

TESIS DOCTORAL



UNIVERSIDAD
DE ALMERÍA

DOCTORADO EN EDUCACIÓN (RD09/11)
ESCUELA INTERNACIONAL DE DOCTORADO

**Efectos fisiológicos del entrenamiento basado en la
variabilidad de la frecuencia cardíaca en corredores
de fondo**

-

**Physiological effect of training based on heart rate
variability in endurance runners**

Doctorando:

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Almería, marzo de 2022

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Como director y codirectora de la tesis doctoral titulada:

**Efectos fisiológicos del entrenamiento basado en la
variabilidad de la frecuencia cardíaca en corredores de
fondo**

Realizada por el estudiante de doctorado D. Alberto González Quílez

CERTIFICAN que la presente tesis doctoral, mediante la modalidad de compendio de publicaciones, reúne las condiciones en cuanto a rigor científico, originalidad y elaboración para su lectura y defensa, pudiendo optar a la obtención del Título de Doctor por la Universidad de Almería.

Y, para que surta los efectos oportunos, firmamos el presente en Almería, marzo de 2022.

Fdo.: Antonio Granero Gallegos

Fdo.: María Carrasco Poyatos

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Índice de abreviaturas

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BMI – Índice de Masa Corporal

CI – Intervalo de confianza

EEP – Escala de esfuerzo percibido

ECG – Electrocardiograma

ES – Tamaño del efecto

FC – Frecuencia cardíaca

FC_{ex} – Frecuencia cardíaca durante el ejercicio

FC_{rec} – Frecuencia cardíaca de recuperación

GC – Grupo control

HIIT – Entrenamiento interválico de alta intensidad

HIIT/MOD – Entrenamiento en intervalos de alta intensidad/ moderada

HRV – Heart Rate Variability (Variabilidad de la Frecuencia Cardíaca)

HR_{pre} – Frecuencia cardíaca previa

HR_{post} – Frecuencia cardíaca posterior

HR_{post1} – Frecuencia cardíaca un minuto posterior

HR_{post3} – Frecuencia cardíaca tres minutos posteriores

HR_{post5} – Frecuencia cardíaca cinco minutos posteriores

HRV-G – Grupo de entrenamiento guiado por el HRV

HRmax – Frecuencia cardíaca máxima

HRV4Training – App medición HRV

Intervalo RR – Tiempo transcurrido entre dos ondas R consecutivas en el electrocardiograma

IAAF – Asociación Internacional de Federaciones de Atletismo

INE – Instituto Nacional de Estadística

Lactate_{pre} – Test previo de lactato

Lactate_{post} – Test posterior de lactato

Lactate_{post3} – Test posterior de lactato tres minutos

Lactate_{post5} – Test posterior de lactato cinco minutos

LnrMSSD – Logaritmo neperiano de las diferencias cuadráticas medias de intervalos RR sucesivos

LnrMSSD_{CV} – Cambio en el coeficiente de variación del logaritmo neperiano de las diferencias cuadráticas medias de intervalos RR sucesivos

LnrMSSD7-d – Promedio semanal del logaritmo neperiano de las diferencias cuadráticas medias de intervalos RR sucesivos

OMS – Organización Mundial de la Salud

PR – Período de preparación cuatro semanas

rMSSD – Desviación estándar de los intervalos RR

RER – Cociente respiratorio

RPE – Escala de esfuerzo percibido

SNA – Sistema nervioso autónomo

SNP – Sistema nervioso parasimpático

SNS – Sistema nervioso simpático

TR – Período de entrenamiento ocho semanas

TRAD-G – Grupo de entrenamiento basado en metodología tradicional

V_{máx} – Velocidad máxima

VT1 – Umbral ventilatorio 1

VT2 – Umbral Ventilatorio 2

VO₂ – Consumo de oxígeno

VO_{2máx} – Consumo máximo de oxígeno

Resumen

Resumen

En el mundo del entrenamiento de resistencia se continúa en la búsqueda del estímulo más eficiente para conseguir las adaptaciones óptimas. La periodización del entrenamiento basada en macrociclos es un procedimiento consolidado en las intervenciones del entrenamiento de resistencia. Sin embargo, en la actualidad, debido a las limitaciones de recursos y tiempo, se está apostando por la planificación basada en microciclos para conseguir mejores resultados a nivel cardiovascular. Este cambio metodológico incide directamente sobre el principio de individualización, ya que una planificación diaria se ajustará mejor al estado fisiológico del deportista para afrontar el entrenamiento. Esto, además, permitirá ajustar mejor la carga de entrenamiento diaria, favoreciendo, presumiblemente, el tiempo de entrenamiento a altas intensidades. Según la literatura publicada hasta el momento, tanto la individualización como el entrenamiento de alta intensidad beneficiarán la optimización del rendimiento a nivel cardiovascular de los deportistas de resistencia.

La monitorización del sistema nervioso autónomo está siendo utilizada actualmente como un método prometedor para optimizar la prescripción del entrenamiento de resistencia. Esto se debe a que las respuestas fisiológicas al entrenamiento están relacionadas con el equilibrio entre la actividad parasimpática y simpática del sistema nervioso autónomo. Por tanto, la variabilidad de la frecuencia cardiaca (HRV, por sus siglas en inglés) está siendo utilizada como el indicador de la activación diaria del sistema nervioso autónomo. Una mayor activación parasimpática se refleja en mayores valores de HRV, lo que indica una mejor recuperación. El HRV es entendido como el intervalo de tiempo entre dos latidos consecutivos. Se obtiene calculando el intervalo de tiempo que transcurre entre dos ondas R consecutivas en un electrocardiograma. Sin embargo, actualmente se han validado otras herramientas en forma de aplicaciones para móviles que facilitan el procedimiento de recogida de datos, como el

HRV4Training. Dado que esta línea de investigación es muy reciente, se hace necesario seguir profundizando en el efecto que esta nueva metodología de prescripción del entrenamiento tiene sobre el rendimiento de los deportistas de resistencia.

Para aportar información al respecto, esta tesis doctoral se ha conformado con tres artículos. El primero, una revisión sistemática con metaanálisis para analizar el efecto del entrenamiento basado en HRV sobre el VO_{2max} de atletas de resistencia. El segundo, el diseño de un protocolo de intervención para optimizar el entrenamiento basado en HRV en atletas de resistencia. Y el tercero, un estudio controlado y aleatorizado en el que se aplicó el protocolo del segundo artículo para analizar el efecto del entrenamiento guiado por HRV sobre parámetros cardiovasculares de atletas de resistencia. De acuerdo con los resultados encontrados en la revisión sistemática con metaanálisis, el entrenamiento guiado por HRV es más eficiente para mejorar el VO_{2max} de deportistas de resistencia que el entrenamiento basado en una metodología tradicional, y los resultados están condicionados por el nivel y el sexo de los deportistas. En este sentido, el protocolo para ensayos controlados y aleatorizados se diseñó para ser llevado a cabo con atletas de élite de género masculino. Tras su puesta en práctica en el estudio controlado y aleatorizado, los corredores que entrenaron en base al HRV lo hicieron a mayor intensidad y menor volumen, obtuvieron mejores resultados a nivel cardiovascular y mejoraron también sus valores de HRV. Por tanto, el entrenamiento basado en HRV es recomendable para la optimización del entrenamiento en deportistas de resistencia.

