESIS DOCTORAL

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Per aspera ad astra.

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ABREVIATURAS, SIGLAS O ACRÓNIMOS

Abreviaturas / Siglas / Acrónimos	Término completo
ATP	Adenosine Triphosphate (Adenosín trifosfato)
ATP-PC	Adenosine Triphosphate–Phosphocreatine System (Sistema de adenosín trifosfato–fosfocreatina)
BMAL1	Brain and Muscle ARNT-Like 1 (Proteína tipo ARNT 1 de cerebro y músculo)
CEA	Stretch-Shortening Cycle (Ciclo estiramiento- acortamiento)
CLOCK	Circadian Locomotor Output Cycles Kaput (Gen reloj circadiano CLOCK)
CMJ	Countermovement Jump (Salto con contramovimiento)
CRY	Cryptochrome genes (Genes criptocromo)
DLMO	Dim Light Melatonin Onset (Inicio de la secreción de melatonina en condiciones de baja luz)
MEQ	Morningness–Eveningness Questionnaire (Cuestionario de matutinidad–vespertinidad)
MCTQ	Munich Chronotype Questionnaire (Cuestionario de cronotipo de Múnich)
NSQ	Suprachiasmatic Nucleus (Núcleo supraquiasmático)
PC	Phosphocreatine (Fosfocreatina)
PER	Period genes (Genes PER o genes “period”)
RFD	Rate of Force Development (Tasa de desarrollo de la fuerza)
SJ	Squat Jump (Salto desde sentadilla)

RESUMEN

La presente Tesis Doctoral se enmarca en el análisis de la influencia del momento del día, el fenotipo circadiano (cronotipo) y el sexo sobre el rendimiento neuromuscular en jugadores de voleibol. El rendimiento en deportes de equipo, y particularmente en disciplinas de carácter intermitente y de alta intensidad como el voleibol, depende de la capacidad para generar elevados niveles de fuerza y potencia muscular en breves intervalos de tiempo, así como de su mantenimiento bajo condiciones de fatiga. Desde una perspectiva integradora, el rendimiento neuromuscular no puede entenderse como una variable aislada, sino como el resultado de la interacción dinámica entre factores fisiológicos, contextuales e interindividuales.

En este marco, la cronobiología ha emergido como un campo de creciente relevancia al evidenciar que múltiples procesos fisiológicos determinantes del rendimiento neuromuscular, como la temperatura corporal, la activación neuromuscular o la secreción hormonal, presentan oscilaciones rítmicas a lo largo del día reguladas por el sistema circadiano. En este sentido, existe evidencia consistente que indica que estos determinantes neurofisiológicos varían a lo largo del día, alcanzando sus valores máximos generalmente en la franja vespertina. No obstante, pese a la solidez del marco teórico existente, la evidencia empírica presenta limitaciones relevantes, ya que proviene principalmente de estudios desarrollados en pruebas de laboratorio aisladas, que no reflejan las demandas reales de la competición o desarrollados en deportes individuales, lo que dificulta su extrapolación a deportes de equipo. Asimismo, la interacción entre el sistema circadiano y factores de variabilidad interindividual, como el fenotipo circadiano (cronotipo) o el sexo biológico, permanece insuficientemente explorada, especialmente en contextos aplicados como el voleibol. La falta de consenso sobre si las variaciones del rendimiento neuromuscular a lo largo del día son similares en todos los deportistas o dependen de características individuales limita la precisión en la evaluación del rendimiento y en la planificación de las cargas de entrenamiento.

En este sentido, la presente Tesis Doctoral analiza la influencia del momento del día sobre el rendimiento neuromuscular en voleibol, así como el papel del cronotipo y del sexo como posibles moduladores de esta respuesta en contextos aplicados de rendimiento. Para abordar este planteamiento, la tesis se estructura como un compendio de cuatro estudios, diseñados de forma complementaria para dar respuesta al problema de investigación.

El **Estudio 1**, consistente en una revisión sistemática con meta-análisis, tuvo como objetivo examinar el efecto del momento del día sobre el rendimiento neuromuscular en deportistas semiprofesionales o profesionales de deportes de equipo. Los resultados mostraron que el rendimiento neuromuscular en tareas explosivas, como el salto con contramovimiento (CMJ) y la agilidad (evaluada mediante pruebas como el *zig-zag* o el *T-test*), es significativamente superior en horario vespertino (16:00-20:00 h) en comparación con el horario matutino (7:00-10:00 h). Por el contrario, no se observaron diferencias significativas en manifestaciones neuromusculares como la fuerza isométrica o el salto sin contramovimiento (SJ). Estos hallazgos sugieren que la influencia del momento del día no es homogénea, sino dependiente del tipo de tarea, siendo más pronunciada en acciones que implican el uso del ciclo de estiramiento-acortamiento y una mayor demanda de coordinación neuromuscular, como los cambios de dirección o los saltos con fase excéntrica previa.

El **Estudio 2** tuvo como objetivo analizar el efecto del momento del día y del cronotipo sobre el rendimiento neuromuscular en jugadoras de voleibol semiprofesionales. Los resultados evidenciaron un rendimiento superior en horario vespertino en variables específicas del juego, como la velocidad de remate, así como en pruebas de rango de movimiento (*Straight Leg Raise Test*), equilibrio dinámico (*Star Excursion Balance Test*) y agilidad (*Modified Agility T-test*). En relación con el cronotipo, no se observaron efectos significativos sobre el rendimiento neuromuscular, por lo que su asociación fue limitada en comparación con el efecto del momento del día. En el contexto específico del voleibol semiprofesional, estos resultados sugieren que la influencia del sistema circadiano se refleja principalmente a través de las variaciones diurnas, con independencia del cronotipo de las jugadoras.

El **Estudio 3** analizó la influencia del fenotipo circadiano (cronotipo) en jugadores masculinos de voleibol, así como su interacción con el momento del día. Los resultados mostraron una influencia limitada del cronotipo sobre el rendimiento neuromuscular, sin observarse interacciones significativas con el momento del día. No obstante, se halló un rendimiento superior en horario vespertino en variables de potencia del tren inferior, específicamente en el SJ, CMJ y el salto de remate (*spike jump*), así como en la velocidad de remate en apoyo (*standing spike test*). Estos resultados sugieren que, en jugadores de voleibol, el momento del día tiene un papel predominante sobre el cronotipo en la expresión del rendimiento neuromuscular.

El **Estudio 4** tuvo como objetivo analizar las diferencias entre sexos en la variación diurna del rendimiento neuromuscular. Los resultados indicaron que, si bien tanto hombres como mujeres alcanzaron su mejor rendimiento en horario vespertino, las diferencias en la variación diurna entre sexos fueron reducidas y dependientes del tipo de tarea, con una variación ligeramente mayor en los hombres en tareas de carácter explosivo, como el CMJ o el salto de remate. Por el contrario, en las mujeres se observó una menor variación del rendimiento neuromuscular a lo largo del día. En conjunto, estos hallazgos indican que, aunque existen diferencias en la variación diurna del rendimiento neuromuscular, el sexo biológico ejerce una influencia limitada sobre la respuesta diurna del rendimiento en jugadores y jugadoras de voleibol.

En conjunto, los resultados de la presente Tesis Doctoral permiten concluir que, entre los factores analizados, el momento del día es uno de los principales determinantes de la variación del rendimiento neuromuscular en voleibol, con un efecto más evidente en tareas que implican la producción de fuerza y potencia, como los saltos y las acciones de remate. Por el contrario, el cronotipo mostró una influencia limitada sobre estas variaciones, mientras que el sexo presentó un efecto reducido y dependiente del tipo de tarea. Desde una perspectiva aplicada, estos hallazgos sugieren la conveniencia de considerar el momento del día en la planificación del entrenamiento y en la evaluación del rendimiento neuromuscular, mientras que el cronotipo y el sexo parecen desempeñar un papel secundario en este contexto.

Palabras clave: Ritmos circadianos; Cronobiología; Rendimiento atlético; Factores sexuales; Fuerza muscular; Deportes de equipo.

ABSTRACT

This Doctoral Thesis is framed within the analysis of the influence of time of day, circadian phenotype (chronotype), and sex on neuromuscular performance in volleyball players. Performance in team sports, particularly in intermittent and high-intensity disciplines such as volleyball, depends on the ability to generate high levels of force and power in short time intervals, as well as to sustain performance under fatigue conditions. From an integrative perspective, neuromuscular performance cannot be understood as an isolated variable, but rather as the result of the dynamic interaction between physiological, contextual, and inter-individual factors.

Within this framework, chronobiology has emerged as a field of growing relevance, as multiple physiological processes that determine performance, such as body temperature, neuromuscular activation, and hormonal secretion, exhibit rhythmic oscillations throughout the day regulated by the circadian system. In this sense, there is consistent evidence indicating that these neurophysiological determinants (Chtourou & Souissi, 2012; Hayes et al., 2010) vary throughout the day, typically reaching peak values in the late afternoon. However, despite the strength of the existing theoretical framework, empirical evidence presents relevant limitations, as it mainly derives from studies conducted in isolated laboratory conditions or in individual sports, which do not reflect the real demands of competition, thereby limiting its extrapolation to team sports. Furthermore, the interaction between the circadian system and inter-individual variability factors, such as chronotype and biological sex, remains insufficiently explored, especially in applied contexts such as volleyball. The lack of consensus regarding whether daily performance variations are similar across athletes or depend on individual characteristics limits the accuracy of performance assessment and training load planning.

In this context, the present Doctoral Thesis aims to analyze the influence of time of day on neuromuscular performance in volleyball, as well as to examine the role of chronotype and sex as potential modulators of this response in applied performance settings. To address this objective, this thesis by compendium was developed, consisting of four studies designed in a complementary manner to address the research problem.

Study 1, consisting of a systematic review with meta-analysis, aimed to examine the effect of time of day on neuromuscular performance in semi-professional or professional

team sport athletes. The results showed that performance in explosive tasks, such as countermovement jump (CMJ) and agility (assessed through tests such as the zig-zag or T-test), is significantly higher in the afternoon (16:00–20:00 h) compared to the morning (07:00–10:00 h). In contrast, no significant differences were observed in neuromuscular manifestations such as isometric strength or *squat jump* (SJ). These findings suggest that the influence of time of day is not homogeneous, but rather task-dependent, being more pronounced in actions involving the stretch-shortening cycle and higher neuromuscular coordination demands, such as changes of direction or jumps with a preceding eccentric phase.

Study 2 aimed to analyze the effect of time of day and chronotype on neuromuscular performance in semi-professional female volleyball players. The results showed superior performance in the afternoon (16:00–20:00 h) in sport-specific variables, such as spike velocity, as well as in range of motion (Straight Leg Raise Test), dynamic balance (Star Excursion Balance Test), and agility (Modified Agility T-test). Regarding chronotype, no significant effects were observed on neuromuscular performance, indicating a limited influence compared to time of day. In the specific context of semi-professional volleyball, these findings suggest that the influence of the circadian system is mainly reflected through daily performance variations, regardless of players' chronotype.

Study 3 analyzed the influence of chronotype in male volleyball players, as well as its interaction with time of day. The results showed a limited influence of chronotype on neuromuscular performance, with no significant interactions observed with time of day. However, higher performance was found in the afternoon in lower-body power variables, specifically in *squat jump* (SJ), countermovement jump (CMJ), and *spike jump*, as well as in ball velocity during the standing spike test. These findings suggest that, in volleyball players, time of day plays a more predominant role than chronotype in the expression of neuromuscular performance.

Study 4 aimed to analyze sex differences in the diurnal variation of neuromuscular performance. The results indicated that, although both men and women reached their peak performance in the afternoon, differences in diurnal variation between sexes were small and task-dependent, with slightly greater variation in men in explosive tasks, such as CMJ or *spike jump*. In contrast, women showed lower variation in performance throughout the day. Overall, these findings indicate that, although differences in diurnal variation exist,

biological sex has a limited influence on the diurnal response of neuromuscular performance in volleyball players.

Overall, the results of this Doctoral Thesis allow us to conclude that, among the analyzed factors, time of day is the main determinant of neuromuscular performance variation in volleyball, with a more pronounced effect in tasks involving force and power production, such as jumps and spiking actions. In contrast, chronotype showed a limited influence on these variations, while sex presented a reduced and task-dependent effect. From an applied perspective, these findings highlight the importance of considering time of day in training planning and neuromuscular performance assessment, whereas chronotype and sex appear to play a secondary role in this context.

Keywords: Circadian rhythms; Chronobiology; Athletic performance; Sex factors; Muscle strength; Team Sports.

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INTRODUCCIÓN

1. INTRODUCCIÓN

1.1 Características del voleibol de competición

El voleibol es una disciplina deportiva practicada en más de 220 países y reconocida como deporte olímpico desde los Juegos Olímpicos de Tokio de 1964 (Reeser, 2003). En el contexto español, este deporte de equipo cuenta con un total de 121.711 licencias federativas, de las cuales 36.054 corresponden a la categoría masculina y 85.657 a la femenina. Esta cifra representa el 2.82% del total de licencias federativas deportivas, situándolo como el tercer deporte de equipo más practicado en España (Anuario de Estadísticas Deportivas, 2025). El voleibol es un deporte colectivo caracterizado por acciones de cooperación-oposición (Moreno, 2005), que enfrenta a dos equipos de seis jugadores en una pista de 18 x 9 m, dividida por una red cuya altura varía según el sexo (2.43 m en categoría masculina y 2.24 m en femenina). El objetivo del juego es enviar el balón al campo contrario con la intención de que toque el suelo, evitando que esto ocurra en el propio campo. Los partidos se disputan al mejor de cinco sets; los cuatro primeros se juegan a 25 puntos y, en caso necesario, un quinto set a 15 puntos, siempre con una diferencia mínima de dos puntos para obtener la victoria (García-de-Alcaraz et al., 2019).

En el contexto competitivo, el voleibol requiere la integración coordinada de componentes técnico-tácticos, físicos y psicológicos para alcanzar niveles óptimos de rendimiento neuromuscular. En este sentido, el voleibol moderno ha experimentado una transformación profunda desde finales de los años noventa. Un hito clave fue la introducción en 1999 del sistema de punto por jugada o *rally-point scoring*, un formato donde cada acción suma directamente al marcador independientemente de quién posea el saque. Este cambio, unido a la especialización de las posiciones de juego (como el líbero o el opuesto), ha derivado en un aumento significativo de la velocidad y la intensidad de las acciones, elevando las demandas técnico-tácticas, físicas y psicológicas en el alto rendimiento (Marcelino et al., 2011).

1.2 Variables antropométricas, físicas y fisiológicas determinantes en el rendimiento en voleibol

Como ocurre en otros deportes, el rendimiento neuromuscular en voleibol está condicionado por una combinación de factores antropométricos, físicos y fisiológicos que influyen directamente en la capacidad del jugador o la jugadora para ejecutar con eficacia y éxito las acciones técnicas y tácticas propias del juego (Sheppard et al., 2008).

Desde una perspectiva morfológica, una mayor estatura, envergadura y longitud de los segmentos corporales constituyen factores determinantes para el éxito en el juego. Estas variables proporcionan una ventaja que permite alcanzar mayores alturas de contacto con el balón y cubrir un espacio superior en la red, optimizando tanto el alcance ofensivo en el remate como la eficacia en la interceptación defensiva durante el bloqueo (Lidor & Ziv, 2010). Debido a estas demandas, la especialización por posiciones ha consolidado perfiles antropométricos diferenciados; mientras que los centrales y atacantes presentan mayores valores de estatura y envergadura para maximizar el juego cerca de la red, los líberos y colocadores tienden a presentar perfiles ligeramente más bajos, priorizando cualidades relacionadas con la agilidad, la velocidad de reacción y la precisión técnica (Lidor & Ziv, 2010; Palao et al., 2014). Más allá de estas dimensiones estructurales, el rendimiento neuromuscular depende también de la composición corporal y del somatotipo, que define la morfología del deportista según el predominio de grasa, músculo o delgadez. Estas variables, a diferencia de las anteriores, son modificables mediante el entrenamiento y la nutrición, permitiendo ajustar el estado físico del jugador a lo largo de la temporada. Los jugadores de élite suelen presentar un somatotipo con predominancia de la mesomorfia, caracterizado por niveles optimizados de masa muscular magra y un bajo porcentaje graso (Duncan et al., 2006; Peña et al., 2018). Si bien estas características no determinan de forma directa la capacidad de producir fuerza o potencia, pueden contribuir a optimizar la producción y transferencia de fuerza en acciones explosivas propias del juego.

Desde una perspectiva física, el rendimiento neuromuscular en voleibol está condicionado por la capacidad para realizar esfuerzos intermitentes de alta intensidad y corta duración, entre los que destacan la capacidad de salto vertical y la velocidad de golpeo como determinantes críticos de la eficacia en el remate, el bloqueo y el saque (Lidor & Ziv, 2010; Sheppard et al., 2008), siendo claves la fuerza y potencia muscular, la velocidad y la coordinación intermuscular e intramuscular (Suchomel et al., 2016). La capacidad de salto vertical ha sido ampliamente identificada como uno de los factores clave tanto en acciones ofensivas (p. ej., remate y saque) como defensivas (p. ej., bloqueo), ya que una mayor capacidad de salto permite contactar el balón a una mayor altura respecto a la red, incrementando el ángulo de ataque en el remate y la capacidad de interceptación en el bloqueo, lo que la convierte en una capacidad física estrechamente relacionada con el rendimiento competitivo (García-de-Alcaraz et al., 2020; Leeuw et al.,

2022; Sheppard et al., 2008). Se estima que un jugador profesional puede realizar entre 60 y 120 saltos por partido, dependiendo de su posición específica en el juego (García-de-Alcaraz et al., 2020; Sattler et al., 2015). Esta elevada carga de saltos requiere un adecuado desarrollo del ciclo de estiramiento-acortamiento (CEA) (Iranpour et al., 2025) así como una elevada tasa de desarrollo de la fuerza (RFD) (Maffiuletti et al., 2016), dado que las acciones de salto implican la producción de altos niveles de fuerza en intervalos temporales muy reducidos. En este contexto, la función de la unidad músculo-tendinosa es fundamental para la reutilización de energía elástica durante el despegue (Suchomel et al., 2016); no obstante, la acumulación de estos esfuerzos a lo largo del encuentro genera una fatiga neuromuscular que puede reducir de forma progresiva la altura máxima de salto (Gathercole et al., 2015; Sheppard et al., 2007). Esta pérdida de capacidad de salto vertical puede comprometer directamente la eficacia técnica en los sets decisivos, limitando la capacidad del jugador para superar el bloqueo o interceptar ataques rivales (García-de-Alcaraz et al., 2020).

Por otro lado, la velocidad de golpeo constituye otro determinante físico relevante del rendimiento en voleibol, ya que influye directamente en la eficacia de las acciones ofensivas, especialmente en el remate y el saque. En particular, el remate es considerado la acción de ataque más determinante, siendo responsable de más del 50% de los puntos obtenidos en un encuentro (Costa et al., 2024; Marcelino et al., 2011; Palao & Valadés, 2012). Desde el punto de vista biomecánico, el remate implica una secuencia compleja de movimientos que integra carrera de aproximación, salto, golpeo y aterrizaje, ejecutados en un intervalo temporal muy reducido ($\approx 1,78$ segundos en hombres y 2,18 segundos en mujeres) (Fuchs et al., 2019). Su eficacia mecánica depende fundamentalmente de la interacción entre la altura de contacto, la trayectoria y, especialmente, la velocidad de salida del balón; un incremento en esta última reduce el tiempo de reacción del sistema defensivo rival (bloqueo y defensa de campo), maximizando así la probabilidad de éxito (Palao & Valadés, 2009; Reeser et al., 2010). De igual manera, la velocidad de golpeo es un factor crítico en el saque, especialmente en el saque en salto en potencia. La velocidad del golpeo del balón tiene por objetivo romper la organización del sistema de recepción contrario, facilitando la consecución directa del punto (*ace*) o limitando las opciones tácticas del colocador rival y por tanto del ataque rival (Reeser et al., 2010). En consecuencia, la capacidad de imprimir altas velocidades al balón en ambas acciones técnicas, el remate y el saque, resulta un factor

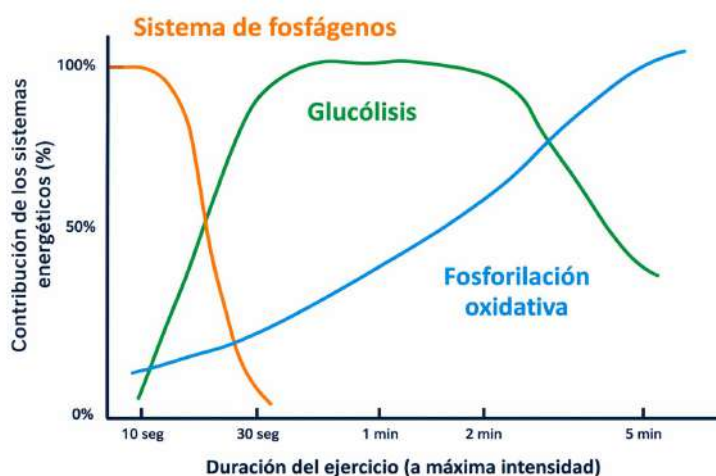
diferencial para el dominio de la fase ofensiva en el voleibol de élite (Baena-Raya et al., 2021).

Desde el punto de vista fisiológico, el voleibol es un deporte de equipo de carácter intermitente y alta intensidad, caracterizado por la alternancia de esfuerzos breves, explosivos y repetitivos con pausas de corta duración (Rebelo et al., 2026). La mayoría de las acciones tienen una duración inferior a 10 segundos y requieren una rápida activación y coordinación del sistema neuromuscular, ya que la ejecución eficaz de gestos explosivos como el salto, el remate o el bloqueo exige producir elevados niveles de fuerza y potencia en intervalos temporales muy breves (Hedrick, 2007). Esto hace que el voleibol sea una disciplina en la que predominantemente se emplea el metabolismo anaeróbico, donde la resíntesis de Adenosín trifosfato (ATP) durante las acciones de juego se produce principalmente mediante vías metabólicas no dependientes directamente de la disponibilidad de oxígeno (Gastin, 2001) como la vía de los fosfágenos (ATP-PC) y la glucólisis. En este sentido, el sistema ATP-PC constituye la principal fuente de energía durante los esfuerzos breves y explosivos característicos del juego, mientras que la contribución de la glucólisis es más limitada (Figura 1). Este predominio del sistema ATP-PC se explica tanto por la corta duración de las acciones como por el carácter intermitente del juego, ya que los breves periodos de recuperación entre puntos (~5–8 segundos) permiten una participación del metabolismo oxidativo suficiente para favorecer la resíntesis parcial de fosfocreatina (Lidor & Ziv, 2010; Tomlin & Wenger, 2001).

Más allá de las demandas metabólicas, durante el transcurso de un partido, los jugadores y jugadoras de voleibol realizan múltiples acciones de alta intensidad, como saltos, aceleraciones, desaceleraciones, bloqueos y remates. Estas acciones implican una elevada demanda neuromuscular repetida de forma continua a lo largo del encuentro, cuya duración puede oscilar entre 60 y 90 minutos (Rebelo et al., 2026). En este contexto, los jugadores deben ser capaces de mantener elevados niveles de producción de fuerza, potencia y resistencia muscular en cada salto, golpeo o desplazamiento (Sattler et al., 2015; Sheppard et al., 2007). Por ello, el éxito en el voleibol depende de que variables clave como el salto vertical, la velocidad (p.ej. de golpeo, reacción y desplazamiento) y la agilidad no solo sean óptimas, sino también sostenibles en el tiempo bajo condiciones de fatiga acumulada propias de la alta competición (Miguel-Ortega et al., 2023; Nasuka, 2020; Sheppard et al., 2007).

Figura 1

Contribución de los sistemas energéticos al suministro de ATP durante el ejercicio



Nota. Adaptada de *Bioenergética de las fibras musculares y ejercicio*, por J. López Chicharro & A. Fernández Vaquero, 2017, Editorial Esteban Sanz.

Desde una perspectiva integradora, el rendimiento neuromuscular no depende únicamente de las características antropométricas o metabólicas del deportista, sino que emerge de la interacción con factores extrínsecos e intrínsecos de alta relevancia. Entre estos factores destacan una correcta higiene de sueño (Charest & Grandner, 2020), la nutrición y el estado de hidratación (Close et al., 2016; Larrosa et al., 2025), y el estrés psicosocial (Gabbett & Georgieff, 2007). Sin embargo, la cronobiología, disciplina de la biología que estudia los ritmos biológicos del organismo (Kuhlman et al., 2018) puede influir tanto en el rendimiento neuromuscular como en la aparición de la fatiga en el voleibol (Gabbett & Georgieff, 2007; Palao et al., 2014; Sheppard et al., 2008). En este contexto, resulta especialmente relevante considerar la influencia de la cronobiología y, en particular, de los ritmos circadianos, ya que son numerosos los procesos fisiológicos implicados en el rendimiento neuromuscular que presentan variaciones a lo largo del día en consonancia con estos ritmos circadianos. Estas fluctuaciones pueden modificar la capacidad de activación neuromuscular del deportista, así como la percepción del esfuerzo y los procesos de recuperación (Chtourou et al., 2012; Waterhouse et al., 2005), lo que justifica la necesidad de profundizar en sus bases fisiológicas y en su interacción específica con el rendimiento neuromuscular. Por tanto, comprender cómo los ritmos circadianos modulan la respuesta fisiológica y el rendimiento neuromuscular resulta clave para optimizar la planificación del entrenamiento y la preparación competitiva en deportes de equipo como el voleibol.

1.3 Cronobiología y sistema circadiano

La cronobiología es la disciplina científica que estudia los ritmos biológicos y su influencia sobre la fisiología y el comportamiento humano (Czeisler & Buxton, 2017). Dentro de estos ritmos biológicos, los ritmos circadianos son oscilaciones biológicas que se repiten en ciclos de aproximadamente 24 horas, y regulan funciones como el sueño, la temperatura corporal y la secreción hormonal (Patke et al., 2020). Estos ritmos están coordinados por el sistema circadiano, compuesto por un reloj central ubicado en el núcleo supraquiasmático (NSQ) del hipotálamo y por relojes periféricos distribuidos en diversos órganos, que sincronizan la actividad fisiológica con los ciclos ambientales de luz y oscuridad (Panda, 2016). Además, la expresión temporal de estos ritmos presenta una marcada variabilidad interindividual, lo que da lugar al denominado fenotipo circadiano (cronotipo), entendido como el conjunto de características biológicas que definen el funcionamiento del reloj interno de un individuo (p. ej., ritmos de sueño-vigilia, temperatura corporal o secreción hormonal). Dentro de este marco, el cronotipo se considera una de las principales manifestaciones conductuales, reflejando cómo esta organización biológica interna se traduce en preferencias y patrones de actividad a lo largo del día (Facer-Childs et al., 2018). Para comprender estas diferencias individuales, resulta necesario analizar, en primer lugar, los mecanismos fisiológicos que regulan el sistema circadiano humano, así como las formas en que estas variaciones se manifiestan y pueden ser evaluadas en contextos de investigación aplicada. En este sentido, la comprensión de estos aspectos resulta fundamental en el ámbito deportivo, ya que permite optimizar entrenamientos, programar competiciones y mejorar la recuperación, ajustando las cargas de trabajo al momento del día en que el organismo se encuentra más eficiente (Vitale & Weydahl, 2017).

1.3.1 Fundamentos fisiológicos y moleculares del ritmo circadiano humano

Los ritmos circadianos (del latín *circa*, “alrededor de”, y *diem*, “día”) son variaciones biológicas periódicas de origen endógeno, con una duración cercana a las 24 horas, que modulan diversas funciones fisiológicas entre las que podemos mencionar el ciclo sueño-vigilia, la temperatura corporal, la secreción hormonal, el metabolismo y el rendimiento neuromuscular (Roenneberg & Merrow, 2016). Estos ritmos se originan mediante mecanismos moleculares autorregulados basados en bucles de retroalimentación transcripcional-traducciona de los denominados genes reloj. Entre ellos destacan CLOCK y BMAL1, que inducen la expresión de los genes PER y CRY, responsables de

generar oscilaciones rítmicas en la expresión génica a lo largo del día (Takahashi, 2017). La regulación de los ritmos circadianos presenta una organización jerárquica encabezada por un reloj biológico central localizado en el NSQ, considerado el marcapasos maestro del organismo (Atkinson & Reilly, 1996). Este reloj central coordina una extensa red de relojes periféricos distribuidos en la mayoría de los tejidos y órganos, incluyendo sistemas fisiológicos como el cardiovascular, el respiratorio o el músculo esquelético (Su & Xiang, 2025; Takahashi, 2017). La interacción entre el reloj central y los relojes periféricos garantiza la coherencia temporal entre múltiples sistemas fisiológicos mediante señales neuro-endocrinas (Mohawk et al., 2012).