Palabras clave: HRV, entrenamiento de resistencia, desempeño del entrenamiento, deportistas de alto nivel, VO_{2max} , correr, rendimiento, variabilidad de la Frecuencia Cardíaca, deportistas de alto nivel, consumo máximo de oxígeno.

Abstract

Abstract

In the world of resistance training, the search continues for the most efficient stimulus to achieve optimal adaptations. Macrocycle-based training periodization is an established procedure in resistance training interventions. However, currently, due to resource and time limitations, microcycle-based planning is being used to achieve better results at the cardiovascular level. This methodological change directly affects the principle of individualization, since a daily planning will better adjust to the physiological state of the athlete to face training. This will also allow better adjustment of the daily training load, presumably favoring the training time at high intensities. According to the literature published so far, both individualization and high intensity training will benefit the optimization of cardiovascular performance in endurance athletes.

Autonomic nervous system monitoring is currently being used as a promising method to optimize the prescription of resistance training. This is because physiological responses to training are related to the balance between the parasympathetic and sympathetic activity of the autonomic nervous system. Therefore, heart rate variability (HRV) is being used as the indicator of the daily activation of the autonomic nervous system. Greater parasympathetic activation is reflected in higher HRV values, indicating better recovery. HRV is understood as the time interval between two consecutive beats. It is obtained by calculating the time interval between two consecutive R waves in an electrocardiogram. However, other tools have currently been validated in the form of mobile applications that facilitate the data collection procedure, such as HRV4Training. Given that this line of research is very recent, it is necessary to continue delving into the effect that this new training prescription methodology has on the performance of endurance athletes.

To provide information in this regard, this doctoral thesis has consisted of three articles. The first, a systematic review with meta-analysis to analyze the effect of HRV-based

training on the VO_{2max} of endurance athletes. The second, the design of an intervention protocol to optimize HRV-based training in endurance athletes. And the third, a controlled and randomized study in which the protocol of the second article was applied to analyze the effect of HRV-guided training on cardiovascular parameters of endurance athletes. According to the results found in the systematic review with meta-analysis, HRV-guided training is more efficient in improving the VO_{2max} of endurance athletes than training based on a traditional methodology, and the results are conditioned by the level and gender of athletes. In this sense, the protocol for randomized controlled trials was designed to be carried out with elite male athletes. After its implementation in the randomized controlled study, the runners who trained based on HRV did so at higher intensity and lower volume, obtained better results at the cardiovascular level and also improved their HRV values. Therefore, HRV-based training is recommended for optimizing training in endurance athletes.

Keywords: HRV, endurance training, training performance, high-level athletes, VO_{2max} , running, performance, heart rate variability, high-level athletes, maximum oxygen consumption

Introducción

Marco teórico

En la literatura especializada se pueden encontrar una gran cantidad de publicaciones científicas y técnicas que han orientado sus objetivos hacia el estudio y control de variables funcionales en el campo del entrenamiento. Su extenso trabajo y experimentación se ha realizado tanto en situaciones de reposo como en aquellas circunstancias en las que el deportista era o es sometido a esfuerzos de diferentes características (intensidad, duración, etc). Como resultado de estas investigaciones, se ha creado un sólido cuerpo de conocimiento que se ha empleado de forma continua en el campo de entrenamiento para el desarrollo de protocolos de trabajo que permitan abordar el proceso de formación y preparación de los deportistas desde una perspectiva cada vez más científica. Es en esta línea en la que se engloba esta tesis doctoral que se centra en el estudio de los procesos de optimización del control del entrenamiento, apoyándose en instrumentos fiables y validados científicamente que pueden ser usados en la práctica diaria por el entrenador y le permita disponer de una herramienta accesible, barata, no invasiva y de uso cotidiano en el proceso de entrenamiento de los deportistas.

Se sabe que el entrenamiento es fundamental para mejorar el rendimiento físico (Hostrup & Bangsbo 2017) y que optimizar el entrenamiento para mejorar el rendimiento de los atletas es un área importante de investigación dentro de la fisiología y medicina deportiva (Bangsbo 2015; Laursen 2010). En este sentido, existen diferentes métodos de entrenamiento, los cuales dan lugar a diversas mejoras en el rendimiento, como el entrenamiento de alta intensidad (Bangsbo, 2015) o las pruebas submáximas (Capostagno, Lambert, & Lamberts 2016). Los componentes clave en cualquier programa de entrenamiento son el volumen, la intensidad, y la frecuencia de las sesiones de ejercicio, siendo la combinación de éstos “impulsos de entrenamiento” la que determina la magnitud de las respuestas adaptativas que

mejoran el estado físico o la condición de un atleta (Hawley, 2008). Combinar estos elementos clave en la búsqueda de una mejor formación y rendimiento representa un área de investigación relevante dentro de la fisiología del ejercicio y la medicina deportiva.

Además, un programa de entrenamiento estándar aplicado a un grupo de atletas puede inducir diversas respuestas en términos de rendimiento y adaptaciones fisiológicas (Bouchard & Rankinen 2001; Vesterinen et al., 2016). Por lo tanto, la individualización se reconoce como un principio de entrenamiento (Hawley, 2008), así como la necesidad de ajustar los estímulos de entrenamiento a la capacidad de carga psicofísica y la tolerancia individual de cada atleta, si las respuestas individuales a las cargas de entrenamiento y recuperación están destinadas a un rendimiento óptimo (Kiviniemi et al., 2010). De esta manera, usar el mismo programa de entrenamiento estandarizado para un grupo de atletas puede provocar una amplia gama de reacciones en términos de rendimiento y adaptaciones fisiológicas (Bouchard & Rankinen 2001; Hautala et al. 2006).

Como afirman Schmitt et al. (2018), un componente importante de la variabilidad interindividual en las respuestas fisiológicas al entrenamiento estandarizado está relacionado con el equilibrio entre la actividad parasimpática (PNS) y simpática (SNS) del sistema nervioso autónomo (SNA) (Aubert, Seps, & Beckers, 2003; Chandola et al., 2010). Según Plews et al. (2013), la variabilidad de la frecuencia cardíaca (HRV, por sus siglas en inglés) es uno de los indicadores que permite el estudio no invasivo de la actividad del SNA en sus ramas simpática y parasimpática.