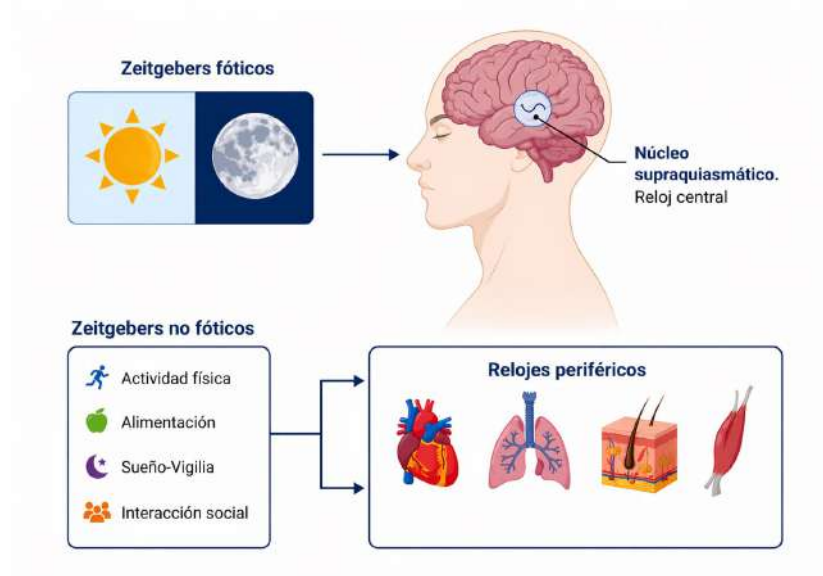
Aunque los ritmos circadianos se generan de forma endógena, su sincronización con el entorno depende de señales externas denominadas *zeitgebers* (sincronizadores). Entre estos, la luz constituye el principal sincronizador fisiológico en humanos, ya que la información luminosa captada por la retina se transmite al NSQ a través del tracto retinohipotalámico, permitiendo el ajuste diario del reloj biológico interno al ciclo ambiental de aproximadamente 24 horas (Figura 2) (Duffy & Czeisler, 2009; Roenneberg & Merrow, 2016). En ausencia de estos sincronizadores ambientales, el reloj interno tiende a expresarse con un periodo ligeramente superior a 24 horas, lo que puede generar un desfase progresivo respecto al entorno (Vitaterna et al., 2001; West & Bechtold, 2015). Además de la luz, existen otros *zeitgebers* secundarios capaces de modular la organización temporal del sistema circadiano, entre los que se incluyen la actividad física, la interacción social o la regularidad de los ciclos sueño-vigilia (Elmore et al., 1994; Mistlberger & Skene, 2004; Youngstedt et al., 2019). En particular, el ejercicio físico ha demostrado actuar como un potente sincronizador circadiano, capaz de inducir avances o retrasos de fase circadiana en función del momento del día, así como de la intensidad y duración del estímulo aplicado (Bennett & Sato, 2023; Youngstedt et al., 2019). A través de estos mecanismos, el ejercicio modula los ritmos hormonales, térmicos y de activación neuromuscular, adquiriendo un papel relevante en la regulación temporal del rendimiento neuromuscular (Lewis et al., 2018; Shen et al., 2023).

En conjunto, esta organización jerárquica y su interacción con los sincronizadores ambientales permiten al organismo anticiparse a las variaciones diarias del entorno, optimizar múltiples procesos fisiológicos y contribuir al mantenimiento de la homeostasis (Augsburger et al., 2022; Panda, 2016; Racinais et al., 2005). No obstante, la expresión funcional de estos ritmos no es idéntica en todos los individuos, lo que da lugar a

diferencias interindividuales en la organización temporal de los procesos fisiológicos y conductuales, fenómeno que se describe mediante el concepto de fenotipo circadiano.

Figura 2

Organización y sincronización del sistema circadiano humano



Nota. Figura de elaboración propia

1.3.2 Cronotipo como expresión del fenotipo circadiano: bases biológicas

La expresión temporal del sistema circadiano presenta una marcada variabilidad interindividual, lo que da lugar al denominado fenotipo circadiano (Facer-Childs & Brandstaetter, 2015). Este concepto abarca la manifestación de los ritmos biológicos tanto a nivel fisiológico (fase de la melatonina, nadir de la temperatura) como conductual (patrones de sueño y alerta) (Allebrandt & Roenneberg, 2008).

Dentro de este marco, el cronotipo constituye una de las principales expresiones del fenotipo circadiano y se define como la predisposición biológica individual hacia la actividad y el rendimiento en horarios matutinos o vespertinos (Mulè et al., 2020). La distribución del cronotipo se describe a lo largo de un continuo de matutinidad–vespertinidad, cuyos extremos corresponden a los tipos matutinos (*morning types*) y vespertinos (*evening types*), mientras que la mayoría de la población se concentra en un rango intermedio (Horne & Ostberg, 1976; Rae et al., 2015). Los individuos con cronotipo matutino tienden a despertarse y acostarse más temprano, presentan mayores niveles de alerta durante la mañana y muestran una fase circadiana adelantada, lo que se refleja en picos más tempranos de temperatura corporal y en la secreción de hormonas circadianas

como la melatonina y el cortisol, siendo su rendimiento neuromuscular y cognitivo más elevado durante las primeras horas del día (Kang et al., 2025). Por el contrario, los sujetos con cronotipo vespertino se caracterizan por horarios de sueño más tardíos y por un retraso de la fase circadiana, lo que conlleva un desplazamiento de sus picos de temperatura corporal, secreción hormonal, alerta y rendimiento neuromuscular hacia la tarde o la noche (Adan, 2013; Vitale et al., 2017). En este sentido, al igual que ocurre con los individuos matutinos en las primeras horas del día, las personas con cronotipo vespertino tienden a mostrar su máximo rendimiento neuromuscular y cognitivo en franjas horarias más tardías. Por su parte, los individuos con cronotipo intermedio presentan características intermedias entre ambos extremos, con horarios de sueño y vigilia más flexibles y una menor amplitud en las variaciones diurnas del rendimiento, lo que se traduce en una distribución más homogénea de sus niveles de alerta y desempeño físico y cognitivo a lo largo del día (Vitale & Weydahl, 2017).

Desde una perspectiva aplicada al rendimiento neuromuscular, esta diferenciación es especialmente relevante, ya que el cronotipo puede determinar el momento del día en el que un deportista alcanza su mayor eficacia cognitiva y su rendimiento neuromuscular óptimo (Vitale & Weydahl, 2017). Considerar esta variable resulta fundamental para interpretar adecuadamente los efectos de la hora del día sobre el rendimiento (Facer-Childs, & Brandstaetter, 2015; Roveda et al., 2020). En este sentido, en el contexto del alto rendimiento neuromuscular, donde las diferencias entre atletas son mínimas, pequeñas variaciones en el rendimiento pueden resultar determinantes para el éxito competitivo. De hecho, se ha observado que mejoras del orden del ~1% pueden ser suficientes para marcar la diferencia entre obtener una medalla o quedar fuera de las primeras posiciones en competiciones de élite como unos Juegos Olímpicos (Malcata & Hopkins, 2014; Pyne et al., 2004). Por tanto, comprender y optimizar factores como el cronotipo y la sincronización circadiana puede suponer una ventaja competitiva clave en contextos donde los márgenes de rendimiento son extremadamente reducidos.

Desde una perspectiva biológica, el cronotipo posee un marcado carácter genético, aunque su expresión final está estrechamente influenciada por factores fenotípicos y ambientales. A nivel molecular, esta base biológica está determinada por la actividad de los genes del reloj circadiano (p. ej., CLOCK, BMAL1, PER y CRY), responsables de regular funciones fisiológicas como la secreción de melatonina y cortisol o la temperatura corporal (Ko & Takahashi, 2006). Son precisamente estas fluctuaciones biológicas las

que sustentan las diferencias conductuales y de preferencia horaria observadas entre los distintos cronotipos (Hofstra & de Weerd, 2008).

Además de la base genética, existen diferentes factores moduladores que influyen en la expresión del cronotipo entre los que podemos mencionar: (i) la edad, que constituye uno de los determinantes más relevantes, observándose una tendencia hacia cronotipos más vespertinos durante la adolescencia y un desplazamiento progresivo hacia cronotipos más matutinos con el envejecimiento (Roenneberg et al., 2004), (ii) el sexo biológico, que puede influir en la distribución y expresión del cronotipo, (iii) factores ambientales y sociales, como la exposición a la luz artificial (Bonmati-Carrion et al., 2014), los horarios escolares o laborales (Wittmann et al., 2006) y la práctica sistemática de ejercicio físico, que puede actuar como un sincronizador circadiano (*zeitgeber*) capaz de inducir desplazamientos de fase en los ritmos circadianos en función del momento del día en que se realiza (Youngstedt et al., 2019). En consecuencia, estos factores pueden modificar la expresión conductual del cronotipo, favoreciendo procesos de adaptación o, en algunos casos, estados de desalineación circadiana persistente. La identificación y caracterización de estas diferencias individuales requiere el uso de herramientas específicas que permitan evaluar la organización temporal del sistema circadiano y estimar el cronotipo en contextos de investigación aplicada.

1.3.3 Métodos de evaluación del cronotipo y de los ritmos circadianos

Para estudiar la organización temporal del sistema circadiano y las diferencias interindividuales asociadas al cronotipo se han desarrollado diversos métodos de evaluación que incluyen marcadores genéticos y fisiológicos, así como cuestionarios conductuales validados (Hofstra & de Weerd, 2008). En relación con los biomarcadores genéticos, se ha observado que determinadas variantes en genes del reloj circadiano se asocian con la predisposición hacia distintos cronotipos. Polimorfismos en el gen PER3 (como el VNTR de repetición 4/5) se han relacionado con una mayor propensión hacia cronotipos matutinos en individuos portadores del alelo largo (Archer et al., 2003), mientras que variantes en CLOCK (p. ej., 3111T/C) y CRY1 se han vinculado con una tendencia hacia cronotipos más vespertinos (Allebrandt & Roenneberg, 2008; Patke et al., 2017). Estos hallazgos ponen de manifiesto la base genética subyacente a la variabilidad circadiana observada entre individuos. Entre los biomarcadores más utilizados destacan la melatonina, el cortisol y la temperatura corporal. La melatonina,

sintetizada por la glándula pineal, constituye uno de los marcadores endocrinos más utilizados para determinar la fase circadiana central, ya que su secreción nocturna se asocia con la denominada “noche biológica” (Shen et al., 2023; Thomas et al., 2020). Este estado representa el periodo de máxima predisposición al sueño, en el cual el sistema circadiano coordina el descenso de la temperatura corporal y la activación de procesos de recuperación, independientemente de los ciclos de luz externos. Por su parte, el cortisol presenta un patrón diurno aproximadamente inverso, caracterizado por un incremento pronunciado tras el despertar que favorece la activación fisiológica matutina (Hayes et al., 2010). En contextos aplicados al deporte, la temperatura corporal ha sido ampliamente utilizada como indicador circadiano debido a su patrón rítmico robusto, con valores mínimos durante la madrugada y un aumento progresivo a lo largo del día hasta alcanzar un pico generalmente localizado en la tarde (Reilly et al., 2007; Waterhouse et al., 2005). Este incremento térmico se ha relacionado con una mayor activación neuromuscular y con mejoras en variables como la producción de fuerza, potencia muscular o velocidad de reacción (Chtourou et al., 2011; Knaier et al., 2022). En deportes de carácter explosivo, como el voleibol, el pico vespertino de la temperatura corporal se ha relacionado de manera consistente con valores superiores de salto vertical, velocidad de reacción y producción de potencia, lo que la convierte en un marcador idóneo para el estudio de las variaciones diurnas del rendimiento neuromuscular (Hesketh & Esser, 2024; Reilly et al., 2007; Souissi et al., 2007).

No obstante, la evaluación directa de estos biomarcadores requiere condiciones experimentales controladas y procedimientos analíticos específicos, lo que limita su aplicación en estudios de campo y en contextos deportivos aplicados (Hofstra & de Weerd, 2008). Por ello, en el ámbito deportivo se recurre con frecuencia a cuestionarios conductuales validados que permiten estimar el cronotipo de forma fiable y no invasiva. Entre estos instrumentos, el Morningness–Eveningness Questionnaire (MEQ) (Horne & Ostberg, 1976) constituye el instrumento más ampliamente utilizado y validado tanto en población general como deportiva. El MEQ consta de 19 ítems que exploran hábitos de sueño, preferencias horarias y niveles de alerta, y permite clasificar a los individuos como matutinos (59-86), intermedios (42-58) o vespertinos (16-41) en función de su puntuación total (Figura 4) (Smith et al., 1989). Por el contrario, el Munich Chronotype Questionnaire (MCTQ) adopta una aproximación conductual y cuantitativa, ya que estima el cronotipo a partir de los horarios reales de sueño en días laborales y libres, calculando indicadores

como el punto medio del sueño (*mid-sleep*) y el *jet lag* social (Roenneberg et al., 2003). De este modo, el MEQ refleja la preferencia circadiana percibida, mientras que el MCTQ proporciona una medida más ecológica del comportamiento real de sueño.

Figura 3

Clasificación del cronotipo (matutino, intermedio y vespertino) según la puntuación obtenida en el MEQ (Horne & Ostberg, 1976)



Nota. Figura de elaboración propia

1.4 Variación diurna del rendimiento neuromuscular: influencia de factores circadianos e interindividuales

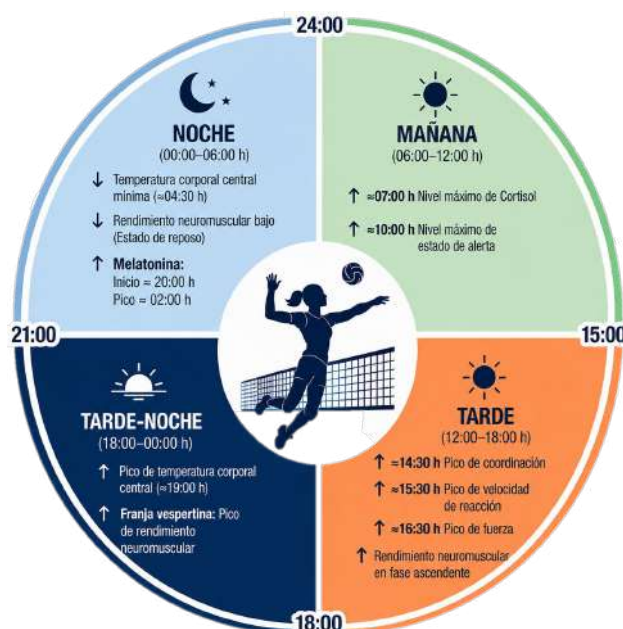
Diversos estudios han demostrado que la organización temporal de los ritmos biológicos influye significativamente en el rendimiento neuromuscular (Vitale & Weydahl, 2017). En particular, la cronobiología aplicada al deporte ha evidenciado que tanto la hora del día como las diferencias interindividuales en la expresión del sistema circadiano pueden modular el rendimiento neuromuscular (Ayala et al., 2021; Vitale & Weydahl, 2017). Esta influencia puede resultar especialmente relevante en deportes de equipo caracterizados por acciones intermitentes, como el voleibol, donde el éxito a nivel competitivo depende de la integración precisa de fuerza, potencia, velocidad de reacción y coordinación motora, y en el que pequeñas variaciones en estas capacidades pueden tener un impacto decisivo en el rendimiento neuromuscular (Mirizio et al., 2020). Por tanto, la variación diurna del rendimiento neuromuscular se ve influenciada no solo por el momento del día, sino también por factores individuales que configuran el fenotipo circadiano del deportista, entre los que destacan el cronotipo y el sexo biológico (Pradhan et al., 2024).

1.4.1 Variación diurna del rendimiento neuromuscular

La evidencia científica acumulada durante las últimas décadas ha demostrado de manera consistente que el rendimiento neuromuscular no permanece constante a lo largo del día (Ayala et al., 2021; Vitale & Weydahl, 2017). En términos generales, la mayoría de las capacidades neuromusculares muestran valores inferiores durante el horario matutino alcanzando sus valores máximos en el horario vespertino o primeras horas de la noche (entre 16:00-20:00 h) (Drust et al., 2005; Kons et al., 2025; Nunes et al., 2021; Reilly et al., 2007) (Figura 4).

Figura 4

Variación circadiana del rendimiento neuromuscular y variables fisiológicas



Nota. Adaptada de *Interactions of cortisol, testosterone, and resistance training: Influence of circadian rhythms*, por L. D. Hayes, et al., 2010, *Chronobiology International*, 27(4), p. 675-705.

Este patrón ha sido descrito de forma especialmente consistente en manifestaciones del rendimiento neuromuscular, como la fuerza máxima, la potencia muscular, la velocidad y la aceleración, tanto en el miembro inferior como en el superior, coincidiendo con mayores niveles de activación neuromuscular (Chtourou & Souissi, 2012; Drust et al., 2005; Racinais et al., 2005). Desde una perspectiva fisiológica, estas variaciones diurnas se han relacionado principalmente con oscilaciones circadianas en la temperatura corporal central y en el nivel de activación del sistema nervioso central. En particular, el incremento progresivo de la temperatura corporal a lo largo del día, con valores máximos en la tarde, se asocia con modificaciones en las propiedades viscoelásticas del músculo y

el tendón, así como con una mayor velocidad de conducción nerviosa (Racinais & Oksa, 2010). A nivel muscular, estas adaptaciones se traducen en un aumento de la capacidad de generación de fuerza/potencia muscular (Coletta et al., 2018), la extensibilidad musculotendinosa y la eficiencia mecánica (Waterhouse et al., 2005), favorecido por una mayor eficiencia en la cinética de los puentes cruzados de actina-miosina y en la actividad de las enzimas implicadas en la hidrólisis del ATP (Decostre et al., 2005). En conjunto, este entorno fisiológico vespertino potencia la extensibilidad muscular y la transmisión de fuerzas, optimizando el rendimiento neuromuscular en acciones explosivas (Racinais & Oksa, 2010).

1.4.2 Fenotipo circadiano como modulador individual del momento óptimo de rendimiento

El cronotipo constituye un elemento a tener en cuenta para la identificación del momento óptimo de rendimiento, ya que refleja la fase circadiana preferente de cada deportista y, con ello, nos proporciona información relevante de sus picos de alerta, activación fisiológica y capacidad neuromuscular (Facer-Childs et al., 2018). Desde esta perspectiva, el rendimiento neuromuscular no depende únicamente de la hora del día, sino del grado de alineación entre el momento de la actividad y la fase circadiana del individuo, lo que constituye una variable explicativa central de la variabilidad interindividual (Rae et al., 2015; Vitale & Weydahl, 2017).

Diversos estudios han sugerido que el rendimiento neuromuscular puede alcanzar sus valores máximos cuando la actividad se alinea con el cronotipo del sujeto (Facer-Childs & Brandstaetter, 2015; Vitale & Weydahl, 2017). A nivel práctico, parece ser que los deportistas con cronotipo matutino suelen rendir mejor en franjas tempranas, aproximadamente entre las 07:00-10:00 h, mientras que los de cronotipo vespertino parecen alcanzar su pico de rendimiento neuromuscular en horarios más tardíos, especialmente entre las 16:00-20:00 h (Facer-Childs, & Brandstaetter, 2015; Vitale & Weydahl, 2017). Por el contrario, los sujetos con cronotipo intermedio se caracterizan por presentar una menor variabilidad diurna en su rendimiento neuromuscular. A diferencia de los perfiles extremos, cuya capacidad funcional fluctúa drásticamente según la hora del día, los tipos intermedios muestran una mayor estabilidad en sus niveles de fuerza y potencia muscular, permitiéndoles mantener un rendimiento más homogéneo a través de

una ventana horaria más amplia (Facer-Childs, & Brandstaetter, 2015). Por el contrario, la realización de esfuerzos en fases biológicamente desfavorables, es decir aquellos con un cronotipo matutino a los que se les exige rendir por la tarde o viceversa, o aquellos con un cronotipo vespertino y se les exige rendir por la mañana suele asociarse con una reducción del rendimiento neuromuscular y una mayor percepción de esfuerzo, especialmente en tareas dependientes de la activación neuromuscular (Facer-Childs, & Brandstaetter, 2015), no obstante, cabe destacar que estas diferencias tienden a reducirse cuando el atleta se habitúa a entrenar en el horario en el que se llevará a cabo la competición, independiente del cronotipo (Chtourou et al., 2012).

Además, es importante señalar que la mayor parte de la evidencia disponible sobre la influencia del cronotipo en el rendimiento neuromuscular procede de estudios realizados en contextos relativamente controlados, principalmente en entornos de laboratorio, y centrados en pruebas físicas estandarizadas (Forster et al., 2025; Tamm et al., 2009). En estos contextos, es más fácil analizar el efecto de los ritmos biológicos, al no estar presentes otros factores propios del deporte de equipo como la táctica, la planificación del entrenamiento o la interacción entre jugadores (Facer-Childs et al., 2018). En disciplinas como el ciclismo, la carrera (Vitale & Weydahl, 2017) o las pruebas de fuerza (Küüsmaa et al., 2016), se ha observado que la alineación entre el cronotipo y el horario de esfuerzo explica una parte relevante de la variabilidad interindividual. Específicamente, se ha cuantificado que el rendimiento neuromuscular puede oscilar entre un 7-26% a lo largo del día en función de la sincronización circadiana (Facer-Childs et al., 2018; Forster et al., 2025). Incluso cuando se controla la hora absoluta del día, los deportistas que realizan el esfuerzo en su ventana biológica óptima muestran una ventaja de rendimiento de hasta un 10% en comparación con aquellos que compiten en desalineación, especialmente en tareas de alta intensidad neuromuscular (Facer-Childs, & Brandstaetter, 2015; Vitale et al., 2017). Estos hallazgos han reforzado la noción de que existe un momento óptimo de rendimiento específico para cada deportista, sin embargo, en deportes de equipo, la evidencia es más limitada y los resultados menos consistentes debido a la complejidad de las demandas físicas, técnicas y tácticas, así como a la rigidez de los horarios de entrenamiento y competición. En este sentido, la exposición sistemática a entrenamientos en una franja horaria fija puede inducir adaptaciones en la fase del ritmo circadiano, un fenómeno de plasticidad fenotípica que permite al deportista optimizar su rendimiento neuromuscular en ese horario específico (Rae et al., 2015). Estas

adaptaciones actúan sobre el fenotipo circadiano funcional logrando atenuar las desventajas de rendimiento neuromuscular asociadas al genotipo circadiano del individuo (p. ej., polimorfismos en genes como CLOCK o PER3). No obstante, aunque el entrenamiento actúa como un potente sincronizador (*zeitgeber*) capaz de desplazar los picos de activación, no logra eliminar por completo la predisposición genética subyacente, persistiendo diferencias significativas entre cronotipos extremos (Ciorciari et al., 2023; Rae et al., 2015).

En definitiva, la desalineación entre el cronotipo y el horario de actividad no constituye un fenómeno inocuo, sino que altera la homeostasis fisiológica del deportista. Esta falta de sincronía reduce la eficiencia neuromuscular al comprometer la tasa de desarrollo de la fuerza (RFD) y la velocidad de conducción nerviosa (Maffiuletti et al., 2016). Asimismo, provoca una alteración del balance hormonal anabólico al modificar el ratio testosterona/cortisol (Hayes et al., 2010) e incrementa la fatiga cognitiva, incrementando los tiempos de reacción y la latencia de respuesta (Tamm et al., 2009). En consecuencia, forzar el rendimiento neuromuscular en fases biológicamente desfavorables aumenta la percepción subjetiva del esfuerzo y el riesgo de fatiga acumulada, limitando la capacidad de adaptación al estímulo deportivo (Vitale & Weydahl, 2017).

1.4.3 Influencia del dimorfismo sexual en la regulación circadiana y el rendimiento neuromuscular

El sexo biológico constituye un modulador relevante en la regulación del sistema circadiano y en su expresión funcional sobre el rendimiento neuromuscular (Mong et al., 2011). A nivel cronobiológico, se ha observado una tendencia intrínseca hacia la matutinidad en las mujeres en comparación con los hombres. Esta diferencia tiene su base fisiológica en un periodo circadiano intrínseco ligeramente más breve; mientras que el ciclo promedio masculino se aproxima a las 24,19 horas, el femenino es significativamente más corto, situándose en torno a las 24,09 horas (Duffy et al., 2011). Esta diferencia biológica se asocia con que las mujeres completen su ciclo biológico en menos tiempo, lo que favorece una fase circadiana adelantada y una activación fisiológica más temprana (Duffy et al., 2011; Walton et al., 2022).

Además de las diferencias en la fase circadiana, el sexo biológico modula la amplitud de los ritmos fisiológicos, es decir, la magnitud de la oscilación entre los valores máximos

y mínimos de una variable. En este sentido, las mujeres suelen presentar una mayor amplitud en variables térmicas y endocrinas, como la temperatura central y la secreción de melatonina, lo que sugiere una organización circadiana que busca la estabilidad de las funciones homeostáticas (Walton et al., 2022). Esta regulación no es meramente genética, sino que depende de un perfil hormonal diferencial entre sexos, donde los estrógenos y la progesterona interactúan directamente con el núcleo supraquiasmático y los relojes periféricos. Los estrógenos, en particular, actúan como un factor modulador que afina la sensibilidad a los sincronizadores externos y optimiza la expresión de los picos de activación neuromuscular (Joyner et al., 2025; Walton et al., 2022).

Desde una perspectiva morfológica, las diferencias fisiológicas entre hombres y mujeres podrían contribuir a explicar patrones diferenciados de variación diurna del rendimiento neuromuscular. Los hombres poseen, por lo general, una mayor sección transversal y una proporción superior de fibras tipo II (rápidas), altamente dependientes de la temperatura corporal central y la activación neural para su funcionamiento óptimo (Haizlip et al., 2015). Dado que estas variables fluctúan a lo largo del día, los hombres muestran una mayor amplitud en la variación diurna (picos más pronunciados por la tarde), especialmente en tareas explosivas y de alta intensidad (Augsburger et al., 2022). Por el contrario, las mujeres tienden a mostrar una mayor densidad de fibras tipo I (lentas) y una eficiencia metabólica oxidativa superior (Hunter & Senefeld, 2024). Estas características metabólicas y estructurales podrían favorecer una mayor estabilidad neuromuscular, provocando oscilaciones diurnas menos acentuadas y un rendimiento neuromuscular más homogéneo a lo largo de la jornada.

En el ámbito del rendimiento, estas diferencias sugieren que el sexo biológico no solo condiciona el rendimiento neuromuscular, sino también el patrón temporal y la magnitud de su variación diurna (Vitale & Weydahl, 2017), no obstante, la evidencia empírica disponible sigue siendo limitada y, en muchos casos, inconsistente. Esta falta de consenso se explica, en parte, por el reducido número de estudios que han analizado específicamente las diferencias entre sexos en la variación circadiana del rendimiento neuromuscular, así como por la heterogeneidad de los protocolos experimentales empleados, los tamaños muestrales limitados y la escasa consideración de variables que pueden tener una influencia determinante, como el cronotipo de los participantes (Costello et al., 2014; Guo et al., 2022). En consecuencia, el análisis del sexo biológico como componente del fenotipo circadiano funcional resulta imprescindible para avanzar

hacia una comprensión más completa y aplicada del rendimiento neuromuscular en deportes de equipo como el voleibol.

En síntesis, a pesar de la sólida fundamentación teórica existente en el ámbito de la cronobiología humana, la evidencia empírica aplicada a los deportes de equipo y, de manera específica, al voleibol resulta limitada. En este contexto, la presente Tesis Doctoral se planteó como objetivo principal contribuir al vacío en la literatura existente.