Según Huang et al. (2018), el HRV se considera la variación en el intervalo de tiempo entre dos latidos consecutivos y se obtiene calculando el intervalo de tiempo entre dos ondas R consecutivas (es decir, fluctuación del intervalo RR) en el electrocardiograma (ECG). Tradicionalmente el HRV se ha medido con ECG (Rodas et al., 2008), aunque actualmente, el desarrollo de nuevas aplicaciones (i.e., smartphone applications: Kubios-HRV, Elite-HRV,

Mobile Lab, o HRV4Training) facilitan las medidas diarias de HRV y su cuantificación y, por tanto, la adaptación individual de cargas de entrenamiento y recuperación. Una de las formas de cuantificar el HRV es a través de rMSSD (la raíz cuadrada media de las diferencias sucesivas entre los intervalos RR adyacentes) (Ortigosa et al., 2018) ya que es un parámetro estadístico temporal que reporta aquellas variaciones que ocurren en el corto plazo entre intervalos RR (Martín-Guillaumes et al., 2018). Valores elevados de HRV se relacionan con una mayor activación del SNS y son un indicador de una mejor recuperación tras el esfuerzo (Ortigosa-Márquez et al., 2017).

El HRV se puede utilizar, por tanto, para la evaluación individual de las respuestas a las cargas de entrenamiento y adaptación a la recuperación (Vesterinen, Nummela, et al., 2016; Williams et al., 2017). En los últimos años, el HRV se ha utilizado para analizar estos desequilibrios entre SNS y SNP en atletas (Lagos et al., 2006) y evaluar diferentes aspectos relacionados con el entrenamiento (Ortigosa-Márquez et al., 2017), como la intensidad del ejercicio y duración (Seiler, Haugen, & Kuffel 2007), recuperación y sobreentrenamiento (Hedelin et al., 2000), carga de entrenamiento (Javaloyes et al., 2019) o perfiles psicológicos (Moreno et al., 2013).

La importancia de la periodización del entrenamiento de macrociclo (semana o meses) es bien reconocida en las intervenciones de entrenamiento de resistencia. Sin embargo, algunos estudios experimentales (Kiviniemi et al., 2007; 2010) destacan la relevancia de la periodización del microciclo (día a día) para lograr mejores adaptaciones cardiovasculares, especialmente cuando se opera bajo las limitaciones de recursos y tiempo limitados. Si el HRV se registra diariamente, se podrían incluir sesiones más ajustadas al estado fisiológico del deportista, lo que favorecería la optimización de la intensidad del entrenamiento, beneficiándose los atletas de mayores adaptaciones fisiológicas periféricas y centrales.

Aunque existen estudios experimentales con diferentes muestras de deportistas de

resistencia, aún existe una falta de consenso en cuanto al diseño del entrenamiento guiado por el HRV (Nuuttila et al., 2017; Ramón Martins et al., 2019), en términos del volumen total alcanzado y la proporción de entrenamiento realizado en intensidades altas, moderadas o bajas. Tampoco parece claro si el entrenamiento guiado por el HRV induce un mejor rendimiento fisiológico, lo que lleva a una mayor velocidad de carrera (Kiviniemi et al., 2007; Nuuttila et al., 2017; Ramón Martins et al., 2019) o mejora el $VO_{2máx}$ (Kiviniemi et al., 2007; 2010; Silva et al., 2019). Además, el HRV no siempre mejora estadísticamente después de una intervención guiada por HRV (Hautala et al., 2006; Kiviniemi et al., 2010; Silva et al., 2019; Vesterinen, Nummela, et al., 2016), y su correlación con los resultados de rendimiento solo se ha analizado en un estudio (Nuuttila et al., 2017).

Como se ha indicado anteriormente, en los últimos años se han realizado diversos estudios experimentales basados en la evaluación del entrenamiento guiado por HRV en atletas de resistencia, tanto en deportistas de élite, en deportes como ciclismo (Javaloyes et al., 2019) y esquí (Schmitt, Willis, Fardel, Coulmy, & Millet, 2018), como con atletas aficionados (Silva et al., 2019; Vesterinen et al., 2016). Pero, hasta nuestro conocimiento, aún no se ha llevado a cabo ningún estudio experimental de estas características con corredores profesionales de resistencia. Con los estudios incluidos en la presente tesis se contribuye a dar respuesta a este vacío en la literatura científica internacional.

Hipótesis y objetivos

Considerando lo anterior, en la presente tesis doctoral se plantearon los siguientes objetivos. En el artículo 1, al tratarse de una revisión sistemática con meta-análisis se plantean preguntas de investigación en lugar de hipótesis y en el artículo 2, al tratarse de un protocolo no se establecen hipótesis.

- Artículo 1:
 - Objetivos:
 - Analizar el efecto del entrenamiento basado en el HRV sobre el VO_{2max} en corredores de resistencia.
 - Preguntas de investigación:
 - ¿El entrenamiento basado en el HRV tiene efectos sobre el VO_{2max} ?
 - ¿El efecto de este tipo de entrenamiento sobre el VO_{2max} es superior al de un entrenamiento tradicional?
 - ¿El nivel de los deportistas es determinante para obtener un efecto sobre el VO_{2max} ?
 - ¿El efecto de este tipo de entrenamiento sobre los valores de VO_{2max} varían según el sexo del atleta?
- Artículo 2:
 - Objetivo:
 - Diseñar un protocolo para la optimización del rendimiento de atletas de alto nivel después de un período de entrenamiento guiado por HRV y otro basado en el entrenamiento tradicional.

- Artículo 3:
 - Objetivos:
 - Analizar la estructura del entrenamiento (volumen e intensidad) realizado por un grupo de entrenamiento guiado por HRV y un grupo de entrenamiento tradicional (HRV-G y TRAD-G), compuestos por corredores de resistencia profesionales.
 - Determinar el efecto de los dos métodos de entrenamiento sobre el rendimiento cardiovascular y la modulación vagal de los atletas.
 - Hipótesis:
 - El HRV-G presentará valores de HRV más altos y entrenará a mayor intensidad y menor volumen, lo que se espera tenga un impacto positivo en su rendimiento cardiovascular y recuperación.
 - Además, se hipotetiza que los atletas del HRV-G conseguirán mejores resultados en las pruebas de rendimiento cardiovascular que los TRAD-G y que las puntuaciones altas de HRV estarán asociadas a un mejor rendimiento de los corredores.

Diseño general de la tesis doctoral

En esta tesis doctoral, en primer lugar, se realizó una revisión sistemática con meta-análisis con el objetivo de estudiar el efecto del entrenamiento guiado por HRV en el rendimiento de deportistas de resistencia. Para este estudio se siguieron las indicaciones de la Colaboración Campbell para revisiones sistemáticas (2019) y se incluyeron ensayos controlados aleatorizados (RCT, por sus siglas en inglés), así como estudios experimentales que utilizaron un método aleatorio para la asignación al tratamiento con el fin de disminuir el riesgo de sesgo de asignación. Tras la búsqueda inicial, se seleccionaron cinco artículos de texto completo que incluyeron 195 participantes (134 hombres y 61 mujeres) que fueron incluidos en el análisis cualitativo y en el meta-análisis. Los resultados sugieren que el HRV es un buen indicador de las respuestas fisiológicas al entrenamiento en atletas de resistencia y la utilización de las puntuaciones diarias de HRV para la individualización del entrenamiento es un método eficaz para la mejora del rendimiento, lo que se refleja en la mejora del $VO_{2máx}$ que, como se ha puntualizado anteriormente, es uno de los principales indicadores de rendimiento del deportista.

A partir de los resultados de esta revisión sistemática con meta-análisis, en segundo lugar, se planteó un protocolo de entrenamiento basado en HRV para corredores de resistencia mediante un RCT por conglomerados. Dado que existe ausencia de un protocolo común a seguir en este tipo de intervenciones, el protocolo propuesto en el segundo artículo que compone esta tesis doctoral contribuye a la literatura científica de varias formas: (i) propone una investigación centrada en deportistas profesionales, una muestra de población para la cual hasta la fecha solo se han realizado dos estudios experimentales; (ii) el protocolo está destinado a investigar corredores de resistencia, de los que no tenemos conocimiento de que se haya realizado ninguna investigación sobre este tipo de muestra y con estas características a nivel internacional. Por tanto, se trata de un trabajo novedoso que pretende proporcionar

apoyo empírico al entrenamiento guiado por HRV en corredores de larga distancia, adaptando el entrenamiento diario a las respuestas fisiológicas de cada atleta individual. Además, para su puesta en práctica se propone el uso de tecnologías emergentes en los campos del entrenamiento y la investigación, como aplicaciones para teléfonos inteligentes; en este caso, HRV4Training, una aplicación validada científicamente que permite calcular la medición diaria del HRV por el propio atleta.

El tercer artículo de esta tesis doctoral es el resultado de la puesta en práctica de la metodología propuesta en el protocolo anterior. Se llevó a cabo un RCT por conglomerados para analizar los efectos del entrenamiento basado en HRV en el rendimiento y la modulación vagal de corredores de resistencia. Se encontró que los atletas que entrenaron en base al HRV (HRV-G) realizaron más sesiones de entrenamiento de alta intensidad que el grupo que realizó un entrenamiento tradicional (TRAD-G), lo que se traduce en que los corredores del HRV-G se recuperaban mejor del esfuerzo, consiguiendo además un mayor aumento en su HRV. Aunque ambos grupos mejoraron su rendimiento cardiovascular, HRV-G obtuvo mejores valores en el VO_{2max} , la velocidad de carrera y el cociente ventilatorio. Estos resultados mostraron una correlación significativa con sus puntuaciones de $\ln rMSSD$, reflejando la vinculación del rendimiento con la modulación vagal. Por tanto, se demostró que el control del entrenamiento en base al HRV es una medida que favorece la individualización del entrenamiento y el rendimiento cardiovascular en corredores de resistencia.

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Publicación 1:

**“HRV-Based Training for Improving
VO_{2max} in Endurance Athletes. A
Systematic Review with Meta-
Analysis”**

Estudio 1

HRV-Based training for improving VO_{2Max} in endurance athletes. A systematic review with meta-analysis

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HRV-Based training for improving VO_{2Max} in endurance athletes. A systematic review with meta-analysis

Abstract

Nowadays, heart rate variability (HRV)-guided training is emerging as an alternative for optimizing the training individualization. However, its effect on maximal oxygen uptake (VO_{2max}) is still not clear. This review aimed to synthesize evidence regarding interventions based on HRV-guided training for VO_{2max} improvements in endurance athletes, and to address the issues that have an impact on this performance enhancement. The Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE, CINAHL complete, the Web of Science core collection, Global Health, Current Contents Connect, and the SciELO citation index were searched. Inclusion criteria were: Randomized controlled trials; Studies with trained athletes enrolled in any regular endurance training; Studies that recruited both men and women; Studies on resistance training controlled by HRV; Studies that measured performance with VO_{2max} ; Studies published in English between the years 2010 and 2020. Two authors evaluated the risk of bias. A random-effects meta-analysis calculating the effect size (ES) was used. Moderator analyses (according to the athlete's level and gender) and meta-regression (according to the number of participants in each group) were undertaken to examine differences in ES. HRV-guided training group and control group enhanced the athletes' VO_{2max} ($p < 0.0001$), but the ES for the HRV-guided training group was significantly higher ($p < 0.0001$; $ES_{HRVG-CG} = 0.175$). The amateur level and the female subgroup reported better significant results ($p < 0.0001$) for VO_{2max} . The results were not determined by the sample size. HRV-guided training has a moderate positive effect on endurance athlete performance (VO_{2max}), conditioned by the athlete's level and sex.

Keywords: performance, heart rate variability, high level constant, maximal oxygen uptake.

Introduction

Description of the condition

The key components in any training program are the volume (i.e., how much), intensity (i.e., how hard) and frequency (i.e., how often) of the exercise sessions, and the combination of these 'training impulses' determines the magnitude of adaptive responses that improve the physical condition of an athlete, or increase fatigue [1]. Combining these key elements to optimize training in athletes for better performance represents a relevant area of research within exercise physiology and sports medicine [2], and it is recognized that a standard training program applied to a group of athletes can induce diverse responses in terms of performance and physiological adaptations [3,4]. Therefore, individualization is recognized as a training principle [1] as well as the need to adjust training stimuli to the psychophysical load capacity and individual tolerance of each athlete, if individual responses to training and recovery loads are intended for optimal performance [5]. The maximal oxygen uptake (VO_{2max}) is considered as one of the main indicators for measuring the performance and cardiovascular adaptation of the athlete to training loads [6]. The VO_{2max} is defined as the largest volume of oxygen that a body can capture, use and transport during intense exercise [7, p. 70] and is a determining factor of endurance performance [7,8]. As Vesterinen et al. [4,9] state, although some athletes show great improvements in endurance performance after standardized group training (even up to 40% in VO_{2max}), other athletes show no changes or benefits, and sometimes even show a decrease in resistance performance. In recent years, research has looked at whether heart rate variability HRV-guided training has positive effects on athletic performance, given that this type of training (day-to-day training) allows daily adjustment of the training and recovery stimuli, individually based on HRV records [4,5,10].