OBJETIVOS E HIPÓTESIS

2. OBJETIVOS E HIPÓTESIS

La presente Tesis Doctoral plantea la siguiente hipótesis:

El rendimiento neuromuscular en jugadores de voleibol variará en función del momento del día, mostrando valores superiores en horario vespertino respecto al matutino. Esta variación estará modulada por el cronotipo, de modo que una mayor alineación entre el momento del día y las preferencias circadianas del individuo se asociará con un mejor rendimiento neuromuscular. Además, el efecto del momento del día sobre el rendimiento neuromuscular diferirá según el sexo.

Esta hipótesis fue contrastada mediante el planteamiento de cuatro estudios, cada uno con sus respectivos objetivos e hipótesis.

Estudio 1

Objetivo 1: Examinar el efecto del momento del día sobre el rendimiento neuromuscular (p. ej., la capacidad de salto vertical o la agilidad) en deportistas de equipo de nivel profesional o semiprofesional.

Hipótesis 1: El rendimiento neuromuscular en deportistas de deportes de equipo de nivel profesional y semiprofesional será menor en horario matutino en comparación con el horario vespertino.

Estudio 2

Objetivo 2: Analizar el efecto del momento del día y del cronotipo sobre el rendimiento neuromuscular en jugadoras de voleibol semiprofesionales.

Hipótesis 2: El rendimiento neuromuscular será mayor en horario vespertino en comparación con el matutino, y que esta mejora estará vinculada al cronotipo individual de las jugadoras de voleibol semiprofesionales.

Estudio 3

Objetivo 3: Determinar la influencia del fenotipo circadiano (cronotipo) sobre el rendimiento neuromuscular en jugadores de voleibol masculino.

Hipótesis 3: El rendimiento neuromuscular de los jugadores de voleibol será superior cuando la evaluación se realice en un momento del día alineado con su fenotipo circadiano, mostrando los deportistas de cronotipo matutino un mejor rendimiento en horario matutino y los de cronotipo vespertino en horario vespertino.

Estudio 4

Objetivo 4: Establecer la influencia del sexo y su modulación sobre el efecto del momento del día sobre el rendimiento neuromuscular en jugadores y jugadoras de voleibol.

Hipótesis 4: El rendimiento neuromuscular de los jugadores de voleibol será mayor en horario vespertino en comparación con el matutino en ambos sexos. Asimismo, la magnitud de estas fluctuaciones diferirá entre hombres y mujeres, mostrando los hombres una mayor variabilidad diurna y manteniendo las mujeres un rendimiento neuromuscular más estable a lo largo del día.

**ESTUDIOS
DERIVADOS
DE LA TESIS
DOCTORAL**

3. ESTUDIOS DERIVADOS DE LA TESIS DOCTORAL

La presente Tesis Doctoral, de acuerdo con el informe correspondiente autorizado por los directores y en cumplimiento con la normativa aprobada por el Órgano Responsable del Programa de Doctorado de Biomedicina, Medicina y Ciencias Biosanitarias de la Escuela Internacional de Doctorado de la Universidad Francisco de Vitoria, se presenta siguiendo el modelo de tesis por compendio de artículos, conformado por tres estudios publicados y un cuarto estudio en revisión.

Las referencias completas de los estudios que constituyen el cuerpo de la presente Tesis Doctoral se presentan a continuación:

- I. **Martín-López J.**, Pérez-López, A., Varillas-Delgado D., & López-Samanes, Á. (2025). Influence of time-of-day on neuromuscular performance in team sport athletes: a systematic review and meta-analysis. *Frontiers in Sports and Active Living*, 6:1466050. (Factor de impacto = 2.6, Q2 Sport Sciences (40/134)).
- II. **Martín-López, J.**, Sedliak, M., Valadés, D., Muñoz, A., Buffet-García, J., García-Oviedo, R., Rodríguez-Aragón, M., Pérez-López, A., & López-Samanes, Á. (2022). Impact of time-of-day and chronotype on neuromuscular performance in semi-professional female volleyball players. *Chronobiology International*, 39(7), 1006–1014. (Factor de impacto = 2.8, Q2 Biology (40/92)).
- III. **Martín-López, J.**, Muñoz, A., Montalvo-Alonso, J. J., Quintana-Milla, I., Sánchez-Jorge, S., Conejero, M., López-Samanes, Á., & Pérez-López, A. (2026). Circadian phenotype influence on neuromuscular performance in male volleyball players. *Chronobiology International*, 43 (3): 421-430. (Factor de impacto = 1.7, Q2 Biology (52/107)).
- IV. **Martín-López, J.**, Muñoz, A., Montalvo-Alonso, J. J., Quintana-Milla, I., Sánchez-Jorge, S., Rebelo, A., López-Samanes, Á., & Pérez-López, A. (2026). Sex differences in the effects of diurnal variation on neuromuscular performance in volleyball players. *International Journal of Sports Physiology and Performance* (*en revisión*).

Tabla 1*Características metodológicas y principales resultados de los estudios que conforman esta Tesis Doctoral*

Estudio	Muestra	Diseño experimental	Variables analizadas	Análisis estadístico	Resultados
1	10 estudios (163 deportistas de deportes de equipo)	Revisión sistemática y meta-análisis	Capacidad de salto (CMJ y SJ), fuerza isométrica y agilidad	Meta-análisis efectos aleatorios (Hedges g; I ²)	El rendimiento neuromuscular fue superior en horario vespertino en CMJ y agilidad, sin diferencias en SJ ni fuerza isométrica.
2	15 jugadoras semiprofesionales de voleibol (Superliga 2 española)	Diseño aleatorizado, cruzado y contrabalanceado (09:00 vs 19:00 h)	Velocidad de golpeo, movilidad articular, equilibrio dinámico, capacidad de salto (CMJ, SJ y <i>spike jump</i>), fuerza isométrica de mano y agilidad	T-test pareado, regresión lineal y tamaño del efecto (Cohen's d)	El rendimiento neuromuscular fue superior en horario vespertino en velocidad de golpeo, agilidad, movilidad articular y equilibrio dinámico, sin asociación con el cronotipo.
3	46 jugadores semiprofesionales de voleibol. Cronotipo matutino (n = 12), intermedio (n = 15) y vespertino (n = 19)	Diseño aleatorizado y cruzado según fenotipo (08:00 vs 19:00 h)	Velocidad de golpeo, equilibrio dinámico, capacidad de salto (CMJ, SJ y <i>spike jump</i>), fuerza isométrica de mano y agilidad	ANOVA de medidas repetidas (cronotipo × momento del día), post hoc Bonferroni y tamaños del efecto (η^2 y Cohen's d)	Influencia limitada del cronotipo sobre el rendimiento neuromuscular. Se observaron mejores valores vespertinos en salto y velocidad de golpeo.
4	102 jugadores/as de voleibol competitivo (46 hombres y 56 mujeres)	Diseño aleatorizado y cruzado con comparación por sexo (08:00-10:00 h vs 19:00-20:00 h)	Velocidad de golpeo, equilibrio dinámico, capacidad de salto (CMJ, SJ y <i>spike jump</i>), fuerza isométrica de mano y agilidad	ANOVA de medidas repetidas (sexo × momento del día), post hoc Bonferroni y tamaños del efecto (η^2 y Cohen's d)	El rendimiento neuromuscular fue superior en horario vespertino en salto vertical (SJ, CMJ y <i>spike jump</i>), velocidad de golpeo y fuerza isométrica de mano. Las diferencias por sexo fueron reducidas.

3.1 Estudio 1: Influence of time-of-day on neuromuscular performance in team sport athletes: a systematic review and meta-analysis

ESTUDIO 1

Publicado:

**Martin-López J., Pérez-López, A., Varillas-Delgado D., & López-Samanes, Á.
(2025) Influence of time-of-day on neuromuscular performance in team sport athletes: a systematic review and meta-analysis. Frontiers in Sports and Active Living, 6,:1466050.**

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Influence of time-of-day on neuromuscular performance in team sport athletes: a systematic review and meta-analysis

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Introduction: Although circadian rhythms have been shown to influence some neuromuscular performance tasks, the time-of-day effect on team sports performance athletes remains equivocal. This study aimed to examine the existing evidence concerning diurnal variations in neuromuscular performance in professional and semi-professional team sports athletes using a meta-analytic approach.

Methods: A literature search was conducted through three different databases: PubMed, SportDiscus and Web of Science. Article selection was made based on the following inclusion criteria: team sports athletes, professional or semi-professional athletes, neuromuscular performance, testing protocols and time-of-day testing times. Neuromuscular performance parameters such vertical jump capacity (i.e., squat and countermovement jump), agility and isometric strength were included in the analysis. Testing protocols that specifically assessed these parameters across morning (AM) and late afternoon/evening (PM) periods were considered were extracted from the selected studies.

Results: Ten studies met the inclusion criteria for qualitative synthesis and five for quantitative synthesis. Meta-analysis indicated lower countermovement jump in the AM compared to with PM (mean difference, -1.44 ; 95% CI -2.80 to -0.08 ; $p = 0.04$) and higher agility performance (mean difference 0.42 ; 95% CI 0.09 – 0.74 ; $p = 0.01$) in PM comparing with AM. No differences were reported in isometric strength and squat jump performance ($p > 0.05$).

Conclusion: Neuromuscular performance is higher in the late afternoon or early evening compared to morning schedules in team sport athletes. Hence, time-of-day variations need to be considered when evaluating neuromuscular performance in professional and semi-professional team sports athletes.

KEYWORDS

time-of-day, neuromuscular performance, team-sports, power, agility

1 Introduction

In team sport athletes, the coordination of muscle recruitment patterns and the control of movements through proprioceptive feedback reflect the efficacy of the neuromuscular system in initiating and directing specific sports actions that determine performance (1, 2). Several aspects could impact neuromuscular performance including nutritional

status (3), hormonal fluctuations (4), muscle fiber typology (5), or time-of-day when training/competitions are developed (6). Referring to the effect of time-of-day, it is worth mentioning the last years exponential increase in the number of scientific investigations that analyzed their impact on neuromuscular performance despite limited evidence in professional and semi-professional athletes can be found (7).

Numerous factors have been proposed to elucidate the potential time-of-day variations in neuromuscular performance such as external factors (i.e., ambient conditions), lifestyle conditions (e.g., time awake and times of training and habitual competition) or internal (endogenous) factors such as chronotype (i.e., manifestation of circadian rhythmicity and predispositions towards morning or evening orientation) or hormonal fluctuations (e.g., cortisol/melatonin) (8, 9). Regarding hormonal rhythms, particularly cortisol and melatonin, play a crucial role in influencing athletic performance by regulating energy levels, alertness, and recovery. Cortisol levels peak in the morning, enhancing physical readiness (10), while melatonin rises at night, facilitating recovery but potentially hindering late-evening performance (11). Athletes' chronotypes, whether they are naturally inclined toward early or late activity, interact with these hormonal cycles, with early chronotypes excelling in the morning and late chronotypes performing optimally in the evening (10, 11). Aligning training and competition schedules with an athlete's chronotype, alongside strategies such as sleep regulation and light exposure, can significantly enhance both performance and recovery, improving mood and cognitive function. In addition to physiological factors, circadian timing affects mood and cognitive function, which are essential for decision-making and situational awareness in team sports. Group-based exercise at specific times has been shown to improve mood and reduce depressive symptoms (12). Furthermore, morning exercise has been found to counteract cognitive impairments caused by partial sleep deprivation offering practical insights for optimizing athletic performance, especially under challenging or irregular schedules (13). However, the majority of the studies published on this topic have been developed on endurance (e.g., swimming or cycling) (14, 15) or power/strength disciplines (16, 17), existing limited evidence in specific tasks of team-sports athletes.

The identification of time-of-day fluctuations in neuromuscular performance among team-sports athletes may provide useful information for coaches and performance staff to schedule training and competitions. Nevertheless, a reduced number of studies have been developed in team-sports athletes and most of them are developed in young athletes (18, 19) or amateur/recreational team sports players (20, 21), with only a few focusing on professional and semi-professional players engaged in team sports athletes (22, 23). Since minimal differences in performance ($\approx 1\%$) might determine success in high-level athletes (24) the understanding of the potential time-of-day influence on neuromuscular performance would contribute to maximizing professional and semi-professional performance. Hence, the aim of this systematic review and meta-analysis was to examine the time-of-day effect in neuromuscular performance (e.g., vertical jump or agility capacity) on professional or semi-professional team sports athletes. Based on previous

findings we hypothesized that neuromuscular performance related to team sports disciplines will be less pronounced in the morning in comparison to late afternoon and early evening in professional and semi-professional team sports athletes.

2 Methods

The entire systematic review and meta-analysis process followed the guidelines outlined in the Preferred Reporting Items for Systematic Review and Meta-Analyses (PRISMA) (25). The review was registered in July 2022 and updated in November 2023 on PROSPERO (ID = CRD42022347351).

2.1 Search strategy, information sources and data managements

A detailed search of the literature was explored using the databases Medline (PubMed), SPORTDiscus, and Web of Science using all the available articles until November 2023. The search strategy was designed to explore areas related to circadian rhythms, chronotype and sports performance in professional and semi-professional sports teams players using a combination of the following keywords: ("professional player" OR "sports player" OR "high-performance player" OR "semi-professional player" OR "soccer players" OR "football players" OR "basketball players" OR "baseball players" OR "hockey players" OR "handball players" OR "rugby players" OR "volleyball players" OR "elite players" OR "athlete") AND ("time of day" OR "time-of-day" OR "morning" OR "evening" OR "morningness-eveningness", OR "circadian rhythms" OR "chronobiology" OR "chronotype") AND ("performance" OR "physical performance" OR "exercise performance" OR "sports performance" OR "neuromuscular performance" OR "body temperature").

2.2 Eligibility criteria

The PICOS model (26) was utilized to establish the inclusion criteria being (a) studies carried out in team sports athletes; (b) professional or semi-professional team sports athletes; (c) studies comparing the morning vs. later afternoon or early evening performance on team sports athletes; (d) studies with experimental designs, specifically randomized crossover trials, that assessed neuromuscular performance in controlled conditions. Exclusion criteria were: (a) studies involving individual sports players; (b) amateur/recreational athletes; (c) jetlag or sleep deprivation studies; (d) studies that applied any dietary supplement (e.g., melatonin) that may affect sports performance; (e) retrospective studies.

2.3 Data collection process and data items

Two researchers independently extracted data for each study that met all inclusion criteria, and a third author resolved any

discrepancies. The information extracted from each study included sample size, participants' characteristics, experimental design and procedure, neuromuscular performance measurements taken, and main morning vs. afternoon outcomes.

2.4 Statistical analysis

The collected data was presented as either the mean $\pm$ standard deviation (SD) or mean (SD). Meta-analysis was performed by comparing each dependent variable between morning (7:00–10:00 h) and afternoon/evening conditions (16:00–20:00 h) using Review Manager software (RevMan, version 5.4; The Cochrane Collaboration, Copenhagen, Denmark) to perform statistical analyses. The random effects model was chosen because of the small sample size and the expected heterogeneity among the chosen studies. The study heterogeneity was explored using the I^2 static and interpreted as “low” if $<50\%$, “moderate” if I^2 is between 50% and 75%, and “high” if I^2 is $>75\%$. The effect size of comparison for each dependent variable was determined by Hedges to reduce the potential bias associated with small sample size. For all analyses, the statistical significance threshold was set at $p < 0.05$.

3 Result

3.1 Search results

The databases search identified 975 potential full-text research articles. After eliminating duplicate articles and those that did not align with the exclusion and inclusion criteria, 22 articles remained and were selected for full-text screening. Another 12 studies were removed after full-text screening because the (a) abstract were not relevant (b) outcomes were not applicable. Finally, 10 studies were selected for the final review and 5 for the meta-analysis. The flow chart shown in Figure 1 provides the results of the literature search.

3.2 Characteristics of included studies and methodological quality

Table 1 outlines the experimental characteristics of 10 selected studies that included 163 healthy professional or semi-professional team sport athletes, aged between 17 and 28 years, being approximately 73% males and 27% females (22, 23, 27–34). All studies employed a similar randomized, crossover experimental design, in which the same participants underwent an experimental trial on at least two occasions, once in the morning (AM) and once in the evening (PM), with the exception of one retrospective study (Heishman et al., 2017) that analyzed previously collected data to evaluate performance. The testing periods for AM and PM trials ranged from 7.00–10.00 and 16.00–22.00 h, respectively. The exercise protocols implemented in these studies were designed to assess the influence of time-of-day on neuromuscular performance (Figure 1).

3.3 Qualitative systematic review

Nine used a longitudinal experimental randomized cross-over study (Cherif et al., 2022; Hammouda et al., 2013; Mhenni et al., 2017, 2021; Pavlovic et al., 2018; Pullinger et al., 2014; Martin-López et al., 2022; Ünver & Atan, 2021; West et al., 2014) and one was a retrospective analysis (Heishman et al., 2017). Different neuromuscular tests were performed at various times throughout the day in the 10 studies selected. In five studies the time-of-day effect was assessed in vertical jumping capacity (23, 27, 29, 32, 33). A sample of 79 team-sports athletes, comprising 15 females and 64 males, realized different vertical jumping tests (i.e., squat and countermovement jump) in two different time-of-day periods: morning (7:30–9:00) and evening (17:00–20:00). The results of a pooled analysis of our meta-analysis indicated a diurnal variation, with superior performance in vertical jumping capacity in the afternoon compared to morning values in countermovement jump (5%), but not in the squat jump (5%). Isometric handgrip strength was measured in three of the 10 articles selected (22, 29, 31). Participants (45 females) performed an isometric handgrip strength with the dominant limb test in a neutral position, and the pooled analysis of the studies selected reported no significant differences in the afternoon comparing with morning values (9%) (22, 29, 31). Regarding agility capacity was analyzed in four of the 10 studies (22, 23, 29, 31). Team sports athletes (45 female and 16 male) carried out different validated agility tests (i.e., modified agility *t*-test, zig-zig test). Consistent with earlier findings, the team sports athletes demonstrated enhanced performance in the afternoon tests compared to the morning values (7%). Among the studies reviewed, four specifically investigated the impact of time-of-day on movement speed in short distances (i.e., 0–30 m). Each of these studies examined how neuromuscular metrics varied between morning and afternoon sessions, offering insights into the optimal times for peak athletic performance (23, 30). Both studies were with 36 team sports athletes analyzing velocity in short distances (i.e., 5-m, 10-m, 20-m and all-out 30s sprints) to assess the speed of athlete movement and demonstrated consistent improvements in performance during late afternoon and early evening. Finally, the only study (15 males) that measured muscular endurance found no significant differences between the performance obtained in the morning and the results obtained in the late afternoon and early evening (34).

3.4 Assessment of risk of bias

The Cochrane Collaboration's tool for assessing risk of bias (RoB) was used to evaluate the study risk of bias within the included randomized controlled trials (35). The risk of selection bias, related to a lack of random sequence generation and allocation concealment, was low in two studies using random group allocation (22, 29). Three studies used random allocation, but the method was unclear (23, 32, 33). The methodology for blinding participants, personnel, and outcome assessors was not clearly reported in any of the studies (22, 23, 29, 32, 33), whereas

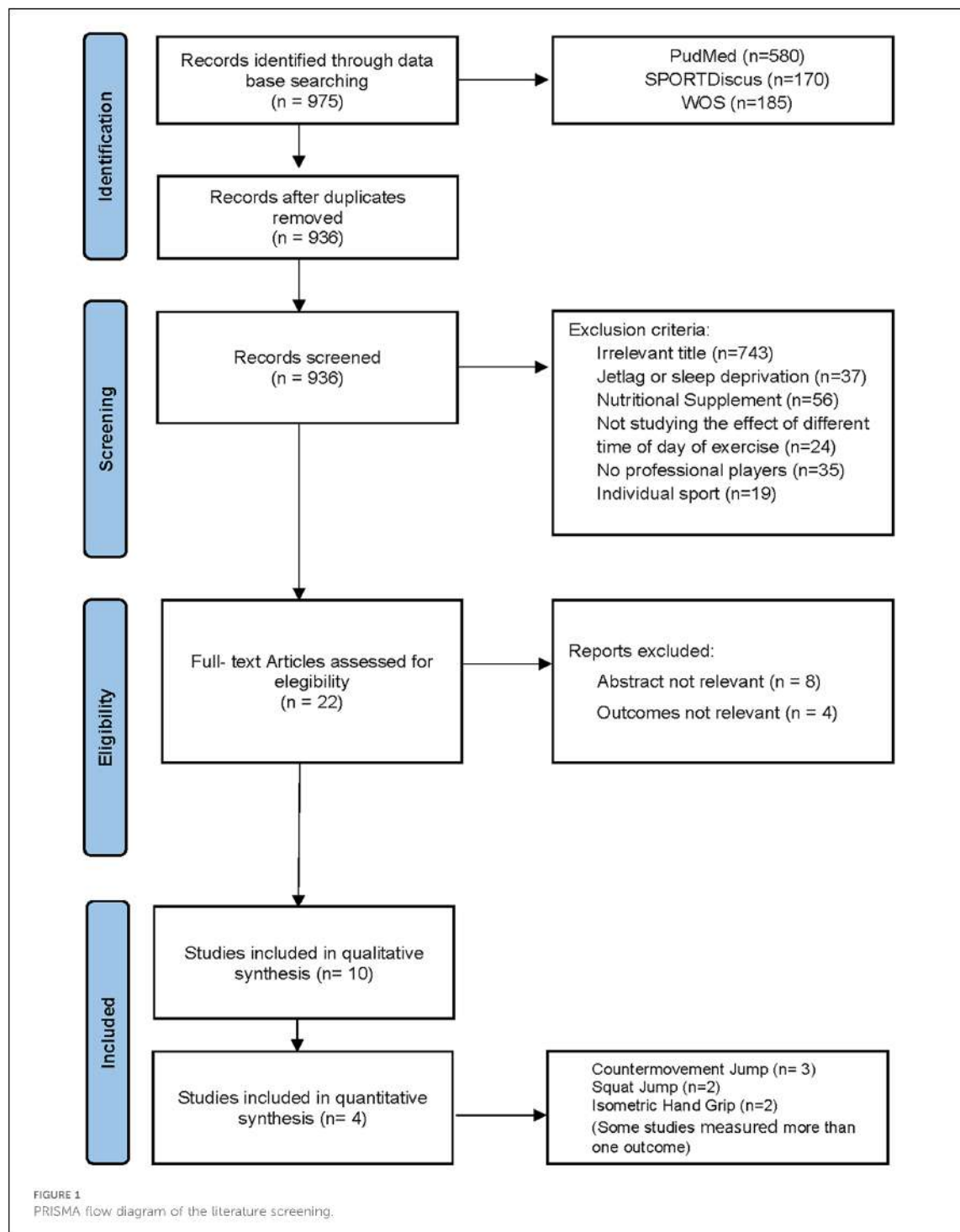


TABLE 1 Effects of time-of-day on neuromuscular performance in professional elite team players.

Study	Participants	Time of day	Design	Main Outcome	Morning	Evening	Key findings
Heishman et al. (27)	10 Elite male NCAA Division 1 basketball player (20.9 ± 6 1.2 years)	7:00–9:00 h vs. 13:45–16:00 h	Retrospective analysis	Countermovement jump height (cm)* Countermovement jump power (W)*	58.3 ± 6 1.4 6,378.0 ± 131.2	61.1 ± 61.9 6,622.1 ± 172.0	CMJ ($p = 0.008$) and Power ($p = 0.004$) were significantly reduced in the morning compared with the afternoon.
Pavlovic et al. (23)	16 elite male handball (25.4 ± 5.8 years)	8:00–9:30 h vs. 18:00–19:30 h	Randomized and cross-over	Zig-zag test (s)* 5 m sprint (s)* 10 m sprint (s)* 20 m sprint (s)* Countermovement jump height (cm)* Squat jump height (cm)* Countermovement jump with arm Swing height (cm)*	4.55 ± 0.16 1.19 ± 0.10 2.03 ± 0.13 3.50 ± 0.25 32.11 ± 4.33 30.54 ± 4.12 38.00 ± 4.52	4.28 ± 0.16 1.06 ± 0.07 1.80 ± 0.09 3.18 ± 0.19 35.75 ± 4.99 33.52 ± 4.09 42.66 ± 4.58	Evening measurements revealed significant increases in jump height for the countermovement jump (CMJ) without ($p < 0.001$) and with arm swing ($p < 0.001$), as well as for the squat jump ($p < 0.001$). Significant improvements were also observed in zig-zag test performance ($p < 0.001$) and sprint times for 5-m, 10-m, and 20-m distances ($p < 0.001$).
Unver & Ann (28)	20 male soccer players (22.20 ± 3.14 years)	9:00 h vs. 14:00 h vs. 19:00 h	Randomized, counter-balance and cross-over	Wingate Anaerobic Power Test (W/Kg)	10.79 ± 1.06	11.30 ± 1.19	No significant differences were found in peak power at 19:00 compared to 9:00 or 14:00 h ($p > 0.05$).
Martin-López et al. (29)	15 female volleyball players (Superliga 2) (22.3 ± 7.2 years)	9:00 h vs. 19:00 h	Randomized, counter-balance and cross-over	Standing spike test (km h ⁻¹)* SEBT posterolateral NO-DOM (cm) Modify Agility T-test (s)* Countermovement jump (cm) Squat jump (cm) Spike jump (cm) Isometric handgrip (N) Repeated sprint ability	64.2 ± 4.8 71.8 ± 6.0 5.8 ± 0.3 30.8 ± 4.4 28.1 ± 3.6 40.2 ± 4.0 288.8 ± 56.8 14.1 ± 1.5	67.1 ± 4.9 75.4 ± 6.5 5.7 ± 0.3 31.0 ± 4.7 28.2 ± 6.6 40.0 ± 4.2 290.9 ± 47.0 15.2 ± 1.6	Maximal ball velocity in the standing spike test was significantly higher in the evening than in the morning ($p = 0.002$). Additionally, the time to complete the modified T-test was lower in the evening compared to the morning ($p = 0.049$).
Pullinger et al. (30)	20 highly motivate male athletes (21.0 ± 2.2 years)	7:30 h vs. 17:30 h	Randomized, counter-balance and cross-over	Distance covered (m)* Average power (W)* Peak velocity (W) Average velocity (Km h ⁻¹)*	2,353 ± 288 19.8 ± 1.2 16.7 ± 1.8	2,537 ± 286 20.4 ± 1.4 18.0 ± 1.9	Distance covered ($p < 0.001$), average power ($p = 0.001$), peak velocity ($p = 0.001$), average velocity ($p < 0.001$) were significantly higher in the evening compared to the morning
Mhenni et al. (31)	15 female Tunisian premier female team handball league (age 20.1 ± 1.5 years)	7:00–8:30 h vs. 17:00–18:30 h	Randomized and cross-over	Isometric handgrip (kg)* Ball-throwing velocity (km.h ⁻¹)* Modify Agility T-test (s)* Sprint Time (s)* Countermovement jump (cm)	28.53 ± 6.34 65.7 ± 7.1 7.61 ± 0.64 6.050 ± 0.278 22.56 ± 3.50	33.46 ± 6.74 68.8 ± 4.7 6.68 ± 0.48 5.568 ± 0.071 23.40 ± 3.29	Isometric handgrip ($p < 0.001$), ball-throwing velocity ($p = 0.027$), and modify Agility T-test ($p < 0.001$) performance decreased in the morning compared to the afternoon/early evening. RSSJA, STbest ($p < 0.001$) and STmean ($p < 0.001$), performance were higher in the evening than in the morning.
Moncef Cherif et al. (32)	22 elite male handball players (Tunisian Pro League champion)	8:00 h vs. 18:00 h	Randomized and cross-over	Ball-throwing velocity (m.s ⁻¹)* IRM BH Squat (Kg)* Countermovement jump (cm)* Repeated sprint ability (s)	22.86 ± 2.18 247.95 ± 33.07 32.97 ± 3.85 6.13	26.21 ± 2.21 272.68 ± 45.05 34.46 ± 4.09 6.05	Significant diurnal variations were observed in the performances of the entire population in BTV ($p < 0.001$), 1-RM BH squat test ($p < 0.001$), and the CMJ test ($p < 0.01$).
Mhenni et al. (22)	15 female team handball players (20.1 ± 1.5 years)	7:00–8:30 h vs. 17:00–18:30 h	Randomized and cross-over	Isometric handgrip (kg)* Ball-throwing velocity (km.h ⁻¹)* Modify Agility T-test (s)* Countermovement jump (cm)	28.53 ± 6.34 65.7 ± 7.1 7.61 ± 0.64 22.56 ± 3.50	33.46 ± 6.74 68.8 ± 4.7 6.68 ± 0.48 23.40 ± 3.29	The best performances of HG ($p = 0.0013$), BTV ($p = 0.027$) and MAT ($p < 0.001$) were recorded in the evening compared to the morning.
West et al. (33)	16 Elite male professional rugby union players (21 ± 4 years)	10:00 h vs. 17:00 h	Randomized and cross-over	Countermovement jump (W)*	5,248 ± 366	5,413 ± 361	Peak power output increased from morning compared to evening ($p < 0.001$)
Hammouda et al. (34)	15 male football players in first division Tunisian league (17.3 ± 0.3)	7:00 h vs. 17:00 h	Randomized and cross-over	Yo-Yo intermittent recovery test (m)*	1,736.64 ± 533.48	2,043.64 ± 533.5	The total distance covered during the Yo-Yo test significantly increased in the evening compared to the morning ($p < 0.01$)

cm, centimeters; N, newtons; Kg, kilograms; W, watts; s, seconds; m, meters; CMJ, Countermovement jump; RSSJA, Repeated shuttle-sprint; ST, Sprint time; BTV, Ball-throwing velocity; 1RM BH squat test, One repetition maximum back half squat at 90; MAT, Modify Agility T-test.