Description of the intervention

HRV is an indicator that enables the non-invasive analysis of autonomic nervous system activity in both its sympathetic and parasympathetic branches [11]. This is relevant if we consider that an important component of the inter-individual variability in physiological responses to training is related to the balance between the parasympathetic (PNS) and sympathetic (SNS) activity of the autonomic nervous system (ANS) [12]. According to Huang et al. [13], HRV is considered as the variation in the time interval between two consecutive heartbeats, and is obtained by calculating the time interval between two consecutive R waves (i.e., RR interval fluctuation) in the electrocardiogram (ECG). Since the elapsed time between beats is not constant, high HRV values are associated with efficient ANS, promoting behavioural adaptation and cognitive flexibility during stress [14], whilst low HRV is indicative of an inefficient ANS, resulting in maladaptive responses to stress and perceived threats [13, p. 46]. HRV analysis is considered a useful method for measuring the heart's ability to adapt to endogenous and exogenous loads [15], therefore, it can be used for the individual assessment of responses to training loads and recovery adaptation [4,16]. High HRV measurements indicate more parasympathetic than sympathetic activation, which is indicative of better recovery and preparedness for facing high-intensity training sessions [17]. HRV-guided training starts with a preparation period of about four weeks, which serves as a standardized data collection phase to obtain the baseline HRV values (e.g., $\ln rMSSD$; the Neperian logarithm of the square root of the mean value of the sum of the squares of the differences between the adjacent RR intervals) and their range of normality (upper and lower limits) for each athlete [9,18]. Once the normal range of HRV measurements has been established, the training prescribed (moderate or high intensity session) is based on this calculation, which is normally updated weekly [19]. Traditionally, HRV has been measured with ECG [20], and quantified by means of $rMSSD$ [17]. Currently, the development and validation of new applications (i.e.,

smartphone applications: Kubios-HRV, Elite-HRV, Mobile Lab, or HRV4Training) facilitate daily HRV measurements and their quantification and, thus, the individual adaptation of training loads and recovery.

How the intervention might work

Bellenger et al. [21], in a recent systematic review with meta-analysis, highlighted the need to use monitoring systems that accurately reflect the athletes' adaptations to the training stimulus. Although there have been numerous research studies using the HRV measure to check wellness and training adaptation in athletes [22,23], these have not focused on performance improvement based on HRV-guided training, but have followed training interventions based on a traditional and non-individualized methodology.

In contrast, evidence does exist supporting the use of HRV-guided training for improved performance in endurance athletes. With this type of training monitoring, some studies have found significant VO_{2max} improvements in athletes who have developed individualized endurance training programs based on daily HRV values. These studies alternated moderate-intensity sessions with high-intensity sessions [4,10] or even with rest sessions, vigorous-intensity training and moderate-intensity exercise [5]. However, Javaloyes et al. [18], in a program with similar characteristics developed with professional cyclists, found no significant improvements in VO_{2max} . Likewise, significant improvements have been found among athletes following HRV-guided training in other variables, for example, for lactate in maximal test [10], speed in maximal test [4], time in maximal test [4,10], or muscle strength [24]. At the level of perceived recovery, significant improvements have also been found in variables such as general stress, emotional stress, lack of energy, and even in overall mood disturbance [25]. HRV-guided training may, therefore, function as an alternative method for improving performance in resistance athletes.

Why is this review important?

In the search to improve athletic performance, different training methods have been tried and studied, such as intensified training [2] or submaximal test [26]. However, it has also been recognized that the same training program followed by a group of athletes can provoke a wide range of reactions in terms of performance and physiological adaptations [3] and that overuse injuries occur due to repetitive submaximal loading of the musculoskeletal system when there is inadequate rest to allow for structural adaptation to take place [27]. In recent years HRV-guided training has shown itself to be a promising method for improving different performance variables (e.g., VO_{2max}) compared to predefined training (traditional training), through the monitoring and individualization of endurance athletes' training [4,28]. HRV-guided training has been investigated in randomized trials on samples from different endurance sports, such as skiers [28], runners [4,25] and cyclists [18]), as well as on athletes of different ages and levels: elite [18,28] and recreational endurance athletes [5,24,25]. It is therefore important to carry out a systematic review and meta-analysis of the different experimental studies conducted so far on endurance athletes in order to assess whether HRV-guided training is an effective method for performance improvement.

Objectives

As mentioned above, this review aimed to analyse the effect of HRV-guided training on VO_{2max} in endurance athlete.

We asked the following research questions regarding HRV-guided training in endurance athletes:

- Research Question 1: Does HRV-based training have an effect on VO_{2max} ?
- Research Question 2: Is the effect of this type of training superior to that of traditional training?

- Research Question 3: Is the level of the athletes decisive in obtaining an effect on the VO_{2max} ?
- Research Question 4: Does the effect of HRV-guided training determine VO_{2max} scores according to the gender of the athlete?

Methods

The methods detailed below are reported in accordance with The Campbell Collaboration policies and guidelines for systematic reviews [29].

Criteria for considering studies for this review (eligibility criteria)

Types of studies

We included randomized controlled trials (RCTs) and the first period of cross-over RCTs and experimental studies using a random method for the treatment assignment in order to reduce the risk of allocation bias. We restricted study eligibility by language. We did not restrict study eligibility by publication status.

Types of participants

We included studies with trained athletes enrolled in any regular endurance training (e.g., runners, triathletes, skiers, and cyclists). We included studies that recruited both men and women, or men and women separately.

Types of interventions

We included studies on endurance training controlled by heart rate variability to improve the athletes' performance. We considered designs comprising any dose, frequency and duration. We also included studies that compared endurance training controlled by HRV to no treatment, or to another training treatment. The types of comparisons included were as follows:

- Endurance training controlled by HRV versus no specific training intervention (e.g., habitual physical activity).

- Endurance training controlled by HRV versus another training intervention (e.g., traditional endurance training or another type of traditional training).
- Endurance training controlled by HRV versus another training intervention (i) versus a further training intervention (ii).
- Endurance training controlled by HRV (i) versus endurance training controlled by HRV (ii) versus another training intervention versus no specific training intervention.

Types of outcome Measures

- Primary
 - Maximal oxygen consumption (VO_{2max}).

Search methods to identify the studies

Electronic searches

The register contains studies identified from the Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE, CINAHL complete, the Web of Science core collection, Global Health, Current Contents Connect, and the SciELO citation index.

The search is up to date as of 15th June 2020. The language was restricted, considering only English or Spanish. Studies were included that were published up until 10 years ago. The terms used to search the databases were: (amateur OR elite OR train *) AND (HRV-guided OR "heart-rate variability guided")

Searching other resources

We checked the reference lists of all the included studies and systematic reviews for additional references. We contacted experts in the field and the authors of the included studies to identify additional unpublished studies. We also checked the results of completed trials registered on the US National Institutes of Health Ongoing Trials Register, ClinicalTrials.gov, the World Health Organization International Clinical Trials Registry Platform (WHO ICTRP) and

proceedings of conferences for relevant research.