*Statistically significant compared to morning session $p < 0.05$.

in three studies, only the outcome assessment was blinded (23, 29, 32). The risk of attrition bias was deemed unclear in only 2 studies because they lost participants in the follow-up that were needed for analysis (23, 32). The risk of reporting bias was unclear in only 1 study of the meta-analysis (23) (Figure 2).

3.5 Heterogeneity

Heterogeneity was assessed using the I^2 statistic, interpreted according to standard thresholds. For metrics with moderate to high heterogeneity ($I^2 > 50\%$), such as agility ($I^2 = 85\%$), a random-effects model was applied to mitigate the influence of differences between studies. This approach allowed us to estimate a conservative and generalizable average effect while acknowledging the inherent variability in study designs and populations. The high heterogeneity observed in agility metrics, for instance, can be attributed to differences in test protocols (e.g., modified agility T-test vs. zig-zag test) and sample characteristics, including variations in age, sex, and competition level (professional vs. semi-professional athletes). By employing a random-effects model, we accounted for these discrepancies, ensuring that the results reflect the variability across studies while maintaining robustness in the meta-analysis conclusions.

We evaluated the heterogeneity of the four studies included in the meta-analysis. Statistical heterogeneity was calculated for continuous variables (squat and countermovement jump, isometric handgrip strength and agility values). In the three studies that analyzed the agility capacity, heterogeneity was high ($I^2 = 85\%$; $p = 0.001$) (22, 23, 29). Heterogeneity was also high for the two studies including isometric handgrip strength ($I^2 = 57\%$; $p = 0.13$) (22, 29). Heterogeneity was low for three studies that

measured the countermovement jump ($I^2 = 10\%$; $p = 0.33$) (23, 29, 32) and for the two studies that included squat jump ($I^2 = 29\%$; $p = 0.23$) (23, 29). No studies were excluded due to a high risk of bias that could influence the presented heterogeneity.

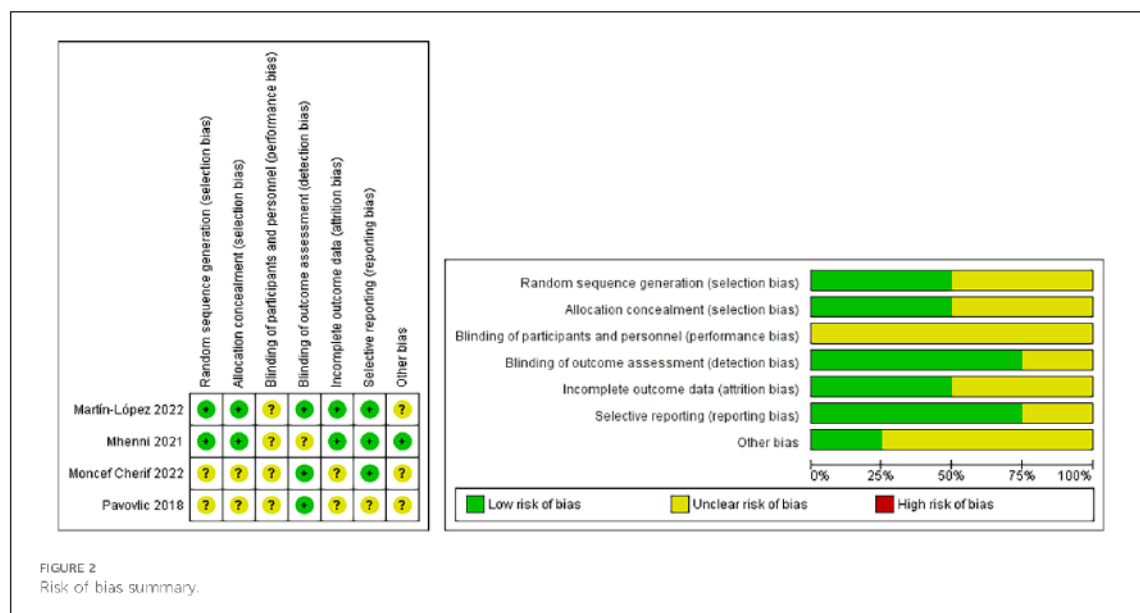
3.6 Quantitative meta-analysis

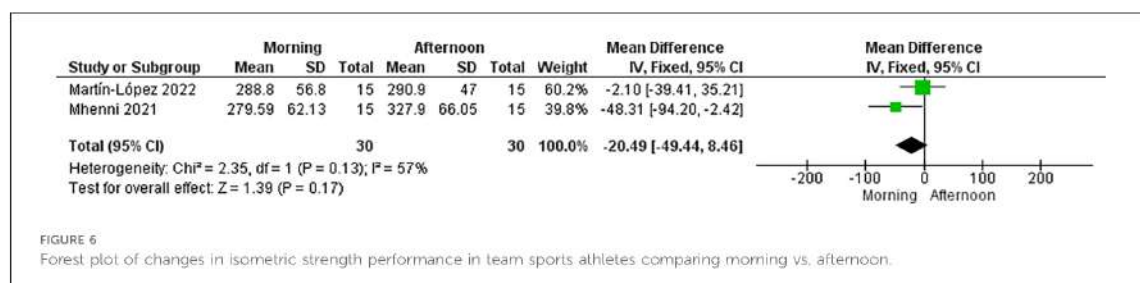
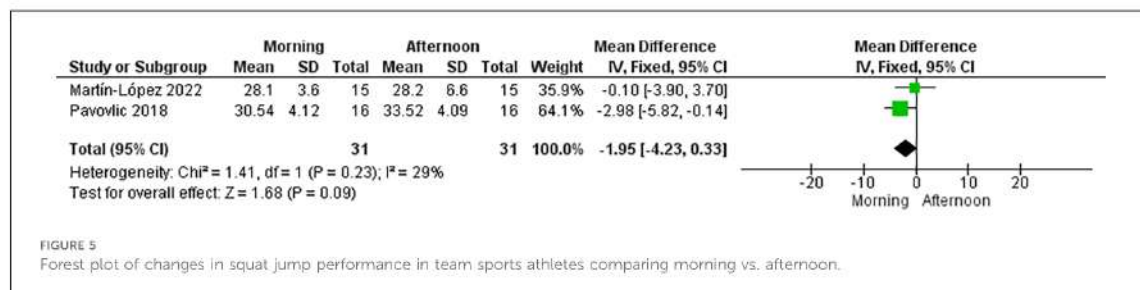
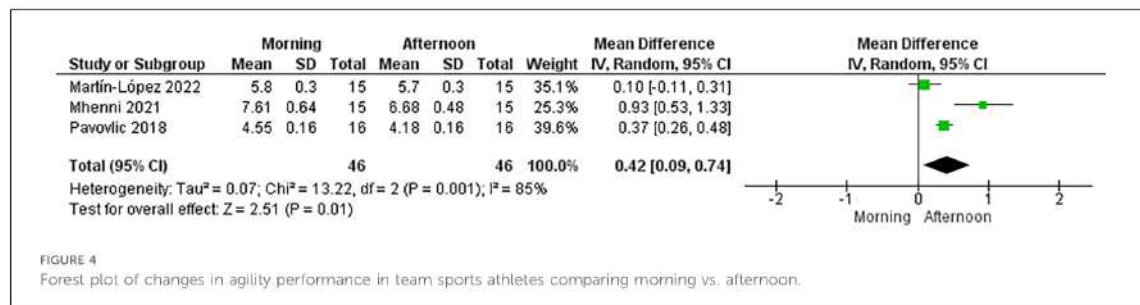
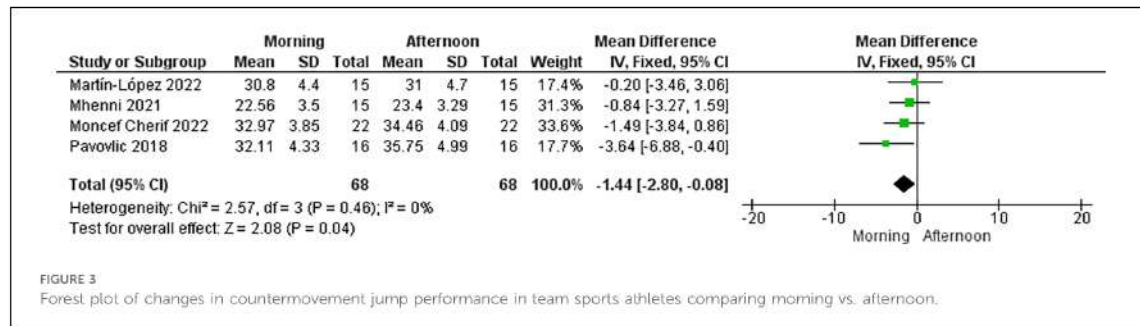
A total of four studies involving 68 athletes (38 males and 30 females) were included in the quantitative meta-analyses. For statistical analysis, data was categorized according to two time-of-days: morning (AM, 7:00–10:00 h) and late afternoon or early evening (PM, 16:00–20:00 h). These categories were applied to each of the following continuous outcomes: countermovement jump (4 studies, $n = 68$ subjects) (22, 23, 29, 32), squat jump (2 studies, $n = 31$ subjects) (23, 29), isometric handgrip strength (2 studies, $n = 30$ subjects) (22, 29) and agility test (3 studies, $n = 46$ subjects) (22, 23, 29). Countermovement jump presented lower values in the AM than in the PM (mean difference, -1.44 ; 95% CI -2.80 to -0.08 ; $p = 0.04$) (Figure 3) and MAT also reported higher values in the PM comparing to AM difference (0.42; 95% CI 0.09 to 0.74; $p = 0.01$) (Figure 4).

No differences were reported between the time-of-day conditions in squat jump (mean difference, -1.95 ; 95% CI -4.23 to 0.33 ; $p = 0.09$) (Figure 5) as same as in isometric handgrip strength (mean difference, -20.49 ; 95% CI -49.44 to 8.46 ; $p = 0.17$) (Figure 6).

4 Discussion

The aim of this systematic review and meta-analysis was to investigate the diurnal variation in neuromuscular performance





of professional or semi-professional team-sports athletes. Our findings indicate a significantly higher performance in the late afternoon and early evening in vertical jumping capacity (i.e., countermovement jump) and agility values compared to morning while no statistically significant differences were obtained in isometric handgrip strength and squat jump, in the meta-analysis that included a population of 68 athletes (38 males and 30 females).

Vertical jump performance is a critical factor in the success of athletes in team sports, as it plays a significant role in various athletic disciplines (Suchomel et al., 2016). This performance is typically evaluated using tests such as the countermovement jump and squat jump, which allow for the measurement of lower limb strength and power output. In our study, three of the four studies included in the meta-analysis showed higher mean vertical jump capacity in the countermovement jump during the late afternoon and early evening compared to the morning (22, 23, 32) while one did not report diurnal differences between protocols (29). Thus, according to our data, there was a consistent pattern despite the limited number of studies that support the fact that countermovement jumps capacity test peaks in the late afternoon and early evening compared to the morning values. Referring to the squat jump, two of the three studies included in this meta-analysis consistently showed higher mean vertical jump capacity not reaching statistical significance. These differences between morning to afternoon and late evening values in vertical jump capacity could be attributed to the typical pattern of increased core temperature that occurs late in the afternoon or early evening, a result of transient changes in heat generation and loss throughout the day (36). This increase in body temperature may induce a passive warm-up effect (37), enhancing metabolic nerve conduction velocity (38) and affecting muscle contractile function (39).

Agility is one of the determining factors in neuromuscular performance in team sports athletes, as comprehend the ability to quickly accelerate, decelerate, change direction and maintain a proper balance (40). According to several studies, agility performance tends to show a peak during the late afternoon and early evening hours. In our study, two of three studies included in this meta-analysis showed an enhancement in agility performance in afternoon-evening compared to morning protocols (23, 31), while the remaining study did not observe this difference (29). Recent findings suggest that elite or professional athletes may be more influenced by time-of-day compared to athletes at other level (e.g., semi-professional athletes) due to elite levels presented higher neural activation and muscular strength/power values that could be more affected by temperature changes and neural recruitment patterns along the day affecting specially in elite athletes comparing to amateur or recreational athletes (41).

Isometric handgrip strength is frequently utilized as an indicator of enhanced force production capacity across both upper and lower body musculature (i.e., weightlifting, paddling), as well as proficiency in executing intricate movements (i.e., rackets or stick skills) (42). Therefore, isometric handgrip is a

cheap and easy test to implement among team sports athletes and can be related to various specific actions involving the upper body actions (e.g., blocking, hitting) that occurs in different teams' sports disciplines (Cronin et al., 2017). In our study, one of the two studies included in this meta-analysis showed statistical diurnal differences (31), while another study did not report differences (29). The differences reported between these studies could be attributed to the specific demands of the team sports disciplines measured (i.e., handball vs. volleyball). Isometric strength demands are notably elevated in handball due to the continuous need for catching, passing, and controlling the ball, which occurs frequently during training and competitions, compared to volleyball where the contact with the ball in hitting, passing, digging or blocking actions occurs in thousandths of a second. Additionally, as previously mentioned, the different levels of competition (i.e., professional vs. semi-professional) may also impact the outcomes presented. The findings of this study reinforce the importance of considering time-of-day variations in neuromuscular performance when designing training and competition schedules for team sport athletes. Beyond the physical aspects, the psychological readiness of athletes also plays a critical role in optimizing performance (12, 13). These findings suggest that coaches should adopt a dual approach, considering both physical and psychological factors when scheduling training sessions. By aligning training schedules with both the physiological and psychological needs of athletes, practitioners can ensure a more comprehensive optimization of performance.

Our systematic review and meta-analysis have several limitations that should be mentioned to enhance its applicability to real sports context scenarios. First limitation of this systematic review and meta-analysis is the scarcity of experimental studies examining the relationship between diurnal variations and neuromuscular performance in professional or semi-professional team sport athletes and that the small sample sizes increase the risk of statistical error and may not capture the full variability in neuromuscular performance across athletes, especially when considering individual differences such as chronotype or sex. Second, is the heterogeneity in study designs, methodologies, and outcome measures across the included studies. Third, the meta-analysis included both male and female participants, which, while necessary due to limited evidence, may have obscured potential gender-specific differences in time-of-day effects on performance. Future research should investigate gender-specific physiological and neuromuscular responses to time-of-day variations to develop tailored performance and training strategies. Fourth, individual differences in chronotype, such as morningness or eveningness preferences, were not fully accounted for in this study. Chronotypes influence performance, with morning-oriented athletes excelling earlier in the day and evening-oriented athletes performing better later. Future research should explore these variations to develop personalized training and competition schedules that align with athletes' natural rhythms. This variability makes it challenging to directly compare and combine the results, leading to potential inconsistencies and reduced generalizability.

5 Conclusion

Neuromuscular performance in professional and semi-professional team-sport athletes is influenced by the time of day, with higher performance levels observed in vertical jump capacity and agility performance during the afternoon and evening (4–8 PM) compared to the morning time (7–10 AM).

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

JM-L: Data curation, Methodology, Writing – original draft, Writing – review & editing, Conceptualization, Formal Analysis, Project administration, Supervision, Validation, Visualization. AP-L: Data curation, Methodology, Writing – original draft, Writing – review & editing. DV-D: Writing – original draft, Writing – review & editing, Formal Analysis, Software. ÁL-S: Writing – original draft, Writing – review & editing, Data curation, Methodology.

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Conflict of interest

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3.2 Estudio 2: Impact of time-of-day and chronotype on neuromuscular performance in semi-professional female volleyball players

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Impact of time-of-day and chronotype on neuromuscular performance in semi-professional female volleyball players

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ABSTRACT

This study aimed to determine if time-of-day could influence physical volleyball performance in females and to explore the relationship between chronotype and volleyball-specific performance. Fifteen young female athletes participated in a randomized counterbalanced trial, performing a neuromuscular test battery in the morning (9:00 h) and the evening (19:00 h) that consisted of volleyball standing spike, straight leg raise, dynamic balance, vertical jump, modified agility T-test and isometric handgrip tests. Chronotype was determined by the morningness-eveningness questionnaire. Compared to the morning, an increased performance was found in the standing spike (4.5%, $p = .002$, ES = 0.59), straight leg raise test (dominant-limb) (6.5%, $p = .012$, ES = 0.40), dynamic balance (non-dominant-limb) (5.0%, $p = .010$, ES = 0.57) and modified T-test (2.1%, $p = .049$, ES = 0.45) performance in the evening; while no statistical differences were reported in vertical jump tests or isometric handgrip strength. Moreover, no associations were found between chronotype and neuromuscular performance ($r = -0.368$ – 0.435 , $p = .052$ – 0.439). Time-of-day affected spike ball velocity, flexibility in the dominant-limb, dynamic balance in the non-dominant-limb and agility tests. However, no association was reported among these improvements and the chronotype. Therefore, although the chronotype may not play critical role in volleyball-specific performance, evening training/matches schedules could benefit performance in semi-professional female volleyball players.

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Introduction

Circadian rhythms refer to the biological variations that occur in the human body during approximately 24-h cycles being regulated by the central pacemaker (i.e., suprachiasmatic nuclei) (Atkinson and Reilly 1996). These time-of-day differences are influenced by several mechanisms such as external (i.e., environmental factors), psychobiological (i.e., lifestyle), internal (i.e., adjustment to time zone), and chronobiological factors (i.e., expression of circadian rhythmicity in an individual) (Douglas et al. 2021; Reilly and Waterhouse 2009; Vitale and Weydahl 2017). These cyclical variations influence some physiological mechanisms such as body temperature (Refinetti and Menaker 1992), glucose/lipid metabolism (Kumar Jha et al. 2015) or hormones regulation (i.e., cortisol, melatonin) (Selmaoui and Touitou 2003), which affect the molecular clock in skeletal muscle (Schroder and Esser 2013) influencing muscle

contraction potentially by causing a reduction in maximal force production (Andrews et al. 2010; Chtourou et al. 2012). Thus, different time-of-day schedules may alter neuromuscular performance in humans (Chtourou and Souissi 2012).

In humans, resistance and short-duration maximal exercise performance are influenced by diurnal fluctuations in metabolism, observing peak of performance at the evening (i.e., 16:00–20:00 h) compared to morning schedules (i.e., 6:00–10:00 h) (Grgic et al. 2019; Mirizio et al. 2020; Pallares et al. 2014; Zarrouk et al. 2012) and effect that seem to occur locally in skeletal muscle nor affecting neural structures (Sedliak et al. 2008). Nonetheless, active warm-ups with or without music, exposures to warm and humid environments, fasting conditions or prolonged training periods at morning hours seem to minimize these time-of-day differences in muscle force and power production (Mirizio et al. 2020). This time-of-day effect on muscle strength and

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power production has been mostly investigated in male athletes, with residual evidence performing in team-sports female athletes (Mhenni et al. 2017).

In addition, chronotype is the expression of circadian rhythmicity and reflects predisposition towards morningness or eveningness, reporting differences between individual or team-sports disciplines (Lastella et al. 2016). Chronotype is defined by three categories: morning type, evening type, or neither type; that indicates the time of that day at which several psychophysiological variables reach their peaks (Vitale and Weydahl 2017). These peaks in psychophysiological variables have been shown to affect sports performance (Vitale and Weydahl 2017; Zarrouk et al. 2012); thus chronotype categorization should be considered to determine the optimal time for training and competition in sports athletes (Vitale and Weydahl 2017).

Volleyball is an intermittent sport characterized by short-duration maximal and explosive movements (i.e., jumps, blocks) developed during continuous defensive or offensive actions (Sattler et al. 2012). The number of on-court players (i.e., six players) and the limited dimension of the court (9 × 18 m) determine that elite volleyball players possess a combination of excellent physical, technical and tactical skills (Lidor and Ziv 2010; Sheppard et al. 2008). Besides anthropometric demands in volleyball (Grgantov et al. 2006), high values of upper/lower strength/power and agility (Paz et al. 2017), jumping capacity (Sattler et al. 2015), flexibility (Lee et al. 1989) and balance (Eylen et al. 2017) are critical for volleyball success.

The impact of time-of-day variances in volleyball performance was previously investigated in male players (Rai and Tiwari 2015), no reporting time-of-day differences in several physical parameters. However, no studies have explored morning-evening performance in female volleyball players and limited evidence has been presented about the association between chronotype and female team-sports performance (Lastella et al. 2021). Thus, the aim of this investigation was to determine the effect of time-of-day and chronotype on neuromuscular performance in semi-professional volleyball female players. It is hypothesized that despite the neuromuscular performance and time-of-day variability previously reported in female team sports (e.g. handball) (Mhenni et al. 2017), time-of-day will affect strength and power production of semi-professional female volleyball players. It is hypothesized a higher performance would be found in the evening compared to the morning trial, and that improvement would be related to the chronotype of the female volleyball players.

Methods

Participants

Fifteen female volleyball players of the Superliga 2 (Second Spanish National Division League) participated in this study (age 22.3 ± 7.2 years, height 1.72 ± 0.04 m, body mass 64.5 ± 5.9 kg, body mass index 21.9 ± 1.9 , volleyball experience 12.0 ± 6.6 years and training per week 6.9 ± 1.2 hours). Semi-professional female volleyball players between 16 and 40 years old who routinely train (> 5 h/week), compete in both the morning ($\sim 9:00$ hours) and evening ($\sim 19:00$ hours) and were able to perform their maximum effort in trials were eligible for inclusion. Exclusion criteria were: (a) impossibility of being tested due to previous upper/lower limbs injuries, (b) experiencing lower limb muscle soreness at time of the testing session, (c) taking any medication, (d) consuming stimulants during the 48 h before the trial (e.g. caffeine, tea). Nine participants were tested during the follicular phase of their menstrual cycle, and six were tested during the luteal phase according to a mobile application (Mycalendar; Period Tracker, Singapore) that identifies main events occurring during the menstruation cycle (de Jonge, 2003). The players and parents/guardians (5 participants were under 18 years old) were informed of all experimental procedures and informed consent was completed before participation. The study was approved by the Francisco de Vitoria Bioethics Commission University (13/2021), which complied with the recommendations of the Declaration of Helsinki.

Experimental design

A randomized and counter-balance design was used in this investigation. The required sample size was obtained by an effect size of 0.78 in Cohen's *d* units (statistical power of 80% with type I error set at 5%) and also considering the members of the volleyball team. Female volleyball players performed the same neuromuscular test battery on two occasions in the morning at 9:00 h and the evening at 19:00 h. The time-of-day order was randomized for each participant. These two times of the day (9:00 h and 19:00 h) were chosen according to the reported habitual training session (19:00 h) and competition schedules (9:00–10:00 h and 19:00 h). Before testing, participants underwent one familiarisation session with the experimental test battery to prevent bias from progressive learning 3 days before the first day of the study at 19:00 h according to the time-of-day selected in our study and female participants realized all the tests included in the neuromuscular battery. To ensure standardisation of the measurements,

all tests were completed at the same volleyball court, where participants used to train and compete, using the same testing devices, handled by the same researcher, in the same order. During the 3 sessions, the air temperature was $6.6 \pm 0.5^\circ\text{C}$ and relative humidity was $45 \pm 3\%$ measured with a portable weather station (Meteorological Station, Küken, Spain). The temperature conditions were determined by the Spanish COVID-19 policy in sports centers that stipulates that a constant airflow renovation must be accomplished in the building facilitated by the opening of all windows.

Experimental protocol

During the 24 h before testing, volleyball players abstained from any strenuous activity and were encouraged to refrain from all dietary sources of caffeine, alcohol, and stimulants 48 h before the experimental trials (Mora-Rodríguez et al. 2015). During the familiarization session, participants completed the Horne–Ostberg Morningness–Eveningness personality questionnaire (Horne and Ostberg 1976) to assess their chronotype. The night before the morning testing session, participants ingested an easily digestible meal with a total energy intake of 759 kcal (50% carbohydrates, 30% fats and 20% proteins) at 21:00 h. Before morning trials, at 7:00 am participants ingested a standardised breakfast of 453 kcal (53% carbohydrates, 42% fats and 5% proteins) and for the evening testing sessions, participants

ingested a menu of 639 kcal (35% carbohydrates, 42% fat and 23% proteins) at 13:00-hours. Athletes were encouraged to sleep 7 to 9 hours the night before trials (Charest and Grandner 2020; Fullagar et al. 2015).

The day of the trial, the participants arrived at their habitual training facility at 8:30 h (morning session) or 18:30 h (evening session). Upon arrival, body composition was assessed by electric bioimpedance (Tanita 901, Tanita Corp., Tokyo, Japan) and tympanic temperature was evaluated in triplicate using a portable thermometer (Thermoscan 6520, Braun, Germany). Besides, hydration status was analysed using urine-specific gravity (Master-s28M, Atago Company, Tokyo, Japan), before instructing participants to drink 500 mL of water 1 h before the trial to ensure euhydration status (< 1.020) (Osterberg et al. 2009).

After initial tests, a 20 minutes standardized warm-up was performed. The warm-up protocol consisted of 5 minutes of continuous running, followed by upper- and lower-body dynamic stretching and specific volleyball exercises (Perez-Lopez et al. 2015; Perez-Lopez and Valades 2013). Then, participants underwent a battery of neuromuscular tests consisting of standing and jumping spike test, straight leg raise test, modified star excursion balance, vertical jumps tests (squat jump, countermovement and spike jump), isometric handgrip strength and modified agility T-test (Figure 1). Finally, during the 30 minutes after the neuromuscular test battery finalization, rating of perceived exertion (RPE) scale (0–10) was determined based on the methodology previously described (Foster et al. 2001).

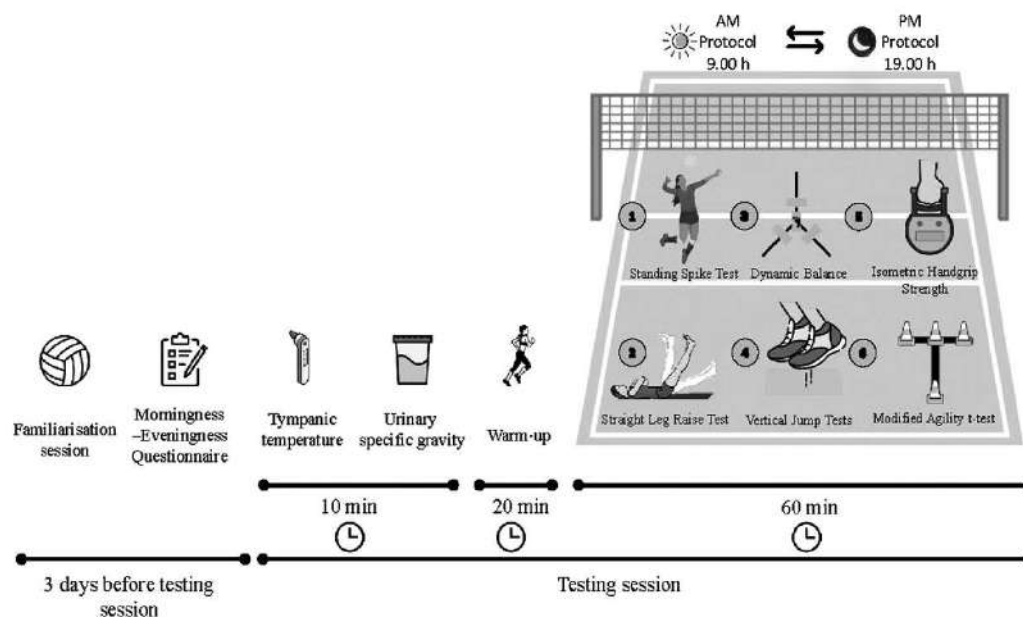


Figure 1. Experimental design.