Data collection and analysis

We conducted the following data collection and analysis in accordance with the recommendations in the Cochrane Handbook for Systematic Reviews of Interventions [30].

Selection of studies

Two review authors independently screened the titles and abstracts of all the retrieved references in Excel (Microsoft Excel 2018 for Windows). The full-text study reports were retrieved for all the citations that at least one review author considered potentially relevant. Two review authors independently screened the full-text articles and identified studies for inclusion; they also identified and recorded the reasons for excluding studies in the excluded studies characteristics. Any disagreements were resolved through discussion. The selection process is detailed in a PRISMA flow diagram [31].

Data extraction and management

We used a standardized piloted data collection form in Microsoft Excel 2018 for Windows and extracted the following study characteristics and outcome data: (i) Methods: study design; (ii) Participants: randomized number, study participants mean age or age range, study location and setting, recruitment methods, inclusion and exclusion criteria, and type of endurance sport; (iii) Interventions: a description of the training intervention characteristics, the dose and duration of the training intervention, a description of the comparison intervention characteristics, the length of follow-up, the number of withdrawals and the reasons for withdrawal; (iv) Outcomes: a description of the primary and secondary outcomes in the review that were reported in the trial, and a listing of other outcomes collected in the trial; (v) Notes: the trial funding and notable conflicts of interest of the trial authors; (vi) a 'risk of bias' assessment. Two review authors independently extracted the outcome data from the included studies into Microsoft Excel 2018 spreadsheets and compared the data to identify any

discrepancies in the data entries. Any disagreements were resolved by consensus. In the Characteristics of Included Studies section, we noted down if a trial did not report outcome data in a usable way. We then transferred all the outcome data into the Comprehensive Meta-Analysis software, version 2.2.064 [32].

Risk-of-bias assessment in the included studies

Two review authors (MCP, AGG) independently assessed the risk of bias for each included trial using the Cochrane risk-of-bias tool [30]. Any disagreements were resolved by discussion. The risk of biases was assessed for the following domains: random sequence generation (selection bias), allocation concealment (selection bias), blinding of participants and personnel (performance bias), blinding of outcome assessment for each outcome (detection bias), incomplete outcome data (attrition bias), selective outcome reporting (reporting bias) and other biases (such as the validity of outcome measure and baseline comparability). Each potential source of bias was assessed as either high, low or unclear, and a quotation from the study report was provided together with a justification for the judgment in the 'risk of bias' tables. The judgments across the different studies were summarized for each of the domains listed.

Treatment effect Measures

The outcome data for each study were uploaded into the data tables of the Comprehensive Meta-Analysis software to calculate the treatment effects. We used the mean difference (MD) for continuous outcomes reported on the same scale, and the standardized mean difference (SMD) for continuous outcomes measured on different scales in different trials. Uncertainty was expressed with 95% confidence intervals (CIs) for all the effect estimates.

Assessment of heterogeneity and reporting bias

Heterogeneity was assessed qualitatively between studies in three ways: a visual examination of the forest plots, the Chi² test ($p \leq 0.10$) for heterogeneity, and the I² statistic. The implications of the observed I² statistic value were considered as follows: 0% to 40%: might

not be important; 30% to 60%: may represent moderate heterogeneity; 50% to 90%: may represent substantial heterogeneity; 75% to 100%: considerable heterogeneity [30]. Publication bias was assessed by examining the asymmetry of a funnel plot using Egger's test. If studies were distributed symmetrically around the mean effect size (ES), there was an absence of publication bias [33]. Subgroup analysis was carried out using the outcome for athlete level (elite vs amateur) and sex ('men vs women' vs 'men and women'). Meta-regression was used to assess the relationship between the studies and the variable sample size.

Sensitivity analysis

A sensitivity analysis was carried out to check whether the results varied according to the endpoint data.

Results

Description of the studies

Search results

The search produced a total of 31 studies, with 221 additional records identified through other sources. The removal of duplicates resulted in nine studies, which were screened by the two authors based on the title and abstract. Three studies were excluded. Six full-text articles were assessed for eligibility. One more study was excluded and five studies were included either in the qualitative analysis or in the quantitative meta-synthesis. The PRISMA flow chart illustrates the search and selection process (Figure 1).