Standing spike test

Standing spike test was performed according to the protocol previously described (Palao and Valades 2009b, 2016). In brief, in the standing spike test, female players hit the ball with maximal strength toward a target zone (1.5 × 1.5 m). Player self-tossed the ball and they spiked without jumping or moving their feet of the ground. A researcher was located laterally to the player to monitor the execution of both tests. If any execution aspect was incorrect, the test was repeated (e.g., not hitting the ball directly toward the target area). The same ball was used during the trial (Molten V5M5000) and in the target zone a Stalker Solo 2 Radar Gun (Applied Concepts, Texas, USA) was situated to measure ball velocity. Ball pressure, and radar orientation and calibration were monitored on each test. Each player had three trials and the maximum speed reached in the test was recorded (Palao and Valades 2009a).

Straight leg raise test

Passive hip flexion range of motion (ROM) values with the knee extended were evaluated with the Straight Leg Elevation test such as previously described (Moreno-Perez et al. 2021). Participants made two maximum passive attempts for the dominant and non-dominant leg, when the difference between one attempt and another was greater than 5%, a third attempt was made, selecting the highest value of the two attempts whose results were similar for further statistical analysis (Moreno-Perez et al. 2021). A unilevel inclinometer ISOMED (Portland, OR, USA) with a telescopic was used for the measurements and the inclinometer was placed approximately on the external malleolus and the distal arm was aligned parallel to an imaginary bisecting line of the extremity (Lopez-Samanes et al. 2021).

Dynamic balance

Dynamic balance was assessed using the OctoBalance device (OctoBalance, Check your Motion, Albacete, Spain), a modified version of the Star Excursion Balance Test (SEBT) measuring in three distinct directions of anterior, posteromedial and posterolateral on both the dominant and non-dominant limbs. The measurement system is based on an extending measuring tape, magnetized in each direction to an octagon-shaped platform. Each trial consisted of pushing the marked point, situated at the top of the measuring tape, with the toes as far as possible in the designated direction (Gonzalo-Skok et al. 2015). Subjects were allowed at least three practice trials in the anterior, posteromedial and posterolateral directions before recording the best of three formal trials in each plane. The maximal

reach distance was recorded where the most distal part of the foot was reached based on the measurement device.

Countermovement jump, squat jump and maximal spike jump test

Participants performed three maximal vertical jumps of repetitions of a countermovement jump, squat jump and spike jump with 45 seconds of passive recovery between repetitions. Countermovement jump and squat jump were performed according to standard methodology (Bosco et al. 1983), while for spike jump, a basic spike technique consisted of 2 approach steps, jump and land was used. For the three test, maximal jump height was recorder using an infrared jump system (Optojump, Microgate, Italy).

Isometric handgrip strength

Participants performed a isometric handgrip strength test with 0 degrees of shoulders flexion, 0 degrees of elbow flexion and the forearm and hand in a neutral position, as previously reported in other studies (Lopez-Samanes et al. 2017). Two maximum isometric voluntary contractions were measured from the dominant and non-dominant hand using a calibrated handgrip dynamometer (Takei 5101, Tokyo, Japan). The highest value out of two attempts was recorded as the maximum voluntary handgrip strength.

Modified agility T-test

The agility T-test required female players to move through a modified T-shape circuit to simulate the fast movements performed in volleyball (e.g., digs or receptions) (Sassi et al. 2009). Participants began the test with both of their feet behind the starting point and sprinted forward to cone B and touched its base with the right hand, facing forward and without crossing feet, they shuffled to the left to cone C and touched its base with the left hand. Then, participants shuffled to the right to cone D and touched its base with the right hand. They shuffled back to the left to cone B and touched its base. Finally, female players ran backward as quickly as possible and returned to line A. The best performance out of two repetitions separated by a 2 minutes recovery period was recorded for subsequent analysis (Munoz et al. 2020). Time to complete the T-shape circuit was measured using two electronic time sensors (Microgate, Polifemo Radio Light, Italia) set 1 m above the surface and positioned 3 m apart facing each other on either side of the starting line. Participants began each test 1 m behind the starting line, and the timer started when they passed the first gate.

Morningness-eveningness questionnaire

The morningness-eveningness questionnaire (Horne and Ostberg 1976) was used to determine the participants' chronotype (morning, intermediate or evening). The questionnaire consists of 19 questions and yields scores ranging from 16 to 96, with lower scores indicating participants' preference toward evening activities and higher scores indicating participants' toward morning activities. Chronotype scores were determined using the Horne and Ostberg classification system (16–41 = evening type; 42–58 = intermediate type; 59–86 = morning type).

Statistical analysis

Data are presented as means and standard deviations. Shapiro–Wilk test was used to assess the normal distribution of data. All variables presented a normal distribution in the test ($p > .05$); hence, differences between experimental condition (morning vs. evening) were assessed using a paired *t*-test. Linear regression analysis was used to examine the relationship between performance variables and the morningness-eveningness questionnaire. The significance level was set at $p \leq .05$. Cohen's formula for effect size (ES) was used and the results were based on the following criteria: trivial (0–0.19), small (0.20–0.49), medium (0.50–0.79) and large (0.80 and greater) (Cohen 1992). All the statistical analyses were done using the SPSS software version 26 (SPSS Inc., Chicago, IL, USA).

Results

Environmental temperature, tympanic temperature, urinary specific gravity and rate of perceived exertion

Tympanic temperature was significant elevated by 1.2% in the evening versus morning trial (35.9 ± 0.5 vs. $36.3 \pm 0.3^\circ\text{C}$, respectively; $p = .006$; ES = 1.00). Therefore, similarly hydration status measured by urinary specific gravity (1.017 ± 0.01 vs 1.018 ± 0.01 , $p = .697$; ES = 0.07) and rate of perceived exertion values 4.1 ± 1.5 vs 4.1 ± 1.6 points ($p = .818$; ES = 0.03) were reported between morning and evening conditions.

Morningness-eveningness questionnaire

Regarding the participants' chronotype assessed by the Morningness-eveningness questionnaire, nine participants (60%) scored as "intermediate type," five participants (33.3%) as evening type and one participant (6.6%) as morning type.

Table 1. Neuromuscular performance differences between morning and evening protocols in semi-professional volleyball players.

Neuromuscular variables	Morning	Evening	ES	Descriptor
Standing spike test ($\text{km}\cdot\text{h}^{-1}$)	64.2 ± 4.8	$67.1 \pm 4.9^*$	0.59	Moderate
Straight Leg Raise Test DOM ($^\circ$)	84.3 ± 13.6	$89.8 \pm 13.2^*$	0.41	Small
Straight Leg Raise Test NO-DOM ($^\circ$)	83.3 ± 14.8	88.3 ± 11.6	0.38	Small
SEBT anterior DOM (cm)	64.9 ± 6.5	65.0 ± 5.3	0.02	Trivial
SEBT posterolateral DOM (cm)	73.4 ± 4.9	75.5 ± 6.6	0.36	Small
SEBT posteromedial DOM (cm)	75.7 ± 6.8	77.9 ± 7.1	0.32	Small
SEBT anterior NO-DOM (cm)	64.9 ± 4.4	65.4 ± 2.8	0.14	Trivial
SEBT posterolateral NO-DOM (cm)	71.8 ± 6.0	$75.4 \pm 6.5^*$	0.57	Moderate
SEBT posteromedial NO-DOM (cm)	75.8 ± 6.7	75.6 ± 7.8	-0.03	Trivial
Countermovement jump (cm)	30.8 ± 4.4	31.0 ± 4.7	0.05	Trivial
Squat jump (cm)	28.1 ± 3.6	28.2 ± 6.6	0.02	Trivial
Spike jump (cm)	40.2 ± 4.0	40.0 ± 4.2	-0.06	Trivial
Isometric handgrip strength DOM (N)	288.8 ± 56.8	290.9 ± 47.0	0.07	Trivial
Isometric handgrip strength NO-DOM (N)	278.8 ± 49.6	264.6 ± 47.0	-0.29	Small
Modified T-test (s)	5.8 ± 0.3	$5.7 \pm 0.3^*$	-0.45	Small

Abbreviations: DOM = dominant; NO-DOM = non-dominant; ES = Effect size; SEBT = Star Excursion Balance Test; $^\circ$ = degrees; cm = centimeters; N = newtons; * = statistically significant compared to morning session

Standing spike, straight leg raise test and star excursion balance test

In the standing spike test, maximal ball velocity was significantly higher in the evening compared to the morning session ($4.5 \pm 4.4\%$, $p = .002$, ES = 0.59). Besides, we found statistically significant differences in the straight leg raise test in the dominant limb during the evening session compared to the morning session ($6.5 \pm 9.4\%$, $p = .012$, ES = 0.41), but not in the non-dominant limb ($6.1 \pm 16.1\%$, $p = .091$; ES = 0.38) (Table 1). Moreover, no statistically significant differences in the SEBT morning vs evening were found dominant limb in the anterior direction (0.2%, $p = .929$, ES = 0.02), posterolateral (2.9%, $p = .605$, ES = 0.36) and posteromedial (3.0%, $p = .233$, ES = 0.32). Finally, when comparing the evening vs morning session, statistically significant differences were found in the SEBT posterolateral direction of the non-dominant limb (5.0%, $p = .010$, ES = 0.57) but not in the anterior (0.8%, $p = .063$, ES = 0.14) or the posteromedial (-0.3% $p = .831$, ES = 0.03) (Table 1).

Countermovement jump, squat jump test and maximal spike jump tests

No statistically significant differences were found in countermovement jump test (0.7%, $p = .621$, ES = 0.05), squat jump (0.4%, $p = .756$, ES = 0.02) and

maximal spike jump test (-0.6% , $p = .610$, $ES = 0.11$) when comparing evening versus morning sessions (Table 1).

Isometric handgrip strength and modified T-test

Comparing morning and evening trials, there were no statistical differences for the maximal isometric handgrip strength in the dominant hand ($1.4 \pm 15.0\%$, $p = .714$, $ES = 0.07$) and non-dominant hand ($-5.1 \pm 10.0\%$, $p = .095$, $ES = 0.29$). Finally, the time required to complete the modified T-test in the evening was $2.1 \pm 3.9\%$ lower comparing morning ($p = .049$, $ES = 0.45$) (Table 1).

Correlations

None of the correlations between morningness-eveningness questionnaire scores and the time-of-day related differences in the neuromuscular battery tests reached statistical significance: spike test ($r = 0.043$; $p = .439$), straight leg raise test dominant ($r = -0.066$; $p = .407$) and non-dominant ($r = 0.365$; $p = .090$), dynamic balance dominant (i.e., anterior, posteromedial and posterolateral) ($r = 0.184-0.435$; $p = .052-0.254$) and non-dominant ($r = 0.055-0.178$; $p = .262-0.421$), countermovement jump ($r = -0.368$; $p = .088$), squat jump ($r = -0.264$; $p = .171$), spike jump test ($r = 0.316$, $p = .125$), isometric handgrip strength dominant ($r = -0.008$; $p = .384$) and non-dominant ($r = 0.310$, $p = .095$) and T-test ($r = -0.030$, $p = .357$).

Discussion

The aim of this study was to assess the effect of two different time-of-day schedules (9:00 vs 19:00 hours) on short-duration maximal physical performances, which could be determinant for the physical volleyball performance and to determine. Secondly, to assess if individual chronotype is associated with sports performance in semi-professional volleyball female players. Our findings showed higher neuromuscular performance in standing spike test, flexibility, dynamic balance and agility performance (ranging from 2.1% to 6.5%) in the evening versus morning session, while no difference was reported in vertical jump capacity and isometric handgrip strength.

Compared to the morning, moderate improvements in the evening were observed in the standing spike test (4.5%). To our knowledge, no study has examined the time-of-day effect on this particular but essential aspect of volleyball performance that is particularly important for hitters volleyball players

(Duncan et al. 2006). However, our data are in agreement with previous studies in other intermittent sports that have reported similar improvements evening versus morning in specific hitting tests (4.0–5.4%) of different sport modalities that involve overhead strokes such as serve velocity in tennis (Lopez-Samanes et al. 2017) or throwing velocity in handball (Mhenni et al. 2017).

Vertical jump height is another important factor related to volleyball performance success, involved in some specific movement patterns in defensive (i.e., block jumps) or offensive actions (Sattler et al. 2012). In addition, it has been reported that volleyball players realized a mean of 37–88 jumps per training session depending on volleyball playing role (Garcia-de-alcaraz et al. 2020), thus, establishing the effects that time-of-day provoked in jump capacity could be relevant for volleyball performance. According to our data, no time-of-day effects were reported in jump capacity in the different tests selected for this study (e.g., countermovement jump, squat jump and maximal spike jump test) (0.4–0.7%) that is in agreement with previous studies performed in female team-sports athletes (i.e., handball) in best jump performance (i.e., countermovement jump) (Mhenni et al. 2017). However, our data are controversial with other studies in team or individual male sports that reported improvements in jump capacity in the evening compared with the morning conditions (i.e., 4.5–9.8%) (Lopez-Samanes et al. 2017; Pavlovic et al. 2018). Thus, players' sex, competition level or experience could be some of the variables that explain the lack of statistically significant effects caused by the two time-of-day protocols used in female semi-professional volleyball players.

An adequate range-of-motion (Panoutsakopoulos et al. 2021) and dynamic balance (Hudson et al. 2016) in the lower limbs is essential in volleyball performance due to the continuous jumps, landings, hits and multidirectional movements that volleyball players realized during training and competitions. Previous data have reported statistically significant time-of-day effects on flexibility values in the evening versus morning conditions in the straight leg raise test (Gifford 1987) that are in agreement with the data reported in the dominant (6.5%) and a tendency in the non-dominant limb (6.1%). Limited literature has explored the effect of time-of-day on dynamic balance, especially in team sports athletes. In this case, we only found statistical time-of-day differences in the no dominant limb in a posterolateral direction (5.0%), not reporting differences in the anterior (0.3–0.8%). In addition, no statistical differences in the dominant limb were reported (0.2–3.0%). Our results

agree with previous studies in the literature that established that time-of-day had a minimal effect on dynamic balance in athletes (Heinbaugh et al. 2015).

Our study reported no time-of-day effects in isometric handgrip strength in dominant and non-dominant limbs (1.4–5.2%). These findings agree with previous studies that did not report statistical differences in Olympic combat athletes or competitive tennis players (Lopez-Samanes et al. 2017; Pallares et al. 2016), but in contrast to the differences observed in male handball athletes (Mhenni et al. 2017). This differences between studies could be attributed to contrasting participants involved and isometric handgrip protocols used.

Agility has been related to volleyball performance, during game actions, volleyball players need to realize quick forward/backward movements and continuous change of direction (Paz et al. 2017). According to our findings, the time-of-day effect on agility performance with statistically significant differences in the evening compared to the morning session (–2.1%). Our data agree with previous studies reported in male/female team sports athletes that found lower times in different agility tests in the evening than morning conditions (i.e., soccer and handball) (Mhenni et al. 2017; Pavlovic et al. 2018; Reilly et al. 2007). Thus, agility performance seems to be influenced by time-of-day showing better performance in the evening compared morning conditions.

Morningness and evening questionnaire is a commonly used for detecting morningness, intermediate and eveningness chronotypes. However, our data reported no significant correlations with neuromuscular performance parameters measured in semi-professional female volleyball players. In addition, our results agree with previous literature studies in other intermittent sports that reported no significant relationship chronotypes and neuromuscular performance (Lopez-Samanes et al. 2017). Besides limited number of participants, another possible explanation may stem from the fact, that questionnaires used for chronotype detection aim rather for sleep and psychological factors that for maximum physical performance. Indeed, the study of Kuusmaa et al. (2015) identified the high morning performance types, the high evening performance types and the neutral types who showed significantly different diurnal rhythms in force production, irrespective of their actual chronotype. Therefore, the questionnaires designed to determine the chronotype may not always be sensitive enough to determine the “morningness” or “eveningness” in maximal neuromuscular performance (Kuusmaa et al. 2015). Finally, although some previous studies have reported that chronotype could influence the RPE scores to submaximal physical test (e.g., self-pace walking,

submaximal cycle test) performed in the morning (Kunorozva et al. 2014; Vitale et al. 2013), according to our data RPE values could not influence in maximal physical task (i.e., spike test, modified T-test) in semi-professional female volleyball athletes.

Aside from its strengths, the current investigation has several limitations that should be discussed to enhance its applicability to real sports contexts. First, we only studied the effect of time-of-day and chronotype in semi-professional female volleyball players. Second, we only tested two times (9:00 and 19:00 h) of the whole circadian rhythm spectrum and maybe the differences in performance could be larger if other times of day are included (i.e., 06:00 and 14:00 h). Third, although the sample size recruited ($N = 15$) was based on power calculation and the number of athletes that formed a semi-professional volleyball team, future studies should involve a large number of female athletes with similar training routines to corroborate the results obtained in this study.

Conclusion

Time-of-day affected spike ball velocity, flexibility in the dominant-limb, dynamic balance in the non-dominant-limb and agility tests, observing a higher performance in the evening compared to the morning trial. However, no association was reported among these improvements and the chronotype. Therefore, although the chronotype may not play critical role in volleyball-specific performance, evening training/matches schedules could maximize physical performance in semi-professional female volleyball players.

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3.3 Estudio 3: Circadian phenotype influence on neuromuscular performance in male volleyball players

ESTUDIO 3

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Circadian phenotype influence on neuromuscular performance in male volleyball players

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ABSTRACT

Circadian phenotype (i.e. chronotype) may affect athletic performance across different times of day, but its role in team sports is underexplored. The aim of this study was to assess the impact of chronotype on neuromuscular performance in male volleyball players. Forty-six male volleyball players (age: 26.8 ± 7.9 years) completed the Morningness-Eveningness Questionnaire and were classified as morning-type ($n = 12$), intermediate-type ($n = 15$) or evening-type ($n = 19$). Then, in a randomized and cross-over design, participants completed a neuromuscular test battery comprising vertical jumps (squat jump, countermovement jump, spike jump), spike tests (standing and jump spike), agility (modified T-test), isometric handgrip strength, and dynamic balance; performed in the morning (8:00–10:00 AM) and the evening (18:00–20:00). No significant circadian phenotype by time-of-day interaction was found ($p = 0.133$ – 0.743 , $\eta_p^2 = 0.09$ – 0.14). However, time-of-day effect was found in squat jump, countermovement jump, spike jump, standing spike test, and non-dominant handgrip strength ($p = 0.001$ – 0.046 , $\eta_p^2 = 0.09$ – 0.41), observing an evening improvement in performance ranging from 1.8% to 3.0% ($p = 0.001$ – 0.035 , $d = 0.47$ – 1.16). circadian phenotype had limited impact on neuromuscular performance while diurnal variation enhanced evening performance with a significant role in vertical jump ability and spike velocity among male volleyball players.

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strength/power; volleyball

Introduction

Circadian phenotype (i.e. chronotype) describes individual variations in daily patterns of sleep-wake cycles, hormone secretion, body temperature fluctuations, feeding behaviour or cellular metabolism that occurs in each individual (Facer-Childs and Brandstaetter 2015). Circadian phenotype is usually evaluated using self-report questionnaires, such as the Morningness-Eveningness Questionnaire (Horne and Ostberg 1976) which consists of a 19-item questionnaire assessing preferences for sleep and engagement in various activities related to physical activity or work, which classifies individuals as morning-types, evening-types, or neither-(intermediate)-types based on their preferred times for activity (Mulè et al. 2020). These questionnaires reflect natural patterns of peak alertness and energy, as circadian phenotype significantly influences an individual's efficiency and productivity throughout the day. Thus, circadian phenotype could play a key role

in determining optimal times for physical performance (Roenneberg et al. 2003; Facer-Childs et al. 2018). This is because the body's internal processes, including hormonal secretion, body temperature regulation, and metabolism, are naturally synchronized with environmental cycles, such as light and darkness. These biological rhythms regulate periods of peak alertness and energy, which, in turn, influence cognitive and physical performance across the day (Roenneberg et al. 2007). By aligning bodily functions with environmental cues, individuals can optimize their physical and cognitive output. In addition, circadian phenotype arises from a complex interplay between genetic (Carpen et al. 2006; Pedrazzoli et al. 2010) and environmental factors (Allebrandt and Roenneberg 2008), both of which significantly influence an individual's tendency toward morningness or eveningness. This, in turn, contributes to variations in cognitive (Schmidt et al. 2012) and physical performance preferences (Vitale and Weydahl

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2017) at different times of the day among individuals. Although evidence remains limited, recent studies suggest that circadian phenotype may play a significant role in driving these diurnal physical performance variations (Küüsmaa et al. 2015; Mulè et al. 2020; Roveda et al. 2020).

Thus, exploring chronotype questionnaires combined with time-of-day performance analysis help to understand how circadian rhythm affects performance. In this regard, strength and power tasks (e.g. isometric strength) have shown to vary throughout the day, with most participants exhibiting higher performance in the afternoon than the morning. Also, rating perceived exertion analysis revealed that morning types perceive higher effort in the afternoon session than the morning; by contrast, evening types were more fatigued in the morning session (Mulè et al. 2020).

While circadian rhythms and phenotypes are known to influence physiological and cognitive performance, their relevance must be examined within the specific demands of specific sports (e.g. volleyball). Volleyball is a team sport played on a court of limited dimensions (9 × 18 m) with six players per team. At the elite level, performance requires a combination of repeated bouts of explosive power, rapid anaerobic energy production, and precise neuromuscular coordination (Sheppard et al. 2009). During volleyball training/competitions key actions such as spikes, vertical jumps, or blocks may critically affect specific outcomes and performance. Despite these demands, training sessions and competitive matches are often scheduled without reference to athletes' circadian phenotypes, highlighting the need to investigate how biological rhythms intersect with the sport's distinct physiological and performance requirements (Sattler et al. 2012). To the best of the authors' knowledge, only one previous study in team sports athletes developed by Roveda et al. (2020) has examined the impact of different circadian phenotypes (i.e. morning-types, evening-types, and intermediate-types) on motor skill performance in 141 adolescent soccer players. The findings of this study showed that morning-types exhibited better neuromuscular performance during morning sessions, whereas evening-types performed better in the evening, while no significant differences in performance were observed for intermediate-types across different times of the day (Roveda et al. 2020). Since sports like volleyball and basketball involve competition at any time of day, investigating chronotype effects in these contexts is of considerable practical importance. Thus, the first aim of this study was to examine the influence of circadian phenotype on neuromuscular performance in male volleyball players.

The second aim was to determine diurnal variations in neuromuscular performance in male volleyball players. Based on previous evidence, athletic performance tends to align with individual phenotype preferences (Küüsmaa et al. 2015; Vitale and Weydahl 2017; Roveda et al. 2020), thereby we hypothesized that neuromuscular performance in volleyball players would be enhanced at the time of day corresponding to each athlete's circadian phenotype. Specifically, morning-types were expected to perform better in morning sessions (08:00–10:00 h), evening-types in evening sessions (19:00–21:00 h).

Methods

Participants

Forty-six male volleyball players (age: 27 ± 8 years; height: 184 ± 7 cm, body mass: 81.4 ± 11.7 kg, body mass index: 23.9 ± 2.7 ; volleyball experience: 12.4 ± 7.8 years; training per week 6.7 ± 3.3 h) were included in this study. Participants were instructed to maintain their daily habits throughout the intervention period. All participants had a minimum of five years of federated competition experience and regularly engaged in both morning (i.e. ~9:00) and evening (i.e. ~19:00) training sessions, which were the time points selected for the present study. The exclusion criteria included: (a) inability to undergo testing due to previous upper or lower limb injuries, (b) presence of lower limb muscle soreness during the testing session, (c) current use of medication, (d) chronic diseases and (e) consumption of stimulants, such as caffeine or tea, within 48 h prior to the trial. Participants were fully informed about the experimental procedures, and formal written consent was obtained before their inclusion in the study. Five participants were under 18 years old; in these cases, minors and their parents or guardians were fully informed about the experimental protocol and signed an informed consent form. The study received ethical approval from the Francisco de Vitoria Bioethics Commission University (13/2021) in accordance with the principles outlined in the Declaration of Helsinki.

Experimental Design

This investigation employed a randomized and cross-over design. Participants' circadian phenotypes (i.e. morning, intermediate, or evening) were assessed using the Morningness-Eveningness Questionnaire (MEQ) (Horne and Ostberg 1976). This 19-item questionnaire yields scores from 16 to 86, where lower scores indicate a preference for evening activities and higher

scores for morning activities. Circadian phenotype classification followed the Horne and Ostberg questionnaire system: establishing categories between evening type (16–41), intermediate type (42–58), and morning type (59–86) (Horne and Ostberg 1976). Each participant completed two testing sessions, one in the morning (08:00–10:00) and one in the evening (18:00–20:00), separated by at least 48 h. The order of testing sessions (morning vs. evening) was determined using a computer-generated randomization procedure in blocks according to participants' chronotype. Neither the evaluators nor the athletes were aware of each participant's circadian phenotype during data collection, ensuring blinding and minimizing potential bias. To ensure measurement consistency, all assessments were conducted in the same sports facilities using identical devices and a predetermined standardized sequence, directed by the same researcher with expertise in volleyball performance evaluation. Volleyball players wore the same athletic footwear on both testing days to eliminate any potential performance variability due to differences in shoe design. Additionally, all athletes were instructed to maintain a consistent sleep pattern (i.e. sleep between 7–9 h) the night before each test, replicate the diet the 24 h before each trial and to refrain from intense physical activity for 48 h prior to the assessment (Martín-López et al. 2022). The air temperature (18.4 ± 5.2 °C) and relative humidity ($45.7 \pm 11.7\%$) were replicable for each participant during the tests conducted in both the morning and afternoon, as measured by a portable weather station (Meteorological Station, Küken, Spain).

Experimental Protocol

On the day of testing, athletes' body composition was assessed using electronic bioimpedance analysis (Tanita 901, Tanita Corp., Tokyo, Japan), and tympanic temperature was measured in triplicate with a portable thermometer (Thermoscan 6520, Braun, Germany). To ensure a euhydrated state ($USG < 1.020$) prior to testing, participants were instructed to consume 500 mL of water one hour before the assessment (Osterberg et al. 2009), after which hydration status was evaluated through urine-specific gravity using a refractometer (Masters28M, Atago Company, Tokyo, Japan). Participants then completed the Horne–Ostberg Morningness – Eveningness questionnaire (Horne and Ostberg 1976) to assess their circadian phenotype.

Subsequently, the players engaged in a standardized 15-minute warm-up composed of continuous running, lower and upper body strength exercises, and specific volleyball exercises (Pérez-López and Valades 2013).

Following the warm-up, participants underwent a neuromuscular test battery, including three vertical jumps (i.e. squat jump, countermovement jump, and spike jump), spike (i.e. standing spike and jumping spike tests), agility performance (i.e. modified agility T-test), isometric handgrip strength and dynamic balance tests (i.e. modified star excursion balance test). The reliability of these measurements was assessed with the coefficient of variation (CV) ranging from 1.6–10%. Rating of perceived exertion (RPE) was recorded within 30 minutes after the completion of the neuromuscular test battery, following the methodology previously described by Foster et al. (Foster et al. 2001) (Figure 1).

Measures

Vertical Jump Test

Vertical jump height was assessed for each jump using an infrared system (Optojump, Microgate, Bolzano, Italy) following standardized procedures (Bosco et al. 1983). Players performed three types of vertical jumps in the following sequence: squat jump, countermovement jump without arm swing, and spike jump with arm swing. During the squat jump and the countermovement jump without arm swing, players kept their hands on their hips. In contrast, the spike jump involved an

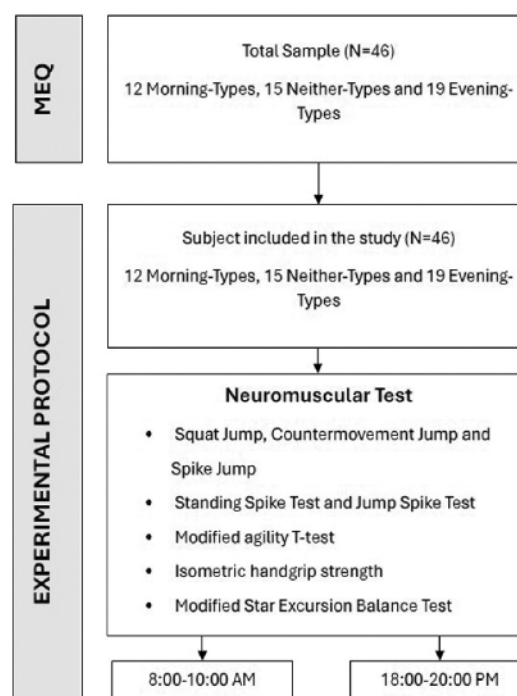


Figure 1. Study protocol.

approach and arm swing to generate momentum and maximize jump height. Each player performed two maximal attempts for each jump type, with 45 seconds of passive recovery between attempts. Players were allowed to choose a comfortable squat depth for all jumps, while a 5-second pause was required before initiating the squat jump to prevent any countermovement (Gaos et al. 2024). The highest recorded jump height (cm) for each jump type was used for analysis.

Spike Tests

Two volleyball-specific ball-hitting tests were conducted. The standing spike test followed a previously established protocol (Palao and Valades 2012). In this test, players self-tossed the ball and then spiked it with maximum force toward a designated target zone (1.5 m × 1.5 m), ensuring their feet lost contact with the ground. The jumping spike test required a coach or an experienced competitive volleyball player to toss the ball 2–3 meters into the air, allowing the player to jump and then spike the ball with maximum force toward the target zone and over the net. A researcher positioned laterally to the players monitored the execution of both tests, ensuring proper ball tossing and spike execution. The test was repeated if any aspect was performed incorrectly, or the fixed positioning was violated. Each player completed three attempts per test, with a 30-second passive standing rest between trials. The same ball (Molten V5M5000) was used throughout all trials, and peak ball velocity ($\text{km}\cdot\text{h}^{-1}$) was measured using a Pocket Radar (Model PR1000-BC, Inc., Santa Rosa, California) and used for analysis in each test (Valades et al. 2007).

Agility Performance Test

The modified agility T-test required players to complete a multidirectional movement circuit (i.e. forward, lateral in both directions, and backward) to simulate explosive actions commonly performed in volleyball (e.g. digs or receptions) (Sassi et al. 2009). Players started in a split stance, with the toes of their lead foot positioned 1 m behind the starting point to prevent inadvertent timing activation. They sprinted 5 m forward to touch the base of cone A with their right hand, then shuffled left (while facing forward and without crossing their feet) to touch the base of cone B with their left hand. Next, they shuffled 5 m to the right to touch the base of cone C with their right hand, shuffled 2.5 m back to the left to touch cone A with their left hand, and finally backpedaled to the finish line (same as

the starting point). Time to complete the test was recorded using single-beam electronic timing sensors (Microgate, Polifemo Radio Light, Italy) set 1 m above the ground and positioned 3 m apart at the starting line. Two trials were performed, separated by a 2-minute passive standing recovery, with the fastest time used for subsequent analysis.

Isometric Handgrip Strength

The isometric handgrip strength test was performed with 0° of shoulder and elbow flexion, while the forearm and hand remained in a neutral position, following previously established protocols (López-Samanes et al. 2017). Two maximal isometric voluntary contractions were performed for both the dominant and non-dominant hand using a calibrated handgrip dynamometer (Takei 5101, Tokyo, Japan). The highest peak force (N) recorded for each hand was used for subsequent analysis.

Dynamic Balance (Modified Star Excursion Balance Test)

Dynamic balance was evaluated using the OctoBalance device (Check your Motion, Albacete, Spain), applied with a modified version of the Star Excursion Balance (SEBT) (Gonzalo-Skok et al. 2015). The measurement system involved an extended measuring tape, magnetized in each direction to an octagon-shaped platform. Each trial consisted of pushing the marked point, situated at the top of the measuring tape as far as possible with the toes in the designated direction (anterior, posteromedial, and posterolateral) (Gonzalo-Skok et al. 2015). The maximal reach distance (cm) was measured by marking the tape measure with erasable ink at the point where the most distal part of the foot reached. Players completed three trials in each of the anterior, posteromedial, and posterolateral directions with the dominant and non-dominant limbs separated by 60 s of passive standing rest. The greatest distance reached was recorded for subsequent analysis in each direction for each limb.

Statistical Analysis

Sample size was calculated using G*Power (version 3.1.9.2; University of Düsseldorf; Düsseldorf, Germany). Previous studies in this population (Martín-López et al. 2022) revealed that fifteen participants are sufficient to detect time-of-day differences with an effect size of 0.4–0.6 ($\alpha = 0.05$ and

$1-\beta=0.8$). However, in the present study circadian phenotype was also considered; therefore, the required sample size was larger. Based on an effect size of 0.4 ($\alpha=0.05$ and $1-\beta=0.95$) a total of 42 participants were estimated to be necessary, assuming a 5% dropout (≈ 2 participants per chronotype group). Ultimately, 46 athletes were recruited, and all completed the study.

Descriptive data are presented as mean and standard deviation (SD). Normal distribution of data was evaluated using the Shapiro – Wilk test, while homogeneity of variances was assessed with Levene's test. For repeated-measures analyses, the assumption of sphericity was verified using Mauchly's test, and Greenhouse – Geisser corrections were applied when it was violated. Non-normally distributed variables were logarithmically transformed (e.g. isometric handgrip, T-test, and Modified Star Excursion Balance Test). Then, repeated measures ANOVA with two factors circadian phenotype (i.e. morning-type vs intermediate-type vs evening-type) and time-of-day (i.e. morning vs evening) was used to compare the effect of circadian phenotype on neuromuscular performance in the morning and evening conditions. Bonferroni correction was used as a *post hoc* test when significant differences were detected. Additionally, a chi-square (χ^2) test was performed to analyze differences in phenotype percentage distributions and differences between trials on tympanic temperature, urinary specific gravity and rate of perceived exertion were evaluated using a Student's t-test. Partial eta-squared (η_p^2) was calculated to measure the effect size in the ANOVA, with the following ranges: small (0.01), medium (0.06), and large (0.14) (Cohen 1988); while Cohen's d (d) was used in pairwise comparison based on the following criteria: trivial (0.019), small (0.20–0.49), medium (0.50–0.79) and large (0.80 and greater) (Cohen 1992). The level of significance was set at $p < 0.05$. All the statistical analyses were done using the SPSS software version 26 (SPSS Inc., Chicago, IL, USA).

Results

Tympanic Temperature, Urinary Specific Gravity, and Rate of Perceived Exertion

During the morning trials, tympanic temperature was $36.1 \pm 0.3^\circ\text{C}$, while in the evening trials, it increased significantly to $36.5 \pm 0.4^\circ\text{C}$ ($p < 0.001$; $d =$

0.84). Hydration status, assessed by urinary specific gravity, was 1.020 ± 6 in the morning and 1.019 ± 8 in the evening ($p = 0.051$; $d = 0.29$) and the RPE remained similar (3.91 ± 1.52 vs. 4.12 ± 1.52 points; $p = 0.458$; $d = 0.11$) when comparing morning and evening measurements.

Morningness – Eveningness Questionnaire (MEQ)

Based on the phenotype classification using the Morningness-Eveningness Questionnaire (Horne and Ostberg 1976), twelve participants (26.1%) were identified as morning-type, fifteen participants (32.6%) as intermediate-type, and nineteen participants (41.3%) as evening-type. However, no significant differences were observed between groups ($p = 0.447$; $\eta_p^2 = 0.043$).

Effect of Circadian Phenotype and Time-of-Day on Neuromuscular Performance

Differences in neuromuscular performance according to circadian phenotype and time-of-day are shown in Table 1. Time-of-day differences were found in countermovement jump, squat jump, spike jump, standing spike test and isometric handgrip strength with the non-dominant hand ($p < 0.05$, $\eta_p^2 = 0.09$ – 0.41) but not in the jumping spike test, modified T-test or handgrip strength with the dominant hand ($p = 0.117$ – 0.429 ; $\eta_p^2 = 0.02$ – 0.06). Moreover, no neuromuscular performance variables revealed circadian phenotype plus time-of-day interaction ($p > 0.05$; $\eta_p^2 = 0.01$ – 0.14).

When comparing morning vs. evening trials, in the squat jump, differences were found in the morning (1.8%, $p = 0.019$, $d = 0.93$), intermediate (2.6%, $p < 0.001$, $d = 1.16$) and evening circadian phenotypes (2.0%, $p = 0.002$, $d = 0.62$) in the evening compared to the morning trial. In the countermovement jump, higher performance levels were found in the evening trial in participants with intermediate (2.0%, $p = 0.001$, $d = 0.88$) and evening circadian phenotype (1.9%, $p < 0.001$, $d = 0.90$), but not in morning phenotype (0.5%, $p = 0.425$, $d = 0.25$). Similarly, in spike jump, higher performance in the evening was found in the intermediate (2.5%, $p = 0.011$, $d = 1.01$) and evening circadian phenotype participants (3.0%, $p = 0.001$, $d = 0.67$) but not in participants with morning phenotype (1.1%, $p = 0.288$, $d = 0.38$). Besides, in the standing spike test, improvements in evening compared to morning trial performance were found in the evening

Table 1. Differences in circadian phenotype and time-of-day in neuromuscular performance (means and standard deviations).

	Circadian phenotype	Morning	Evening	Time-of-day effect		Time-of-day × circadian phenotype interaction	
				<i>P</i> value	η_p^2	<i>P</i> value	η_p^2
Countermovement jump (cm)	Morning	41.60 ± 9.36	42.09 ± 9.53	<0.001	0.33	0.133	0.09
	Intermediate	38.80 ± 6.19	40.85 ± 7.09*				
	Evening	42.17 ± 6.96	44.02 ± 7.57*				
Squat jump (cm)	Morning	37.61 ± 9.45	39.45 ± 9.50*	<0.001	0.41	0.736	0.01
	Intermediate	35.88 ± 6.79	38.44 ± 6.86*				
	Evening	39.22 ± 6.26	41.19 ± 7.78*				
Spike jump (cm)	Morning	55.00 ± 11.40	56.11 ± 11.83	<0.001	0.28	0.356	0.05
	Intermediate	51.17 ± 7.99	53.64 ± 8.91*				
	Evening	55.96 ± 10.08	59.00 ± 11.22*				
Standing spike test (km·h ⁻¹)	Morning	78.66 ± 10.35	80.83 ± 10.07	<0.001	0.14	0.498	0.03
	Intermediate	81.84 ± 9.31	82.66 ± 9.86				
	Evening	83.31 ± 11.12	86.26 ± 11.67*				
Jumping spike test (km·h ⁻¹)	Morning	78.25 ± 10.32	76.83 ± 8.79	0.332	0.02	0.256	0.06
	Intermediate	80.40 ± 11.40	82.13 ± 12.50				
	Evening	83.63 ± 10.11	86.31 ± 8.67				
Modified T-test (s)	Morning	5.73 ± 0.37	5.74 ± 0.22	0.429	0.02	0.488	0.03
	Intermediate	5.94 ± 0.24	5.92 ± 0.34				
	Evening	5.90 ± 0.39	6.04 ± 0.37				
Isometric handgrip strength DOM (N)	Morning	421.15 ± 67.87	437.40 ± 74.68	0.117	0.06	0.743	0.14
	Intermediate	467.52 ± 82.86	478.76 ± 79.62				
	Evening	475.19 ± 86.10	479.27 ± 76.60				
Isometric handgrip strength NO-DOM (N)	Morning	396.00 ± 76.40	418.13 ± 75.92	0.046	0.09	0.274	0.06
	Intermediate	438.06 ± 69.67	456.87 ± 75.76				
	Evening	448.06 ± 95.18	447.24 ± 79.44				

Values are means ± SD. Results derived from two-way repeated-measures ANOVA (time-of-day × phenotype). Abbreviations: **p* < 0.05 compared with morning session within the same chronotype. η_p^2 = partial eta squared; cm = centimetres; N = newtons; DOM = dominant; NO-DOM = non-dominant.

* significant differences (*P* ≤ .05).

Table 2. Differences in circadian phenotype and time-of-day in the Star Excursion Balance test performance (means and standard deviations).

	Circadian phenotype	Morning	Evening	Time-of-day effect		Time-of-day × circadian phenotype interaction	
				<i>P</i> value	η_p^2	<i>P</i> value	η_p^2
SEBT anterior DOM (cm)	Morning	61.25 ± 5.19	61.25 ± 4.41	0.646	0.01	0.417	0.04
	Intermediate	62.83 ± 5.60	62.33 ± 5.57				
	Evening	63.73 ± 7.42	65.13 ± 7.03				
SEBT anterior NO-DOM (cm)	Morning	62.37 ± 6.08	60.66 ± 4.42	0.840	0.01	0.218	0.07
	Intermediate	63.16 ± 4.06	63.16 ± 5.54				
	Evening	63.47 ± 8.32	64.76 ± 8.20				
SEBT posterolateral DOM (cm)	Morning	83.54 ± 9.85	82.79 ± 8.81	0.926	0.00	0.491	0.03
	Intermediate	80.93 ± 12.71	82.70 ± 12.06				
	Evening	84.60 ± 9.49	83.86 ± 7.32				
SEBT posterolateral NO-DOM (cm)	Morning	81.87 ± 8.61	80.66 ± 7.76	0.380	0.02	0.478	0.03
	Intermediate	80.20 ± 8.57	80.83 ± 8.84				
	Evening	84.86 ± 11.00	83.23 ± 6.98				
SEBT posteromedial DOM (cm)	Morning	79.45 ± 9.68	77.04 ± 6.59	0.096	0.06	0.447	0.04
	Intermediate	78.83 ± 13.43	78.90 ± 11.95				
	Evening	81.44 ± 10.25	78.84 ± 8.22				
SEBT posteromedial NO-DOM (cm)	Morning	81.54 ± 8.80	77.29 ± 8.02	0.080	0.07	0.224	0.07
	Intermediate	80.43 ± 12.91	81.26 ± 8.40				
	Evening	82.31 ± 10.64	79.15 ± 8.64				

Values are means ± SD. Results derived from two-way repeated-measures ANOVA (time-of-day × phenotype).

Abbreviations: **p* < 0.05 compared with morning session within the same chronotype. η_p^2 = partial eta squared; cm = centimetres; N = newtons; DOM = dominant; NO-DOM = non-dominant.

phenotype (3.0%, *p* = 0.036, *d* = 0.47), but not in the morning (3.1%, *p* = 0.071, *d* = 0.45) or the intermediate circadian phenotype (0.8%, *p* = 0.595, *d* = 0.19).

Differences in Star Excursion Balance test performance are shown in Table 2. No time-of-day (*p* = 0.080–0.926, η_p^2 = 0.01–0.07) or time-of-day plus circadian phenotype interaction (*p* = 0.218–0.491, η_p^2 = 0.03–0.08).

Discussion

The principal aim of this study was to examine the relationship between circadian phenotype and time-of-day on neuromuscular performance in male volleyball players. Our findings revealed that although evening performance is improved in vertical jump (e.g. squat jump, countermovement jump or spike jump), spike ball speed (e.g. standing spike test) or isometric strength (e.g. handgrip in the non-dominant hand) compared to morning values, circadian phenotype showed limited (e.g. countermovement and spike jumps) or no effect (e.g. remaining test) on neuromuscular performances. Therefore, these results suggest that diurnal variations, rather than circadian phenotype, may play a critical role in optimizing neuromuscular performance in male volleyball players.

The chronobiological approach to sports and physical performances has gained significant importance in recent years, with a notable increase in scientific studies (Wang and Xia 2023) analysing how circadian rhythms (i.e. diurnal variations) impact athletic performance, training adaptations or recovery, particularly in team sport athletes (Reilly et al. 2007; Mhenni et al. 2017; Martín-López et al. 2022; Gaos et al. 2024). However, the existing evidence regarding the influence of athletes' circadian phenotype on diurnal variation on sport performance remained scarcely studied. Only a few studies conducted in soccer (Roveda et al. 2020), rowing (Brown et al. 2008) or swimming (Rae et al. 2015) have explored this idea. These studies used the morningness-eveningness questionnaire, except in one case where the PER3 genotype was also included (Rae et al. 2015), and revealed that morning-type phenotypes perform better in the morning compared to the evening than intermediate- and evening-type athletes. In contrast, evening performance was higher in evening-type compared to morning- and intermediate-type athletes. Also, intermediate-type athletes often exhibit more stable performance across different times of the day. However, despite these previous shreds of evidence, our study failed to find such a relationship between circadian phenotype and time of day on neuromuscular performance. In fact, our findings on vertical jump capacity and agility performance contradict those reported on adolescent soccer players' performance in vertical jump capacity (i.e. Sargent jump test) or agility performance (i.e. Illinois agility test) (Roveda et al. 2020). Chronotype is modified with age,

particularly in males (Roenneberg et al. 2003; Fischer et al. 2017); thus, the differences observed in neuromuscular performance in both studies may be caused by the transition between chronotypes experimented with age. Moreover, the battery of neuromuscular performance tests differs between studies, particularly in selected vertical jump capacity (i.e. Sargeant vs Spike jump or countermovement jump) and agility performance test (i.e. Illinois agility test vs modified agility T-test) or even the electronic devices employed (i.e. handheld stopwatches vs photoelectric device) could be behind the difference observed between studies. Therefore, Brown et al. (2008) analyzed differences in the exercise type (e.g. endurance vs neuromuscular performance), showed that participants' age can be one of the reasons that explain the opposing results shown compared to our study since they recruited eight male and eight female collegiate rowing participants (e.g. 19.6 ± 1.5 vs 27 ± 8 y), another reason could be the potential sex difference existing in the diurnal variation on performance (Spitschan et al. 2022). Finally, Rae et al. (2015) recruited twenty-six swimmers (18 males and 8 females of 32.6 ± 5.7 y), despite the differences in exercise type, found that habitual training schedule (e.g. morning or evening) is another variable that could mask the influence of chronotype on diurnal variation. This idea is in line with our results since all the male athletes recruited scheduled their volleyball training in the evening, and it was in the evening trial where all chronotypes (e.g. morning-type, intermediate-type, and evening-type) reported higher performance in vertical jumps capacity, spike ball velocity or agility tests. Therefore, future chronotype and time-of-day-focused studies should consider collecting data about the habitual training schedules of their participants.

Furthermore, another study evaluated the interaction between chronotype and time-of-day in isometric strength. Küüsmaa et al. (2015) observed higher evening strength in evening types for lower limb performance (e.g. isometric leg press and knee extension). In contrast, we found higher evening handgrip strength performance in morning-type and intermediate-type athletes in both dominant and non-dominant hands. While evening-type athletes reported a less pronounced evening increase in the dominant hand and even a slight morning increase in the non-dominant hand. These differences may stem from variations in neuromuscular regulation between upper and lower limbs, the distinct circadian modulation of muscle function or differences in the studied populations (trained athletes vs.

untrained individuals). However, isometric strength (e.g. handgrip) is not a critical variable in volleyball performance and, consequently, is not habitually trained by these athletes, which may alter the potential influence of chronotype and time-of-day on this variable.

When considering the time-of-day effects on performance, our results were consistent with previous research indicating that peak muscle strength and endurance typically occur in the early evening (Reilly et al. 2007; Heishman et al. 2017; Pavlovic et al. 2018). This suggests that, while phenotype may play a role in some specific tasks or sports, the influence of time-of-day on performance may be a more applicable factor, especially in tasks demanding sport-specific strength and endurance performance. The analysis of performance changes across different times of day revealed several key findings. Overall, significant improvements were observed in afternoon performance, particularly for intermediate and evening chronotypes. For vertical jump tests, including the countermovement jump, squat jump and spike jump, evening performance was notably enhanced in intermediate-type and evening-type athletes, while morning-type showed less pronounced improvement. These differences were likely driven by the diurnal variations in core and muscle temperature, nervous system activation, and coordination (Kilduff et al. 2013; Heishman et al. 2017; Cherif et al. 2022), which may be critical for these neuromuscular performance tests. However, this was not found in spike ball velocity, potentially due to the difference in the type of movement (e.g. single- vs. multi-joint involvement), muscle groups involved or the use of force (e.g. ground reaction force in jumps and kinetic chain in spikes).

Besides, no significant time-of-day effects were found for agility, with all chronotypes performing similarly in both morning and afternoon trials. These findings align with studies on other team sports, such as basketball (Gaos et al. 2024) and suggest that the time of day may have minimal influence on agility and strength/power muscle performance, as seen in other research on handball and volleyball (Gharbi et al. 2013; Martín-López et al. 2022). Similarly, no substantial time-of-day differences were observed in dynamic balance performance, with only a minor improvement seen in the posteromedial direction for the non-dominant limb in the evening session. These results are consistent with findings in dynamic balance research, which suggest that the time-of-day has a limited impact on tasks requiring controlled balance (Heinbaugh et al. 2015; Martín-López et al. 2022).

Limitations

The study has several limitations that should be considered when interpreting the results obtained. i) data were collected exclusively from male volleyball players; therefore, it would be interesting to determine whether circadian phenotype also has no influence on neuromuscular performance in females. ii) the sample size according to chronotype was small; thus, future studies with larger samples should be conducted to explore its influence on time-of-day performance. iii) Data were collected from a specific sport and a particular ethnic group; therefore, these findings cannot be generalized to other sports disciplines or populations. iv) chronotype was classified using a questionnaire instead of genotyping participants for the VNTR polymorphism within the PER3 gene (Archer et al. 2003; Kunorozva et al. 2012). v) Diet, sleep, and stimulant intake were based solely on self-report; future research should apply more objective monitoring methods. vi) the athletes' habitual evening training schedule may have attenuated potential chronotype-related differences, as regular exposure to training at a fixed time of day can induce partial adaptation of physiological and performance rhythms.

Conclusion

In conclusion, evening performance is enhanced in vertical jump tests (e.g. squat jump, countermovement jump, and spike jump), spike ball speed (e.g. standing spike test), and isometric strength (e.g. handgrip in the non-dominant hand) compared to morning values. However, the influence of circadian phenotype on neuromuscular performance was limited (e.g. countermovement and spike jumps) or negligible (e.g. other tests). These findings indicate that diurnal variations, rather than circadian phenotype, play a more critical role in optimizing neuromuscular performance in male volleyball players. Future research should investigate whether these results can be generalized to other team sports, to athletes of different sexes or ethnic backgrounds, and to individuals with distinct morning- or evening-oriented training habits.

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Disclosure Statement

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3.4 Estudio 4: Sex differences in the effects of diurnal variation on neuromuscular performance in volleyball players

ESTUDIO 4

En revisión:

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Sex-specific effects of diurnal variation on neuromuscular performance in volleyball players

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Keywords:	exercise physiology, strength training

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3 ABSTRACT

4 **Purpose:** Neuromuscular performance is influenced by diurnal variation, but it remains
5 unclear whether sex moderates these patterns in team sport athletes. This study examined
6 whether sex moderates the effect of time of day on neuromuscular performance in
7 volleyball players. **Methods:** One hundred and two volleyball players (46 males, 56
8 females) completed a randomized battery of tests in the morning (08:00–10:00) and
9 evening (18:00–20:00) on separate days. The battery included vertical jumps (squat,
10 countermovement, and spike jumps), spike performance (standing and jumping spike),
11 agility (modified T-test), isometric handgrip strength, and dynamic balance (modified star
12 excursion balance test). **Results:** Specifically, males improved their squat jump
13 performance in the evening ($p=0.002$, $\eta^2=0.09$) and showed higher ball velocity in the
14 jumping spike ($p=0.045$, $\eta^2=0.04$). Females tended to improve agility in the evening
15 ($p=0.027$, $\eta^2=0.05$) and demonstrated better posterolateral balance on the dominant leg
16 ($p=0.005$, $d=0.26$), while males showed no change ($p=0.799$). No significant interactions
17 were observed in other balance directions ($p>0.05$). A significant time-of-day effect was
18 observed, indicating higher evening performance in all jump tests ($p = 0.005$, $\eta^2 = 0.08$ –
19 0.19), ball velocity in spike velocity ($p = 0.002$, $\eta^2 = 0.09$), and isometric handgrip
20 strength ($p = 0.001$, $\eta^2 \approx 0.10$). **Conclusion:** Sex-specific differences were evident only
21 in selected tasks: males tended to show greater improvements in explosive actions, while
22 females showed more stable or agility-related benefits. Nonetheless, the predominant
23 effect was a general improvement in neuromuscular performance in the evening,
24 independent of sex.

25

26 **Keywords:** Circadian rhythms, muscular and strength power output, team athletes,
27 diurnal variation.

28 Sex differences are well established in human physiology, however, most scientific
29 research, particularly in sport and exercise science, has historically focused on male
30 participants ¹ This underrepresentation of females has limited our understanding of sex-
31 specific responses to exercise, although recent work has highlighted a growing scientific
32 and practical interest in identifying the physiological mechanisms underlying potential
33 sex differences ². Males and females differ across multiple physiological systems while
34 males typically display greater lean mass and a higher proportion of type II muscle fibres
35 comparing females^{1,2}; hormonal profiles also diverge, with elevated testosterone
36 concentrations in males versus cyclical fluctuations of estrogen and progesterone in
37 females ³; while at the neural level, females often exhibit higher motor unit discharge
38 rates and recruitment thresholds ⁴. These sex-related differences contribute to variations
39 in human motor performance, influencing muscle strength and power production ² and
40 fatigue resistance ^{5,6}. Importantly, such differences are not solely determined by sex but
41 may also be modulated by external factors, including environmental conditions ^{7,8} and
42 circadian rhythms (i.e., time-of-day) ⁷. The interplay between biological and contextual
43 determinants highlights the complexity of interpreting performance outcomes and
44 emphasizes the need for integrative approaches.

45 Circadian rhythms are endogenous biological oscillations that follow an approximately
46 24-hour cycle, governed by the suprachiasmatic nuclei of the hypothalamus ⁹. They
47 influence a wide range of physiological processes, including core body temperature ¹⁰ or
48 muscle contractile properties and neuromuscular excitability ¹¹. Neuromuscular
49 performance in explosive tasks, such as jumping and sprinting, typically peaks in the late
50 afternoon or early evening (16:00 – 20:00 h) and declines in the early morning (06:00 –
51 10:00 h), with performance differences ranging from 5% to 15% in measures such as
52 vertical jump height and maximal voluntary contraction ^{12,13}. Although circadian

variations in neuromuscular performance are well documented¹⁴, most studies have focused exclusively on males, limiting the generalizability of findings. This is a critical oversight, as sex may modulate time-of-day effects on sport performance^{15,16}. To address this gap in the literature, the present study examines whether sex modulates the effect of time-of-day on neuromuscular performance in team sports athletes (i.e., volleyball players). We hypothesize that neuromuscular performance will be higher in the evening compared to the morning in both sexes; however, the magnitude of these fluctuations is expected to differ, with males showing greater diurnal variability and females maintaining more stable performance throughout the day.