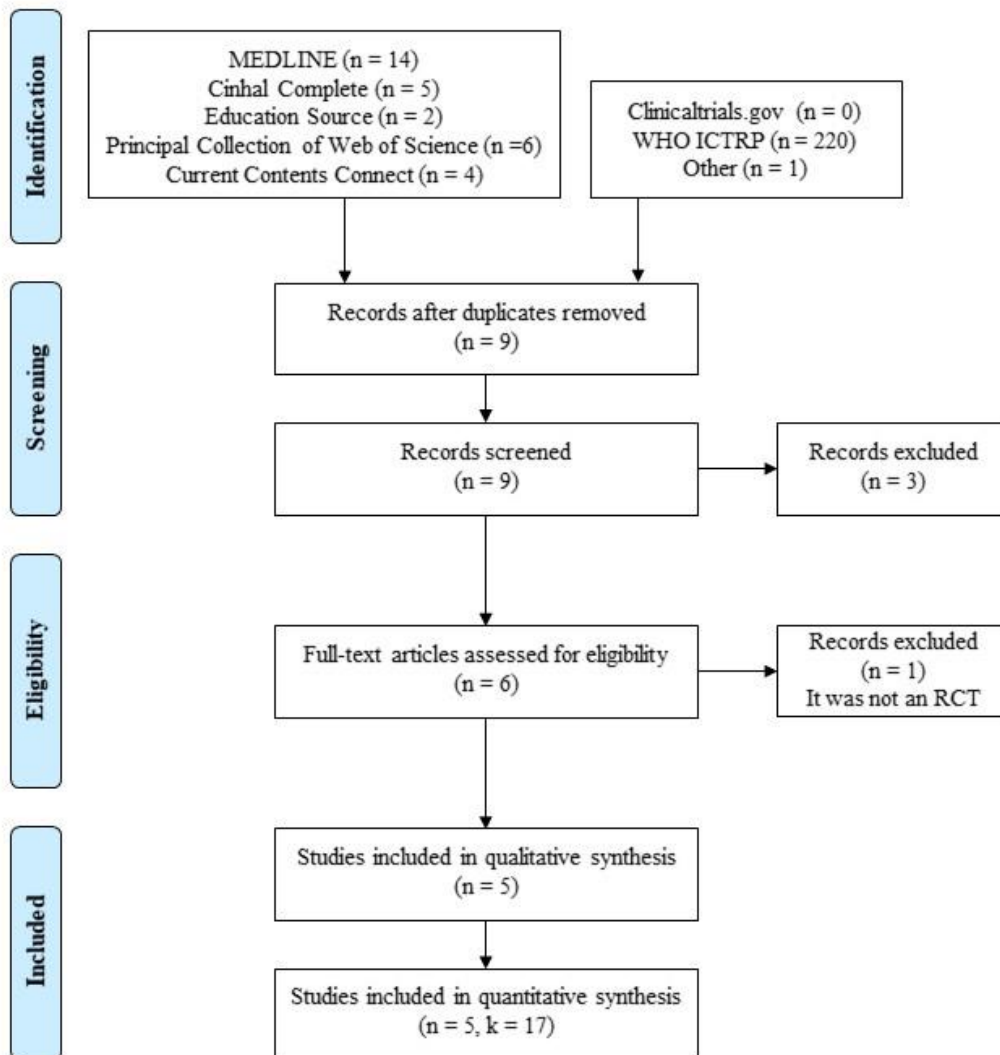


Figure 1. Study flow diagram following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses Guidelines [31]. N number of papers, k number of individual studies.

Included studies

Five studies carrying out HRV-guided training with elite or amateur athletes were included in this review [4,5,10,18,28], which were identified by the first author and the publication date: Javaloyes_2019, Kiviniemi_2010, Nuuttila_2017, Schmitt_2018, and Vesterinen_2016.

- *Study location*

Schmitt_2018 conducted their study at the French National Ski-Nordic Centre, while the locations for the other four studies were not specified.

- *Study design*

Every study included in this review was a randomized controlled trial.

- *Participants*

A total of 165 participants (104 men and 61 women) were included in these studies. Javaloyes_2019 and Nuutila_2017 considered only male samples of 17 and 32 participants, respectively. In the rest of the studies, the samples were composed of men and women: Kiviniemi_2010 included 24 men and 36 women, Schmitt_2018 incorporated 19 men and 5 women, and Vesterinen_2016 assessed 20 men and 20 women. In the studies by Javaloyes_2019 and Schmitt_2018, the samples were composed of professional athletes (cyclists and Nordic-skiers, respectively) while in the other three studies, the samples were of a non-professional level.

- *Interventions*

According to the types of comparisons contemplated in the present systematic review (a. Endurance training controlled by HRV versus no specific training intervention; b. Endurance training controlled by HRV versus other training intervention; c. Endurance training controlled by HRV (i) versus another training intervention (ii) versus another training intervention; d. Endurance training controlled by HRV (i) versus endurance training controlled by HRV (ii) versus other training intervention versus no specific training intervention), Javaloyes_2019, Nuutila_2017, and Vesterinen_2016 were classified in Comparison b; Schmitt_2018 in Comparison c, and Kiviniemi_2010 in Comparison d.

The interventions in the included studies focused on running (Kiviniemi_2010,

Nuutila_2017, and Vesterinen_2016), skiing (Schmitt_2018), and cycling (Javaloyes_2019). They were from 7 to 15-weeks long. In most of the studies, three (Nuutila_2017) or four (Javaloyes_2019, Schmitt_2018 and Vesterinen_2016) low-intensity preparation weeks were followed either by the experimental or the control groups (standard training) before the intervention. An 8-week intervention was carried out in every study except for Schmitt_2018, which considered 15 days of training. The assessment weeks were treated separately from the intervention period only in Javaloyes_2019 and Schmitt_2018.

In every study, the experimental groups trained at moderate or high intensities according to their daily HRV scores. The control groups (standard training) followed a predefined training design at high, moderate and low intensities (Javaloyes_2019), high and moderate intensities (Kiviniemi_2010 and Nuutila_2017), or moderate and low intensities (Vesterinen_2016). The control group (standard training) design was not explained in Schmitt_2018.

- *Outcomes*

The primary outcome analysed in the included studies was VO_{2max} . The secondary outcomes were: ventilatory thresholds (Javaloyes_2019), power in the cycling test (Javaloyes_2019); rMSSD or RR interval (Javaloyes_2019 and Schmitt_2018); basal heart rate (Nuutila_2017; Kiviniemi_2010, and Schmitt_2018); maximal heart rate in the ergometer test (Nuutila_2017); speed in the treadmill test (Nuutila_2017, and Vesterinen_2016); maximal speed in the 10-meters test (Nuutila_2017); time and lactate in the 3000m test (Nuutila_2017); maximal load in the ergometer test (Kiviniemi_2010); and oxygen saturation and VO_2 at the second ventilatory threshold (Schmitt_2018).

Further details about participants, interventions, comparators, and outcomes are provided in Table 1.

Table 1. Overview of the studies included in the review.

Author, Year	Method	Participants	Intervention	Outcomes	Results	Risk of Bias		
						Bias	Author's Judgement	Support for The Judgement
Javaloyes_2019	Randomized controlled trial	Trained male cyclist, mean age of 38.42 years. N = 17: EG = 9 + CG = 8 Location: not specified. Recruited from local clubs Inclusion criteria: at least 2 years of experience in cycling. Exclusion criteria: not specified.	15 weeks (4 weeks of baseline period to capture baseline HRV + 8 weeks of training + 3 weeks of assessments); 4-7 sessions/week; time depended on the training intensity. EG: HRVG-based training before each session; training MICT and HIIT according to HRV. CG: 4 high intensity training sessions + 4 High intensity interval training sessions + 6 moderate intensity training sessions + 2-5 low intensity training sessions/week. No follow-up periods. No withdrawals.	Primary: VO _{2max} (maximal bicycle ergometer test-direct measurement). Secondary: ventilatory thresholds in graded test, peak power output in graded test, rMSSD with heart rate monitor + kubios (LnrMSSD), mean power output during a 40 minutes all-out cycling test.	VO _{2max} : no significant differences intra-groups and inter-groups. Moderate training load: significant inter-group differences (EG = 24%; CG = 27%). VT2: significant improvements in EG (36.11 ± 3.73W). Peak power output: significant improvements in EG (17.45 ± 3.91W). LnrMSSD: significant differences inter-groups for the percentage of change (EG = 0.85 ± 3.21%, CG = -2.02 ± 5.21%). Mean power 40M: significant improvements in EG (17.67 ± 3.03W).	Selection Performance Detection Attrition Reporting Other	Unclear High High Low Unclear Low	Insufficient information about the sequence generation process and allocation to permit judgement of 'Low risk' or 'High risk'. Incomplete blinding, and the outcome is likely to be influenced by lack of blinding The study did not address this outcome No missing outcome data Insufficient information to permit judgement of 'Low risk' or 'High risk' The study appears to be free of other sources of bias