Methods

Participants

One hundred and two volleyball players, 46 male (age: 26 ± 7 years; height: 183 ± 7 cm; body mass: 81.4 ± 11.7 kg; body mass index: 23.9 ± 2.7 ; volleyball experience: 12 ± 7 years; training per week 6 ± 3 hours) and 56 female (age: 23 ± 5 years; height: 171 ± 6 cm; body mass: 65.5 ± 7.5 kg; body mass index: 22.3 ± 2.2 ; volleyball experience: 12 ± 5 years; training per week 8 ± 4 hours) participated in this study. All participants regularly competed and trained in both morning (~09:00 hours) and evening (~19:00 hours) sessions. Exclusion criteria included: (a) any upper- or lower-limb injury within the 3 months before testing, (b) presence of upper or lower-limb muscle soreness at the time of testing, (c) use of medications that could affect neuromuscular performance and (d) consumption of stimulants such as caffeine or tea within the 48 hours before each trial. All participants were informed of the study procedures, and written informed consent was obtained before their inclusion. For minors (<18 years), parental or guardian consent was secured. The study received ethical approval from the University Bioethics Commission (13/2021) in accordance with the principles outlined in the Declaration of Helsinki.

78 ***Experimental design***

79 A randomized crossover design was used. Each participant completed a neuromuscular
80 test battery at two distinct times of day: morning (08:00–10:00) and evening (18:00–
81 20:00), on non-consecutive days separated by a 48-hour interval. The testing order was
82 randomly assigned to minimize potential bias, with participants serving as their own
83 controls for within-participants comparisons. All sessions were performed in the same
84 indoor sports facility, where participants routinely train and compete, under controlled
85 conditions. Average ambient temperature was $18.4 \pm 5.2^{\circ}\text{C}$, and relative humidity was
86 $45.7 \pm 11.7\%$, measured with a portable meteorological station (Meteorological Station,
87 Küken, Spain). To minimize variability, participants wore same athletic footwear during
88 both testing days. They were instructed to follow a consistent sleep schedule (7-9 hours
89 per night) before each testing session to avoid strenuous exercise for 48 hours prior to
90 testing, and to abstain from caffeine, alcohol intake, and other stimulants during the same
91 period¹⁷.

92 ***Experimental protocol***

93 Body composition was assessed using bioelectrical impedance analysis (Tanita 901,
94 Tanita Corp., Tokyo, Japan). Tympanic temperature was measured in triplicate with a
95 digital thermometer (Thermoscan 6520, Braun, Germany). To ensure euhydration (USG
96 < 1.020), participants drank 500 mL of water one hour before testing. Hydration status
97 was confirmed using urine-specific gravity measured with a refractometer (Masters28M,
98 Atago Company, Tokyo, Japan). Participants completed a standardized 15-minute warm-
99 up consisting of continuous running, upper- and lower-body strength exercises, and
100 volleyball-specific drills¹⁷. They then performed a neuromuscular test battery including
101 vertical jump tests (i.e., squat jump, countermovement jump and spike jump), spike
102 performance test (i.e., standing spike and jumping spike), agility performance (i.e.,

103 modified agility T-test), isometric handgrip strength and dynamic balance tests (i.e.,
104 modified star excursion balance test). The reliability of these measurements was
105 confirmed, with coefficient of variation (CV) ranging from 1.6% to 10% depending on
106 specific test. Rating of perceived exertion (RPE) was recorded within 30 minutes after
107 the test battery using a 1-10 scale.

108 ***Vertical jump tests***

109 Players performed three maximal attempts for each of vertical jump tests (squat jump,
110 countermovement jump and spike jump) with 45 seconds of passive recovery between
111 attempts. The squat jump was performed from static squat at $\sim 90^\circ$ knee flexion position,
112 following a 3-second static hold to ensure compliance and then performed a maximal
113 vertical jump. All jumps were executed without countermovement prior to take-off. The
114 countermovement jump was initiated from standing position, with equal body mass
115 distributed over both feet. Participants descended to $\sim 90^\circ$ knee flexion, and then
116 performed a maximal vertical jump, landing in the same position as at take-off¹⁸. The
117 spike jump test consisted of a vertical-specific vertical jump without the ball, involving
118 three to four approach steps and a two-foot take off before performing a maximal vertical
119 jump using arm swing. In all test, jump height was measured with an infrared system
120 (Optojump, Microgate, Bolzano, Italy). Additional trials were performed until two
121 consecutive jumps differed by $<10\%$. The best trial was retained. Previous studies have
122 reported CV values of 2.1–2.8% for these tests in national-level players¹⁹.

123 ***Spike performance tests***

124 Two volleyball-specific ball hitting tests were performed. In the standing spike test,
125 participants self-tossed the ball and spiked it with maximal force toward a designated
126 target zone (1.5×1.5 m) while maintaining constant foot contact with the floor²⁰. In the
127 jumping spike test, a coach or experienced volleyball player tossed the ball 2–3 meters

high, then participants performed a standardized three- to four-step approach before jumping and spiking the ball over the net with maximal force toward the target zone. A researcher was positioned laterally during both tests to monitor proper execution of the spike (and toss) in a fixed position; if any error occurred, the attempt was repeated. Each participant performed three attempts for each test, with 30 seconds of passive recovery between attempts. The same volleyball (Molten V5M5000, Molten, Thailand) was used in all trials. Peak ball velocity ($\text{km} \cdot \text{h}^{-1}$) was recorded with a Pocket Radar device (Model PR1000-BC, Inc., Santa Rosa, California), which has demonstrated excellent reliability ($\text{ICC} = 0.980$)²¹. The best velocity per test was analysed. Reported CV for these tests is 4%²⁰.

Agility performance test

Agility and change-of-direction speed were assessed using the modified T-test, adapted to mimic volleyball-specific movements such as diving and lateral coverage²². The modified agility t-test required players to execute a multidirectional movement circuit (i.e., forwards, laterally in both directions, and backwards) to simulate explosive actions performed in volleyball (e.g., digs, receptions)²². More precisely, players began the test in a split stance with the toes of their leading foot placed 1 m behind the starting point to avoid inadvertent triggering of timing. Players sprinted forward for 5 m and touched the base of a cone (A) with the right hand, before shuffling (facing forward without crossing the feet) to the left to touch the base of another cone (B) with the left hand. Players then shuffled 5 m to the right to touch the base of another cone (C) with the right hand, before shuffling 2.5 m to the left to touch the base of cone A with the left hand, and then backpedalled to the finish line (which is the same as the starting point). Performance time was measured using single-beam timing sensors (Microgate, Polifemo Radio Light,

Italy), positioned 1 m high and 3 m apart. Two trials were completed with 2 minutes of recovery; the fastest time was retained. Reported CV is 2.6%²².

Isometric handgrip strength

Isometric handgrip strength was measured with a Takei 5101 calibrated dynamometer (Takei, Tokyo, Japan). Participants performed the test in a standardized standing posture, with the shoulder in neutral position and the elbow fully extended (0° flexion) and the forearm and hand in a neutral grip position²³. Each hand (dominant and non-dominant) performed two maximal voluntary contractions, with 30 seconds of rest between attempts. Peak force (N) was recorded and analysed. Reported CV is 1.6%²³.

Dynamic balance (Modified Star Excursion Balance Test).

Dynamic balance was assessed using a modified version of the Star Excursion Balance Test (SEBT), with the OctoBalance system (Check your Motion, Albacete, Spain). The system consists of magnetized measuring tapes radiating from a central platform, aligned with three reach directions: anterior, posteromedial, and posterolateral²⁴. Participants were instructed to maintain a stable base of support on the testing leg while reaching as far as possible in the designated direction with the contralateral foot, pushing the sliding indicator along the tape to record their maximum reach distance. Each leg (dominant and non-dominant) was assessed through three attempts per direction, with a 60-second rest between trials. The best reach distance (cm) per direction was used for analysis. Reported CV ranges from 2.5–3.3% in team-sport athletes²⁴

Statistical analysis

An a priori power analysis was conducted using G*Power software (version 3.1.9.2; University of Düsseldorf, Düsseldorf, Germany) to determine the sample size required to detect a moderate sex by time- of- day interaction in a two-way mixed-design repeated-measures analysis of variance (ANOVA). The analysis assumed an effect size of 0.25, an

alpha error of 0.05, and a desired statistical power of 0.90. The calculation indicated that a minimum total sample size of 46 participants (23 females and 23 males) would be required to detect the specified interaction effect with the desired statistical power. Data are presented as mean $\pm$ standard deviation (SD). Normality of all variables was assessed using the Shapiro–Wilk test. As most variables did not meet normality assumptions, performance data were log-transformed before analysis. Then, a two-way repeated-measures ANOVA was conducted for each dependent variable, with time-of-day (morning vs. evening) as the within-subject factor and sex (male vs. female) as the between-subject factor. Bonferroni-adjusted post hoc comparisons were applied to identify within- and between-group differences. Effect sizes were calculated using partial eta-squared (η_p^2) for the ANOVA, with values interpreted as small (0.01), medium (0.06), and large (0.14), and Cohen's d for pairwise comparisons, categorized as trivial (< 0.20), small (0.20–0.49), medium (0.50–0.79), and large (≥ 0.80) (Cohen, 1988). Statistical significance was set at $p < 0.05$. All analyses were conducted using IBM SPSS Statistics for Windows, version 26.0 (IBM Corp., Armonk, NY, USA).

Results

Tympanic temperature, urinary specific gravity and rate of perceived exertion.

No significant sex by time-of-day interaction was found for tympanic temperature or urine specific gravity (USG) ($p > 0.05$, $\eta_p^2 < 0.00$ –0.02). However, a significant main effect of time-of-day was detected for both variables ($p < 0.001$, $\eta_p^2 = 0.11$ –0.43). Specifically, males and females exhibited significantly higher tympanic temperatures in the evening ($p < 0.001$, $d = 0.80$ –0.93). For USG, males showed a significant decrease from morning to evening ($p < 0.001$, $d = 0.53$), whereas the reduction in females did not reach statistical significance ($p = 0.071$, $d = 0.25$). In contrast, for RPE, neither a

201 significant sex by time-of-day interaction ($p = 0.096$, $\eta_p^2 = 0.03$) or a main effect of time-
202 of-day ($p = 0.429$, $\eta_p^2 = 0.01$) was observed.

203 *Vertical jump test*

204 Table 1 shows differences in sex and time-of-day in vertical jump performance. In squat
205 jump, a significant sex by time-of-day interaction ($p = 0.002$, $\eta_p^2 = 0.09$) and time-of-day
206 effect ($p < 0.001$, $\eta_p^2 = 0.19$) were observed in the squat jump. Male participants showed
207 a significant improvement in the evening (5.9%, $p < 0.001$, $d = 0.76$), whereas females
208 did not (1.5%, $p = 0.236$, $d = 0.25$). In the countermovement jump, only time-of-day
209 effect was found ($p = 0.005$, $\eta_p^2 = 0.08$). Males showed significant evening improvements
210 (3.8%, $p = 0.002$, $d = 0.69$) an effect not shown in females (1.2%, $p = 0.403$, $d = 0.10$).
211 Similarly, in the spike jump time-of-day effect was noted ($p < 0.001$, $\eta_p^2 = 0.19$), finding
212 significantly higher performance in the evening in both males (4.4%, $p < 0.001$, $d = 0.64$)
213 and females (3.0%, $p = 0.007$, $d = 0.22$).

214 *Spike performance tests*

215 Table 1 shows differences in sex and time-of-day in spike performance test. In the
216 standing spike test, a time-of-day effect was found ($p = 0.002$, $\eta_p^2 = 0.09$), observing
217 significantly higher ball velocities in males (3.1%, $p = 0.007$, $d = 0.38$), whereas females
218 demonstrated a non-significant improvement (1.6%, $p = 0.080$, $d = 0.22$). In the jumping
219 spike test, sex by time-of-day interaction was found ($p = 0.045$, $\eta_p^2 = 0.04$). Male
220 participants exhibited non-significant improvement in evening trials (2.0%, $P = 0.257$, d
221 $= 0.53$), whereas female participants showed a slight non-significant decrease (-1.5%, p
222 $= 0.082$, $d = 0.27$).

223 *Agility performance test*

224 Table 1 shows differences in sex and time-of-day in agility T-test. A significant sex by
225 time-of-day interaction was observed in the modified agility T-test ($p = 0.027$, $\eta_p^2 = 0.05$).

Male participants showed a non-significant decline in evening performance (-1.2% , $p = 0.113$, $d = 0.68$), whereas females exhibited a non-significant improvement (1.1% , $p = 0.119$, $d = 0.33$).

Isometric handgrip strength

Table 1 shows differences in sex and time-of-day in isometric handgrip strength. A non-significant sex by time-of-day interaction was found for isometric handgrip strength in both the dominant ($p = 0.413$, $\eta_p^2 = 0.007$) and non-dominant hand ($p = 0.560$, $\eta_p^2 = 0.003$). However, time-of-day effect was found in both the dominant ($p = 0.001$, $\eta_p^2 = 0.10$) and non-dominant hands ($p = 0.001$, $\eta_p^2 = 0.10$). Females showed a significant improvement in the evening in both the dominant (4.47% , $p = 0.003$, $d = 0.42$) and non-dominant hands (4.33% , $P = 0.005$, $d = 0.35$), while the improvements in males were not statistically significant in either the dominant (2.8% , $p = 0.096$, $d = 0.25$) or non-dominant hand (3.4% , $p = 0.070$, $d = 0.31$).

INSERT TABLE 1 AROUND HERE

Dynamic balance performance

Table 2 shows differences in sex and time-of-day in the SEBT. Significant sex by time-of-day interactions effect was only found in posterolateral balance on the dominant limb ($P = 0.036$, $\eta_p^2 = 0.043$), while no other interaction ($p > 0.05$, $\eta_p^2 = 0.01-0.04$) or time-of-day effect differences were obtained in the other variable ($p > 0.05$, $\eta_p^2 = 0.01-0.03$). Female athletes demonstrated a significant improvement in posterolateral balance on the dominant limb in the evening (3.4% , $p = 0.005$, $d = 0.26$), whereas male athletes showed a minimal, non-significant change in the same direction (0.60% , $p = 0.799$, $d = 0.57$).

INSERT TABLE 2 AROUND HERE

DISCUSSION

250 The aim of this study was to determine whether sex moderates the effect of time-of-day
251 on neuromuscular performance in volleyball players. The main findings showed
252 significant sex by time-of-day interactions in squat jump, jumping spike test, modified
253 agility T-test and in posterolateral balance on the dominant limb, but not in other vertical
254 jumps (countermovement and spike jumps), standing spike test, isometric handgrip
255 strength or any other balance test. Altogether indicates that diurnal performance
256 fluctuations were not uniform across sexes, which emphasize the importance of tailoring
257 training and competition strategies to both biological sex and time-of-day.

258 Vertical jumping underpins both offensive (e.g., spiking) and defensive (e.g., blocking)
259 actions in volleyball ¹⁹. The squat jump showed a significant sex $\times$ time-of-day
260 interaction, with males presenting a significant evening improvement (5.9%), whereas
261 females did not (1.5%). In contrast, the countermovement jump did not reveal a sex $\times$
262 time-of-day interaction. Instead, a main effect of time-of-day was observed, but this
263 improvement reached significance only in males (+3.8%), confirming previous evidence
264 that circadian variability in explosive tasks is more pronounced in men ^{25,26}. These results
265 are supported by sex-differences physiological and neural factors. Males typically show
266 a greater proportion of type II fibers, which are preferentially recruited during explosive
267 tasks and exhibit diurnal fluctuations in contractile velocity and power ²⁷⁻²⁹. Additionally,
268 men display higher corticospinal excitability and earlier recruitment of high-threshold
269 motor units, further amplifying time-of-day effects ^{4,11}. Conversely, females rely more on
270 type I fibres and distinct neuromuscular strategies, such as higher motor unit discharge
271 rates that support gradual, sustained activation patterns ^{4,29}. These features may buffer
272 performance against circadian modulation, explaining their relative stability.

273 Interestingly, both sexes improved similarly in the spike jump. The added contribution of
274 the arm swing, combined with the stretch-shortening cycle (SSC) also present in the

275 countermovement jump, likely reduced reliance on maximal muscle power while
276 emphasizing elastic energy storage and coordination. This mechanism may explain the
277 comparable time-of-day effect across sexes, despite differences in muscle mass and
278 hormonal rhythms. Overall, our results align with prior work showing enhanced evening
279 performance in vertical jumps and sprinting tasks ^{25,27}, reinforcing the notion that
280 explosive lower-limb performance peaks later in the day.

281 The spike, a decisive high-velocity striking action in volleyball showed sex-dependent
282 patterns. Males improved in the evening, whereas females showed no diurnal variation.
283 Comparable findings have been reported in upper-limb ballistic tasks ²⁵. The greater
284 evening gains in males may reflect increased electromyographic activity, neuromuscular
285 activation, and enhanced musculo-tendinous compliance associated with higher afternoon
286 body temperature and testosterone variability ⁷. Females, by contrast, maintain more
287 stable thermoregulatory and hormonal profiles, attenuating circadian oscillations⁹. The
288 lack of sex differences in the jump spike may be explained by its greater technical
289 complexity, which depends on skillful integration of lower- and upper-limb movement
290 ³⁰. The variability observed here likely reflects the heterogeneous technical proficiency
291 of our semi-professional sample, which may have masked subtle sex- or time-dependent
292 effects.

293 Volleyball players also need to execute swift lateral movements, and to make rapid
294 transitions between accelerations, decelerations, and backpedalling during
295 defensive/offensive play sequences. Thus, modified T-test was used to evaluate
296 participants' agility. A sex by time-of-day interaction was noted in this test, where males
297 showed slower agility times in the evening compared to the morning (+1.17%), whereas
298 females exhibited slightly faster times in the evening (−1.06%). Although changes did not
299 reach statistical significance, the opposite directions suggest that sex modulates how

300 circadian rhythms influence perceptual-motor tasks ¹⁶. This variability may be partly
301 explained by fluctuations in arousal or fatigue later in the day ^{9,31}. Females, however,
302 seemed to maintain or even slightly improve their agility performance, which could
303 reflect greater stability in perceptual-motor processing across the day ²⁹. When compared
304 to previous research, our findings diverge from studies reporting systematic evening
305 improvements in agility ^{9,27}. These findings reinforce the idea that agility, because of its
306 strong dependence on anticipatory control and decision-making, is less consistently
307 affected by circadian rhythms than purely explosive movements ^{25,26}.

308 Isometric handgrip strength test serves in volleyball as a quick, efficient, and reliable
309 proxy for overall upper body strength, with indirect applicability to key performance
310 skills (i.e., spiking, blocking, serving) ^{27,32}. Although no sex by time-of-day interaction,
311 time-of-day effect was found revealing that both sexes exhibited small improvements in
312 evening sessions, more evident in females, where both the dominant (4.5%) and non-
313 dominant hand (4.3%) reached statistical significance. In males, the improvements were
314 smaller and not statistically significant, with an increase of 2.8% in the dominant hand
315 and 3.4% in the non-dominant hand. These findings support prior evidence that isometric
316 strength tasks, relying on maximal force output rather than complex neuromuscular
317 coordination, are influenced by muscle temperature and motor unit recruitment but not
318 strongly moderated by sex ^{27,32}. Dynamic balance is defined as the ability to maintain
319 stability while shifting body mass, particularly when altering the base of support, being
320 highly relevant for volleyball-specific actions such as blocking, or landing ³³. Dynamic
321 balance outcomes demonstrated minimal sensitivity to sex by time-of-day or time-of-
322 effect, except for the posterolateral reach with the dominant limb, where females showed
323 a significant evening improvement. This limited effect reinforces prior observations that
324 balance relies more on sensory integration and postural control mechanisms, which are

less prone to circadian fluctuations³⁴. Unlike explosive tasks that depend heavily on muscle power and corticospinal excitability, balance performance is predominantly mediated by proprioceptive input, vestibular feedback, and visual processing, systems that tend to remain relatively stable across the day³⁵. Therefore, the small effect observed here only female may indicate subtle sex and circadian modulation of sensorimotor integration, though its practical relevance appears limited.

This study is not without limitations. First, the menstrual cycle phase was not controlled in the female sample. Although evidence remains mixed, some studies suggest that neuromuscular performance may fluctuate across the menstrual cycle⁵, and this factor could have influenced variability in female responses. Second, while chronotype and sleep patterns were partially standardized, individual circadian preferences and sleep–wake rhythms may still have affected performance outcomes. Third, the sample consisted almost exclusively of volleyball players from a single region, which limits the generalizability of our findings to athletes with different anthropometric or neuromuscular characteristics. Finally, no direct assessments of hormonal profiles or corticospinal excitability were performed, restricting our ability to confirm the physiological mechanisms underlying the observed sex by time-of-day interactions.

Conclusions

Males exhibited greater fluctuations across the day, with clear evening improvements in explosive lower-limb and spike-related tasks, whereas females showed smaller or negligible variations, except for modest improvements in handgrip strength and dynamic balance. Time of day exerted a significant influence overall, particularly enhancing explosive and force-related performance in the evening.

Practical Applications

349 **For volleyball players and coaches:** Training design, testing, and competition
350 preparation should account for both sex and time of day. Male volleyball players may
351 benefit from scheduling power- and velocity-oriented sessions (e.g., jumping and spiking
352 drills) in the afternoon, when explosive performance appears to be enhanced. In contrast,
353 female players demonstrate more stable performance across the day, allowing greater
354 flexibility in training and testing schedules without compromising performance
355 outcomes. Coaches are encouraged to individualize training loads and assessment timing
356 according to sex-specific circadian responses to optimize performance.

357 **For researchers:** Future studies should further examine the mechanisms underlying sex-
358 specific diurnal variations in neuromuscular performance, including hormonal,
359 neuromuscular, and chronobiological factors. Additionally, research is needed to
360 determine whether these circadian patterns extend to different competitive levels, training
361 status, and sport-specific tasks, as well as to explore their long-term implications for
362 neuromuscular adaptations.

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365 this study for their invaluable contribution to the study.

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475 **LIST OF TABLES / FIGURES**

Table 1. Differences in sex and time-of-day in neuromuscular performance

Table 2. Differences in sex and time-of-day in the Star Excursion Balance test performance.

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For Peer Review

TABLE 1. Differences in sex and time-of-day in neuromuscular performance (means and standard deviations).

Test	Sex	Morning	Evening	Effect Size (d)	Time-of-Day Effect		Sex × Time Interaction	
					<i>P</i> value	η_p^2	<i>P</i> value	η_p^2
Squat jump (cm)	Male	37.7 ± 7.35	39.8 ± 7.91*	0.76	<0.001	0.19	0.002	0.09
	Female	28.0 ± 3.97	28.5 ± 4.59	0.25				
Countermovement jump (cm)	Male	40.9 ± 7.42	42.5 ± 7.92*	0.69	0.005	0.08	0.080	0.03
	Female	30.0 ± 5.02	30.3 ± 5.21	0.10				
Spike jump (cm)	Male	54.2 ± 9.85	56.5 ± 10.7*	0.64	0.001	0.19	0.268	0.01
	Female	37.2 ± 6.75	38.1 ± 6.85*	0.22				
Standing spike test (km·h ⁻¹)	Male	81.4 ± 10.4	83.7 ± 10.7*	0.38	0.002	0.09	0.396	0.01
	Female	65.7 ± 5.30	66.7 ± 6.36	0.22				
Jumping spike test (km·h ⁻¹)	Male	81.2 ± 10.6	82.5 ± 10.6	0.53	0.738	0.01	0.045	0.04
	Female	64.0 ± 8.54	62.6 ± 7.59	0.27				
Modified T-test (s)	Male	5.87 ± 0.349	5.93 ± 0.344	0.68	0.896	0.01	0.027	0.05
	Female	6.26 ± 0.448	6.19 ± 0.504	0.33				
Isometric handgrip strength DOM (N)	Male	459 ± 82.1	468 ± 77.6	0.25	0.001	0.10	0.413	0.01
	Female	296 ± 42.4	307 ± 37.0*	0.42				
Isometric handgrip strength NO-DOM (N)	Male	431 ± 83.8	443 ± 77.2	0.31	0.001	0.10	0.560	0.01
	Female	283 ± 42.5	294 ± 43.6*	0.35				

Abbreviations: cm = centimetres; N = newtons; DOM = dominant; NO-DOM = non-dominant; η_p^2 = partial eta squared; * = statistically significant difference vs. morning session ($p < 0.05$).

TABLE 2. Differences in sex and time-of-day in the Star Excursion Balance test performance (means and standard deviations).

Test	Sex	Morning	Evening	Effect Size (d)	Time-of-Day Effect		Sex × Time Interaction	
					<i>P value</i>	η_p^2	<i>P value</i>	η_p^2
SEBT anterior DOM (cm)	Male	62.8 ± 6.28	63.2 ± 6.09	0.48	0.405	0.01	0.950	0.01
	Female	61.1 ± 7.01	60.6 ± 6.46	0.21				
SEBT anterior NO-DOM (cm)	Male	63.1 ± 6.49	63.2 ± 6.64	0.51	0.306	0.01	0.271	0.01
	Female	61.4 ± 5.45	62.1 ± 4.62	0.23				
SEBT posterolateral DOM (cm)	Male	83.1 ± 10.6	83.2 ± 9.27	0.57	0.083	0.03	0.036	0.04
	Female	72.2 ± 7.13	74.4 ± 7.61*	0.26				
SEBT posterolateral NO-DOM (cm)	Male	82.6 ± 9.68	81.8 ± 7.76	0.50	0.109	0.03	0.782	0.01
	Female	72.4 ± 6.83	73.4 ± 6.93	0.20				
SEBT posteromedial DOM (cm)	Male	80.1 ± 11.1	78.4 ± 9.10	0.54	0.372	0.01	0.197	0.02
	Female	73.9 ± 7.63	73.8 ± 9.58	0.25				
SEBT posteromedial NO-DOM (cm)	Male	81.5 ± 10.8	79.4 ± 8.37	0.56	0.241	0.01	0.260	0.01
	Female	73.2 ± 6.31	73.4 ± 7.99	0.24				

Abbreviations: η_p^2 = partial eta squared; cm = centimetres; N = newtons; DOM = dominant; NO-DOM = non-dominant; * = statistically significant difference vs. morning session ($p < 0.05$).

DISCUSIÓN GENERAL

4. DISCUSIÓN GENERAL

La presente Tesis Doctoral analiza la influencia del momento del día sobre el rendimiento neuromuscular en jugadores de voleibol, considerando el posible efecto modulador del fenotipo circadiano (cronotipo) y del sexo biológico. Para ello, se integraron los resultados de una revisión sistemática con meta-análisis (Estudio 1) y tres estudios experimentales (Estudios 2-4). En conjunto, los hallazgos obtenidos permiten profundizar en la comprensión de los efectos de la cronobiología en el rendimiento neuromuscular en deportes colectivos y aportan evidencia específica en voleibol, un deporte de equipo que hasta ahora había sido escasamente explorado en este contexto específico en la literatura científica.

De manera general, los resultados de esta Tesis Doctoral muestran que el rendimiento neuromuscular tiende a ser superior en horario vespertino en comparación con el horario matutino, especialmente en tareas explosivas y en acciones específicas del voleibol. Este patrón se observó tanto en la revisión sistemática con meta-análisis como en los estudios experimentales realizados en esta Tesis Doctoral. No obstante, es preciso matizar que, aunque el horario óptimo coincidió en ambos sexos, la mejora del rendimiento neuromuscular a lo largo del día no fue igual en hombres y mujeres. Estos resultados coinciden con investigaciones previas que han descrito mejoras en el rendimiento neuromuscular durante la segunda mitad del día en diferentes deportes de equipo como el fútbol, el balonmano o el baloncesto (Gaos et al., 2024; Pavlović et al., 2018; Reilly et al., 2007). En términos generales, estas variaciones diurnas del rendimiento neuromuscular se han atribuido a la influencia de los ritmos circadianos sobre múltiples variables fisiológicas, entre las que destacan la temperatura corporal central (Waterhouse et al., 2005), la excitabilidad neuromuscular (Chtourou & Souissi, 2012), la activación cortical (Reilly et al., 2007) y los procesos metabólicos (Hayes et al., 2010), factores que tienden a alcanzar niveles más favorables en horario vespertino.

Desde una perspectiva fisiológica, uno de los mecanismos tradicionalmente propuestos para explicar las mejoras vespertinas del rendimiento neuromuscular es el incremento progresivo de la temperatura corporal central a lo largo del día. La temperatura corporal presenta una variación circadiana de aproximadamente 1 °C, alcanzando sus valores máximos en horario vespertino (17:00–19:00 h), lo que coincide con los picos de rendimiento neuromuscular observados (Douglas et al., 2021; Refinetti

& Menaker, 1992). Este paralelismo ha llevado a plantear que la temperatura podría contribuir a las diferencias en el rendimiento neuromuscular en función del momento del día. En este sentido, estudios *in vitro* han sugerido que el aumento térmico podría optimizar la interacción entre actina y miosina durante la contracción muscular (Decostre et al., 2005; Gray et al., 2006). Sin embargo, la extrapolación de estos hallazgos al modelo *in vivo* es controvertida. Aunque se ha observado que una disminución de la temperatura muscular se asocia con reducciones en la fuerza (Bergh & Ekblom, 1979), la variación diaria de la temperatura no parece suficiente para explicar por sí sola las diferencias en el rendimiento neuromuscular (Edwards et al., 2013). De hecho, estudios experimentales han demostrado que el incremento artificial de la temperatura corporal en horario matutino, mediante calentamiento pasivo, no elimina las diferencias de rendimiento neuromuscular respecto al horario vespertino (Edwards et al., 2013). En conjunto, estos hallazgos sugieren que, aunque la temperatura corporal fluctúa en paralelo al rendimiento neuromuscular, no constituye un mecanismo explicativo principal, siendo necesario considerar la interacción con otros factores neurales y moleculares en la regulación de la ritmicidad de la fuerza y la potencia en jugadores de voleibol.

Los resultados de esta Tesis Doctoral sugieren que la magnitud de las fluctuaciones diurnas podría depender de las demandas mecánicas de la tarea evaluada. En particular, las mayores diferencias se observaron en pruebas explosivas, como los saltos verticales o las acciones de remate. Estas tareas requieren una alta activación muscular y utilización del ciclo de estiramiento-acortamiento, factores que podrían ser más sensibles a las variaciones de la excitabilidad neural a lo largo del día (Chtourou & Souissi, 2012; Hirono et al., 2024). Por el contrario, las tareas de carácter isométrico, como la fuerza de prensión manual, mostraron resultados menos consistentes. La evidencia sobre las variaciones diurnas en pruebas isométricas no es concluyente, ya que algunos estudios han encontrado diferencias entre momentos del día (Teo et al., 2011), mientras que otros no (Gaos et al., 2024; López-Samanes et al., 2017). Esta falta de consistencia podría estar relacionada con el grado de experiencia y familiarización de los deportistas con la tarea (Teo et al., 2011). En este sentido, los factores neuromusculares que regulan la fuerza máxima pueden verse condicionados por el aprendizaje motor, lo que podría haber atenuado la expresión de un patrón circadiano claro en la fuerza de prensión en la muestra analizada. Asimismo, a diferencia de las acciones explosivas, las tareas isométricas presentan un mayor tiempo de aplicación de fuerza, lo que podría reducir su dependencia de la velocidad de

activación neural y del componente elástico musculotendinoso (Guette et al., 2005). En conjunto, estos hallazgos sugieren que la influencia del momento del día sobre el rendimiento neuromuscular no es uniforme, sino que podría depender de las características mecánicas de la tarea y del perfil de especialización del deportista.

Otro de los aspectos relevantes analizados en esta Tesis Doctoral fue determinar la influencia del cronotipo o fenotipo circadiano como posible modulador de las variaciones diurnas del rendimiento neuromuscular. De manera global, los resultados obtenidos sugieren que la influencia del cronotipo sobre el rendimiento neuromuscular fue limitada o inexistente en los jugadores y las jugadoras de voleibol evaluados. Este hallazgo coincide con investigaciones previas en deportistas entrenados, que tampoco encontraron una relación clara entre las preferencias circadianas evaluadas mediante cuestionarios y el rendimiento neuromuscular máximo (Küüismaa et al., 2015; López-Samanes et al., 2017). En primer lugar, estos resultados podrían explicarse por la disociación existente entre las preferencias subjetivas y los ritmos biológicos. Cuestionarios como el MEQ o el MCTQ evalúan principalmente la dimensión conductual del cronotipo basada en los hábitos de sueño y vigilia, la cual no siempre guarda una correlación directa con la fase circadiana fisiológica que regula el rendimiento neuromuscular (Vitale & Weydahl, 2017). Asimismo, en poblaciones deportivas con horarios de entrenamiento estructurados, los ritmos circadianos pueden verse modulados por los hábitos de entrenamiento realizados a una hora específica del día (Chtourou et al., 2012; Souissi et al., 2002). Se ha sugerido que el entrenamiento repetido en un mismo momento puede inducir adaptaciones cronobiológicas que atenúan o modifican las predisposiciones circadianas individuales (Rae et al., 2015). En este sentido, la regularidad en el horario de entrenamiento podría actuar como un sincronizador externo, favoreciendo la adaptación del rendimiento neuromuscular a las demandas temporales de la práctica deportiva, independientemente del cronotipo inicial. Este hecho podría explicar la ausencia de interacción entre cronotipo y momento del día observada en la presente Tesis Doctoral, dado que los participantes estaban habituados a entrenar en horario vespertino. En consecuencia, el efecto del momento del día observado podría reflejar tanto la influencia de los ritmos circadianos endógenos como la adaptación al horario habitual de entrenamiento. Además, este tipo de muestras suele presentar una menor variabilidad en el cronotipo, con predominio de perfiles intermedios (Lastella et al., 2016).

La presente Tesis Doctoral aporta evidencia sobre la influencia del sexo biológico en la magnitud de la variación diurna del rendimiento neuromuscular. Aunque el perfil de mejora vespertina fue similar en ambos sexos, los jugadores masculinos mostraron una oscilación vespertino–matutino más pronunciada en tareas explosivas, como los saltos verticales y el remate. Este hallazgo coincide con estudios que muestran una mayor variación diaria de la potencia muscular en hombres que en mujeres (Augsburger et al., 2022; Chtourou et al., 2012). Desde una perspectiva neurofisiológica, estas diferencias podrían estar relacionadas con el perfil de activación de las unidades motoras. Los hombres presentan fibras musculares de tipo II de mayor tamaño, lo que favorece el reclutamiento de unidades motoras de alto umbral y una mayor capacidad para generar fuerza explosiva (Trevino et al., 2019). Esta mayor dependencia de mecanismos neuromusculares asociados a la producción rápida de fuerza podría hacer que el rendimiento neuromuscular masculino sea más sensible a las variaciones diarias en la excitabilidad neural (Hunter & Senefeld, 2024; Trevino et al., 2019). Este patrón podría estar relacionado con diferencias en el perfil neuromuscular. En este sentido, las mujeres presentan, en términos relativos, una mayor proporción de fibras tipo I y estrategias de activación orientadas a una mayor resistencia a la fatiga (Trevino et al., 2019; Weber et al., 2006). Estas características se han asociado con una menor producción de potencia, pero también con una mayor resistencia a la fatiga neuromuscular y una respuesta más estable en tareas repetidas o de intensidad submáxima (Ansdell et al., 2019; Roberts et al., 2020). En consecuencia, mientras que el rendimiento neuromuscular masculino parece estar más supeditado a la ventana temporal de reclutamiento de las unidades motoras de alto umbral, el perfil neuromuscular femenino podría favorecer un rendimiento más constante, minimizando el impacto del momento del día en la expresión de la fuerza explosiva.

En conjunto, la presente Tesis Doctoral aporta evidencia específica sobre la cronobiología del rendimiento neuromuscular en voleibol, permitiendo contrastar la hipótesis planteada. Los resultados confirman que el rendimiento neuromuscular varía en función del momento del día, mostrando valores superiores en horario vespertino respecto al matutino, especialmente en tareas de carácter explosivo. Sin embargo, la influencia del cronotipo sobre estas variaciones fue limitada, no observándose una mejora del rendimiento neuromuscular asociada a la coincidencia entre el momento del día y las preferencias circadianas individuales. Asimismo, aunque se observaron diferencias entre

hombres y mujeres en la magnitud de la variación diurna del rendimiento neuromuscular, estas fueron de escasa magnitud y no se manifestaron de forma consistente en todas las variables analizadas. Desde una perspectiva aplicada, estos hallazgos sugieren la conveniencia de estandarizar la hora de las valoraciones físicas para asegurar la comparabilidad de los datos. Además, la programación de entrenamientos de alta intensidad podría realizarse preferentemente en horarios próximos al momento habitual de competición, con fin de optimizar la adaptación neuromuscular a las condiciones reales de juego.

**LIMITACIONES Y
FUTURAS LÍNEAS
DE INVESTIGACIÓN**

5. LIMITACIONES Y LÍNEAS FUTURAS DE INVESTIGACIÓN

5.1 Limitaciones

A pesar del rigor metodológico aplicado en los estudios que conforman esta Tesis Doctoral, es necesario reconocer algunas limitaciones que deben considerarse al interpretar los resultados y su grado de generalización.

En primer lugar, la evaluación del fenotipo circadiano se fundamentó exclusivamente en cuestionarios subjetivos. Esta aproximación metodológica no permitió considerar factores genéticos determinantes, como los polimorfismos en los genes reloj (p. ej., CLOCK, BMAL1, PER o CRY), cuya influencia en la regulación de los ritmos biológicos podría explicar parte de la variabilidad interindividual observada en la respuesta neuromuscular al momento del día. Aunque el uso de cuestionarios está ampliamente validado en la literatura, estos presentan limitaciones inherentes asociadas a la autopercepción y a factores sociolaborales. Al evaluar principalmente patrones conductuales de sueño-vigilia, estas herramientas no permiten determinar con precisión la fase circadiana fisiológica subyacente si no se complementan con medidas objetivas, como el genotipo circadiano o, en su defecto, la actigrafía o marcadores endocrinos (p. ej., melatonina o cortisol) (Kantermann et al., 2015; Roenneberg et al., 2003). En consecuencia, la ausencia de una validación objetiva del cronotipo obliga a interpretar con cautela la interacción entre las preferencias circadianas y el rendimiento neuromuscular.

En segundo lugar, en la muestra de jugadoras de voleibol no se controló de forma estricta la fase del ciclo menstrual ni el uso de anticonceptivos hormonales en el momento de la evaluación. La literatura científica ha señalado que las fluctuaciones hormonales asociadas al ciclo menstrual podrían influir en determinadas respuestas fisiológicas relacionadas con el ejercicio. Sin embargo, la evidencia disponible es heterogénea y, en muchos casos, muestra efectos de magnitud trivial o una elevada variabilidad interindividual (McNulty et al., 2020). En este sentido, la ausencia de un control estricto del ciclo menstrual y del uso de anticonceptivos hormonales podría haber contribuido a parte de la variabilidad observada en el rendimiento neuromuscular de las jugadoras, aunque su magnitud real continúa siendo objeto de debate. Por tanto, futuros estudios deberían considerar el control más preciso de estas variables con el fin de comprender

mejor su posible interacción con el rendimiento neuromuscular en diferentes momentos del día.

En tercer lugar, la evaluación del rendimiento neuromuscular se realizó únicamente en dos momentos del día: uno en horario matutino (9:00 h) y otro en horario vespertino (19:00 h). Aunque este planteamiento es coherente con la literatura previa y permite comparar los extremos funcionales del ciclo diurno, además de ajustarse a los horarios habituales de entrenamiento y competición de las personas participantes, no permite una caracterización completa del perfil circadiano. La utilización de solo dos momentos del día impide identificar posibles patrones no lineales, picos intermedios de rendimiento o diferencias individuales en el momento de máxima expresión del rendimiento neuromuscular, especialmente cuando se consideran variables moduladoras como el sexo, el cronotipo o la base genética (Portaluppi et al., 2010). Si bien un mayor número de evaluaciones distribuidas a lo largo del día habría permitido describir con mayor precisión las variaciones del rendimiento a lo largo del día, esta opción habría incrementado significativamente la carga experimental.

5.2 Líneas futuras de investigación

A partir de los resultados obtenidos en esta Tesis Doctoral y de las limitaciones identificadas, se proponen diversas líneas de investigación futura orientadas a profundizar en la interacción entre la genética y los ritmos circadianos en el rendimiento neuromuscular.

En primer lugar, futuras investigaciones podrían incorporar análisis genéticos que permitan evaluar la influencia de polimorfismos en genes reloj implicados en la regulación del sistema circadiano, como CLOCK, BMAL1, PER o CRY. El estudio de estas variantes genéticas o genotipos circadianos podría ayudar a explicar la variabilidad interindividual observada en la respuesta del rendimiento neuromuscular al momento del día, así como a identificar perfiles genéticos asociados a una mayor o menor sensibilidad a las fluctuaciones circadianas. En esta línea, nuestro grupo de investigación actualmente desarrolla diferentes diseños de estudio orientados a integrar el análisis cronogenético con la evaluación del rendimiento neuromuscular en deportistas, y se ha iniciado la recogida de datos preliminares en esta dirección.

En segundo lugar, sería de especial interés analizar la interacción entre genes relacionados con el rendimiento neuromuscular (p. ej., ACTN3) y el momento del día. Este enfoque permitiría determinar si ciertos genotipos de rendimiento presentan respuestas diferenciales en tareas de fuerza, potencia o agilidad según el horario de evaluación, contribuyendo al desarrollo de modelos explicativos más integradores que combinen cronobiología y genética del rendimiento neuromuscular.

Por último, resultaría de interés avanzar en el desarrollo o adaptación de herramientas específicas que permitan evaluar el cronotipo desde una perspectiva aplicada al rendimiento neuromuscular. Los cuestionarios más utilizados en la literatura científica, como el Morningness–Eveningness Questionnaire (Horne & Ostberg, 1976) o el Munich Chronotype Questionnaire (Roenneberg et al., 2003), se centran fundamentalmente en las preferencias relacionadas con los patrones de sueño y vigilia y en la organización temporal de los hábitos cotidianos. Sin embargo, estos instrumentos no fueron diseñados específicamente para evaluar cómo se manifiestan las diferencias circadianas en el contexto del entrenamiento o del rendimiento neuromuscular. En este sentido, el desarrollo de cuestionarios específicos para población deportista, o la adaptación de los ya existentes, podría incorporar dimensiones relacionadas con la percepción del rendimiento neuromuscular en distintos momentos del día, la tolerancia al entrenamiento en diferentes franjas horarias, la percepción de fatiga asociada al horario de práctica o los hábitos de práctica de actividad física en diferentes momentos del día. La inclusión de este tipo de variables, junto con el estudio del genotipo circadiano, permitiría obtener información más directamente relacionada con la expresión funcional del fenotipo circadiano en el contexto deportivo. Esto facilitaría su aplicación práctica en la planificación del entrenamiento y la programación de la competición.

CONCLUSIONES

6. CONCLUSIONES

La presente Tesis Doctoral ha investigado la influencia del momento del día, el fenotipo circadiano (cronotipo) y el sexo biológico sobre el rendimiento neuromuscular en jugadores de voleibol.

En el **Estudio 1**, se examinó el efecto del momento del día sobre el rendimiento neuromuscular (p. ej., la capacidad de salto vertical o la agilidad) en deportistas de nivel profesional o semiprofesional. En dicha revisión sistemática con meta-análisis se pudo observar que el rendimiento neuromuscular en deportistas de equipo profesionales y semiprofesionales está influido por el momento del día, observándose niveles de rendimiento neuromuscular significativamente superiores en la capacidad de salto vertical y en la agilidad en horario vespertino (16:00–20:00 h) en comparación con el horario matutino (7:00–10:00 h). Estos resultados respaldan parcialmente la hipótesis planteada, al evidenciar una mejora del rendimiento neuromuscular en franjas horarias vespertinas.

En el **Estudio 2**, se analizó el efecto del momento del día sobre el rendimiento neuromuscular en jugadoras de voleibol semiprofesionales. En dicho estudio se observó que el momento del día afectó a la velocidad de golpeo de balón en el remate, a la flexibilidad del miembro dominante, al equilibrio dinámico del miembro no dominante y al rendimiento neuromuscular en las pruebas de agilidad, observándose un mayor rendimiento en la sesión vespertina (19:00 h) en comparación con la sesión matutina (9:00 h). Además, se analizó de forma secundaria la asociación del cronotipo sobre el rendimiento neuromuscular, no encontrándose ninguna relación. En consecuencia, los hallazgos de este estudio apoyan parcialmente la hipótesis planteada, mostrando que el efecto del momento del día sobre el rendimiento se observa de forma consistente, mientras que el cronotipo no parece condicionar el rendimiento neuromuscular.

En el **Estudio 3**, se determinó la influencia del fenotipo circadiano (cronotipo) sobre el rendimiento neuromuscular en jugadores de voleibol masculino. Se observó que el rendimiento en horario vespertino fue superior al rendimiento matutino en las pruebas de salto vertical (*squat jump*, salto con contramovimiento y salto de remate), en la velocidad del balón en el remate (prueba de remate en apoyo) y en la fuerza isométrica (fuerza de presión manual del miembro no dominante). Sin embargo, la influencia del fenotipo

circadiano sobre el rendimiento neuromuscular fue limitada (salto con contramovimiento y el salto de remate) y siendo inexistente en el resto de las variables analizadas (*squat jump*, velocidad del balón en el remate y fuerza de prensión manual). Estos resultados indican que el momento del día, más que el fenotipo circadiano, desempeña un papel más relevante en la optimización del rendimiento neuromuscular en jugadores masculinos de voleibol. En consecuencia, los hallazgos respaldan parcialmente la hipótesis planteada, al confirmar la influencia del momento del día, mientras que el fenotipo circadiano muestra una capacidad moduladora limitada en las variables neuromusculares analizadas.

En el **Estudio 4**, se evaluó la influencia del sexo biológico sobre las variaciones diurnas del rendimiento neuromuscular en jugadores y jugadoras de voleibol. Los resultados de este estudio mostraron que el momento del día ejerció una influencia significativa en el rendimiento neuromuscular, con valores superiores en horario vespertino. No obstante, la magnitud de estas fluctuaciones no se manifestó de forma homogénea; mientras que en los jugadores masculinos se observaron incrementos significativos en tareas de potencia del tren inferior y acciones de remate en horario vespertino, en las jugadoras la respuesta fue más heterogénea y de menor amplitud. Estas diferencias deben interpretarse con cautela debido a la falta de control del ciclo menstrual y de variables hormonales, sin poder atribuirse de forma concluyente a mecanismos sexo-específicos. Por tanto, estos resultados respaldan parcialmente la hipótesis planteada, al confirmar la mejora vespertina en ambos sexos, pero difieren de la misma al no observar el pico de rendimiento matutino esperado en la población femenina.

Por lo tanto, en base a la hipótesis general planteada:

El rendimiento neuromuscular en jugadores de voleibol variará en función del momento del día, mostrando valores superiores en horario vespertino respecto al matutino. Esta variación estará modulada por el cronotipo, de modo que una mayor alineación entre el momento del día y las preferencias circadianas del individuo se asociará con un mejor rendimiento neuromuscular. Además, el efecto del momento del día sobre el rendimiento neuromuscular diferirá según el sexo.

Se eleva a la categoría de tesis y se concluye que:

El rendimiento neuromuscular en jugadores de voleibol varía en función del momento del día, mostrando de forma consistente valores superiores en horario vespertino respecto

al horario matutino, especialmente en tareas de carácter dinámico y explosivo. Por su parte, el fenotipo circadiano (cronotipo) mostró una influencia limitada sobre estas variaciones, sin observarse una mejora consistente asociada a la coincidencia entre el momento del día y las preferencias circadianas individuales. En cuanto al sexo, se identificaron algunas diferencias en la respuesta diurna del rendimiento neuromuscular; no obstante, estas fueron moderadas y poco consistentes entre las distintas variables analizadas.

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ANEXOS

8. ANEXOS

CURRÍCULUM INVESTIGADOR DERIVADO DE TESIS DOCTORAL

La presente sección recoge la actividad investigadora desarrollada en el marco de la Tesis Doctoral. Este trabajo ha dado lugar a una producción científica consolidada en revistas indexadas, la participación en proyectos competitivos y la difusión de resultados en congresos nacionales e internacionales, contribuyendo al avance del conocimiento en el ámbito de las ciencias salud y deporte.

PUBLICACIONES CIENTIFICAS

1. **Martín-López, J.**, Pérez-López, A., Varillas-Delgado, D., & López-Samanes, Á. (2025). Influence of time-of-day on neuromuscular performance in team sport athletes: A systematic review and meta-analysis. *Frontiers in Sports and Active Living*, 6, 1466050. <https://doi.org/10.3389/fspor.2024.1466050>
2. **Martín-López, J.**, Sedliak, M., Valadés, D., Muñoz, A., Buffet-García, J., García-Oviedo, R., Rodríguez-Aragón, M., Pérez-López, A., & López-Samanes, Á. (2022). Impact of time-of-day and chronotype on neuromuscular performance in semi-professional female volleyball players. *Chronobiology International*, 39(8), 1006–1014. <https://doi.org/10.1080/07420528.2022.2057322>
3. **Martín-López, J.**, Muñoz, A., Montalvo-Alonso, J. J., Quintana-Milla, I., Sánchez-Jorge, S., Conejero, M., López-Samanes, Á., & Pérez-López, A. (2026). Circadian phenotype influence on neuromuscular performance in male volleyball players. *Chronobiology International*, 43(3), 421–430. <https://doi.org/10.1080/07420528.2025.2606270>
4. Del Val-Manzano, M., Montalvo-Alonso, J. J., Valadés, D., Ferragut, C., **Martín-López, J.**, López-Samanes, Á., & Pérez-López, A. (2025). Muscular strength and power and endurance performance at loads exceeding 25% of one-repetition maximum are unaffected by time-of-day in resistance-trained male participants. *Chronobiology International*, 42(8), 1039–1047. <https://doi.org/10.1080/07420528.2025.2524520>

5. Berlanga, L. A., López-Samanes, Á., **Martín-López, J.**, De la Cruz, R. M., Garcés-Rimón, M., Roberts, J., & Bertotti, G. (2023). Dietary nitrate ingestion does not improve neuromuscular performance in male sport climbers. *Journal of Human Kinetics*, 87, 47–57. <https://doi.org/10.5114/jhk/161812>
6. Peris, F., Conejero, M., **Martín-López, J.**, Guerrero-Alarcón, R., Franco, A., Martínez-Deza, P., & Martín-Miguel, I. (2026). Efecto de la ingesta de cafeína sobre el rendimiento físico-técnico en jugadores profesionales de pádel masculinos. *Padel Scientific Journal*, 4(1), 13–32. <https://doi.org/10.17398/2952-2218.4.13>

PROYECTOS DE INVESTIGACIÓN

1. **Influencia de los ritmos circadianos y del cronotipo en el rendimiento físico en voleibol**, (Referencia del proyecto: UFV2022-19) financiado por el Vicerrectorado de Investigación de la Universidad Francisco de Vitoria (UFV). Financiación: 9.920 EUR. Periodo: enero de 2022 – diciembre de 2024. Investigador principal: Alejandro Muñoz Moreno.
2. **Efectos de la cafeína en el rendimiento deportivo en jugadores de pádel profesional** (Referencia del proyecto: UFV2023-47), financiado por el Vicerrectorado de Investigación de la Universidad Francisco de Vitoria (UFV). Financiación: 9.500 EUR. Periodo: enero de 2023 – diciembre de 2024. Investigador principal: Rafael Alarcón Guerrero.
3. **Efectos a nivel cardiovascular y marcadores inflamatorios en un programa de 12 semanas de entrenamiento de tenis** (Referencia del proyecto: UFV2024-30), financiado por el Vicerrectorado de Investigación de la Universidad Francisco de Vitoria (UFV). Financiación: 7.700 EUR. Periodo: enero de 2024 – diciembre de 2025. Investigador principal: Julio Martín López.
4. **Influence of time-of-day, sleep and PER3 genotype on neuromuscular and cognitive factors in badminton players**, financiado por la Badminton World Federation (BWF). Financiación: 5.800 EUR. Periodo: noviembre de 2024 – noviembre de 2026. Investigador principal: Álvaro López Samanes.
5. **Evaluación biomecánica y neuromuscular del entrenamiento funcional con Steel Clubs** (Referencia del proyecto: UFV2026-25), financiado por el Vicerrectorado de Investigación de la Universidad Francisco de Vitoria (UFV). Financiación: 19.500

EUR. Periodo: enero de 2026 – diciembre de 2027. Investigador principal: Jorge Gutiérrez Hellín

CONTRIBUCIÓN EN CONGRESOS

1. **XIII Simposio Internacional de Actualizaciones en Entrenamiento de la Fuerza**, celebrado en la Facultad de Ciencias de la Actividad Física y del Deporte (INEF) de la Universidad Politécnica de Madrid, los días 17–18 de diciembre de 2021. Tipo de contribución: póster. Trabajo presentado: Impact of time-of-day and chronotype on neuromuscular performance in semi-professional female volleyball players.
2. **XIV Simposio Internacional de Actualizaciones en Entrenamiento de la Fuerza**, celebrado en la Facultad de Ciencias de la Actividad Física y del Deporte (INEF) de la Universidad Politécnica de Madrid, los días 16–17 de diciembre de 2022. Tipo de contribución: póster. Trabajo presentado: Dietary nitrate ingestion does not improve neuromuscular performance in male rock climbers: a randomized, double-blind, placebo-controlled study.
3. **XII Congreso Internacional de la Asociación Española de Ciencias del Deporte (AECD)**, celebrado en la Universidad Autónoma de Madrid, los días 21–23 de junio de 2023. Tipo de contribución: comunicación oral. Trabajo presentado: Influence of time-of-day on neuromuscular performance in team sport athletes: a systematic review and meta-analysis.
4. **XII Congreso Internacional de la Asociación Española de Ciencias del Deporte (AECD)**, celebrado en la Universidad Autónoma de Madrid, los días 21–23 de junio de 2023. Tipo de contribución: comunicación oral. Trabajo presentado: Análisis de las variables de rendimiento deportivo en jóvenes jugadores de pádel en función de su pico de velocidad de crecimiento, categoría y género.
5. **XII Congreso Internacional de la Asociación Española de Ciencias del Deporte (AECD)**, celebrado en la Universidad Autónoma de Madrid, los días 21–23 de junio de 2023. Tipo de contribución: comunicación oral. Trabajo presentado: Neuromuscular but not technical performance is affected by time-of-day in semi-professional female basketball players.
6. **Congreso Nacional de Pádel, celebrado en la Facultad de Ciencias del Deporte** de la Universidad de Extremadura (UEX), Cáceres, los días 15–16 de septiembre de 2023. Tipo de contribución: comunicación oral. Trabajo presentado: El efecto de la cafeína en el rendimiento deportivo de jugadores de alto nivel de pádel.

7. **XV Simposio Internacional de Actualizaciones en Entrenamiento de la Fuerza**, celebrado en la Facultad de Ciencias de la Actividad Física y del Deporte (INEF) de la Universidad Politécnica de Madrid, los días 15–16 de diciembre de 2023. Tipo de contribución: póster. Trabajo presentado: Impact of beetroot juice ingestion on neuromuscular performance in healthy older women.
8. **29th Annual Congress of the European College of Sport Science (ECSS)**, celebrado en Glasgow (Scotland, UK), del 2 al 5 de julio de 2024. Tipo de contribución: póster. Trabajo presentado: The time of day causes no effect on muscular strength and power performance.
9. **XVI Simposio Internacional de Actualizaciones en Entrenamiento de la Fuerza**, celebrado en la Facultad de Ciencias de la Actividad Física y del Deporte (INEF) de la Universidad Politécnica de Madrid, los días 13–14 de diciembre de 2024. Tipo de contribución: póster. Trabajo presentado: Anthropometric and neuromuscular characteristics differ between elite, sub-elite and amateur female volleyball players.
10. **XIII Congreso Internacional de la Asociación Española de Ciencias del Deporte (AECD)**, celebrado en la Universidad Miguel Hernández de Elche, los días 1–3 de octubre de 2025. Tipo de contribución: comunicación oral. Trabajo presentado: Circadian phenotype influence on neuromuscular performance in male volleyball players.
11. **II Congreso Internacional de Nutrición Aplicada a la Mujer Deportista**, celebrado en la Universidad Pontificia Comillas (Madrid), los días 27–28 de noviembre de 2025. Tipo de contribución: comunicación científica. Trabajo presentado: Sex differences in the effects of diurnal variation on neuromuscular performance in volleyball players.