Kiviniemi_20 10	Randomized controlled trial	Healthy men and women. Mean age of 34.57 years.	8 weeks of aerobic exercise sessions (40 min); vigorous-intensity level: HR between 85% of HRpeak - 5 bpm lower limit; moderate-intensity exercise was 70% of HRpeak - 5 bpm lower limit.	Primary: VO _{2max} (maximal bicycle ergometer direct measurement). Secondary: HR, R-R interval with hearth rate monitor, maximal load in ergometer test.	VO _{2max} : intra-group significant improvements in ST (men subgroup) (pre-test = 50 ± 7; post-test = 53 ± 7ml/kg/min), ST (women subgroup) (pre-test = 35 ± 5; post-test = 37 ± 4ml/kg/min), HRV-I (men subgroup) (pre-test = 50 ± 6; post-test = 54 ± 6ml/kg/min), HRV-I (women subgroup) (pre-test = 36 ± 4; post-test = 39 ± 3ml/kg/min), and in HRV-II (women subgroup) (pre-test = 37 ± 5; post-test = 40 ± 5ml/kg/min). HR: R-R interval: intra-group significant improvements in HRV-I (men subgroup) (pre-tests = 13.7 ± 6.7; post-test = 16.9 ± 8.7ms). Maximal load: intra-group significant improvements in	Selection	High	Random sequence generation and allocation concealment.
		N = 60. Men, n = 24; women, n = 36). ST: standard training (8 men + 8 women) + HRV-I: HRV-guided training for men and women (EG: 8 men + 8 women) + HRV-II: HRV guided training tailored for woman (12) + CG (8 men + 8 women). Location: No specified Recruitment: advertisement local newspaper Inclusion criteria: healthy men and women Exclusion criteria: to smoke, BMI ≥ 30 kg/m ² ; regular physical	HRV-I: if HRV increased or did no change, vigorous-intensity training on that day; if HRV decreased, moderate-intensity exercise or rest. HRV-II: vigorous-intensity exercise only when HRV had increased. ST group: two moderate-intensity and three vigorous-intensity exercises weekly. CG: no intervention Not follow-up period. 7 withdrawals: ST (1 men + 1 women) + HRV-I (7 men + 7 women); CG (7 men + 8 women) + HRV-II (10); 4 because of illness or injury and 3		Performance	High	No blinding of participants and personnel	
						Detection	High	The study did not address this outcome
						Attrition	High	High rates of loss to follow up
						Reporting	Unclear	Insufficient information to permit judgement of 'Low risk' or 'High risk'
						Other	Low	The study appears to be free of other sources of bias

exercise training more than twice a week during the last 3 months, competing athletes, mellitus asthma, or cardiovascular disorders.

because of insufficient compliance.

ST (men subgroup) (pre-test = 275 ± 28W; post-test = 293 ± 35W), ST (women subgroup) (pre-test = 179 ± 32W; post-test = 198 ± 35W), HRV-I (men subgroup) (pre-test = 270 ± 29W; post-test = 300 ± 25W), HRV-I (women subgroup) (pre-test = 174 ± 28W; post-test = 189 ± 25W), and in HRV-II (women subgroup) (pre-test = 177 ± 26W; post-test = 194 ± 23W).

Nuutila_2017	Randomized controlled trial	Males, 19-37 years. N = 24. EG = 13 and CG = 11. Location: not specified. Recruitment not specified. Inclusion criteria: recreationally	11 weeks (3 weeks of control + 8 weeks of training). EG: 2-5 sessions/week; CG: 6 sessions/week; time depended on the training intensity. EG: 4 moderate intensity endurance training sessions + 20 high interval intensity training session. Training MICT and HIIT according to HRV.	Primary: VO ₂ max (maximal treadmill test-direct measurement). Secondary: basal heart rate, maximal heart rate, lactate in treadmill, V _{max} in treadmill test, V _{max} in 10m,	VO ₂ max: significant intra-group changes (EG = 3.1 ± 0.8ml/kg/min; CG = 2.2 ± 0.6ml/kg/min). Basal HR: significant intra-groups decrease (EG = 4.4 ± 0.6bpm; CG = 3.6 ± 0.1bpm). V _{max} in treadmill: intra-groups significant	Selection Performance Detection Attrition Reporting	High High High High Unclear	Allocation based on the results of a laboratory test or a series of tests No blinding of participants and personnel The study did not address this outcome High rates of loss to follow up Insufficient information to permit judgement of 'Low risk' or 'High risk'
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		endurance training. Exclusion criteria: not specified	CG: 22 moderate intensity endurance training sessions + 20 high interval intensity training sessions + 4 high intensity strength training sessions No follow-up periods. 9 withdrawals: illness (n=1), injuries (n=2), personal reasons (n=3), lack of adherence (n=3).	time and lactate in 3000 test, rMSSD with heart rate monitor + Firstbeat. Other: body weight, height in countermovement jump, strength in concentric dynamic leg press, nocturnal heart rate variability, testosterone and cortisol (blood samples), % of fat (InBody 720).	improvements (EG = 0.9 ± 0.1 km/h; CG = 0.5 ± 0.1 km/h). Vmax in 10 m: decreased significantly in CG from pre to mead test (0.08 ± 0.04 m/s). Time in 3000 test: significant intra-groups decrease (EG = 35 ± 2 s; CG = 35 ± 6 s). Lactate in 3000 test: intra-groups significant improvements (EG = $12 \pm 18.4\%$) from mead-to post test; CG = $16-0 \pm 23.5\%$) from pre-to post test. rMSSD: significant improvements in EG (13 ± 3 ms).	Other	Low	The study appears to be free of other sources of bias
Schmitt_2017	Randomized controlled trial	24 elite Nordic skiers (19 men, age 23.3 ± 3.6 ; 5 women, age 22.8 ± 4.1). N = 24; H-HRV, HRV-guided training normobaric hypoxic group (n = 9) + H, sleeping in	Prior to pre-test: 3 low intensity training weeks (base training) with progressive training volume + 1 week recovery; Intervention: pretest + 15 days training (training load was organized into four training zones depending on the intensity and	Primary: VO2max (maximal treadmill test-direct measurement). Secondary: basal HR, peripheral oxygen saturation	VO2max: intra-group significant changes in H-HRV ($3.8 \pm 3.1\%$). Basal HR: inter-groups significant differences (H-HRV = 55.38 ± 10.02 vs H = 55.59 ± 4 bpm; H-HRV = $55.38 \pm$	Selection Performance Detection Attrition	High High High Low	Allocation based on the results of a laboratory test or a series of tests No blinding of participants and personnel The study did not address this outcome No missing outcome data.

normobaric hypoxia group (n = 9); N, normoxia group (n = 6).	quantified as in Mujika et al. (1996), adapted to Nordic skiing (the threshold for training adjustment was chosen as 30% of the mean of the previous day) + posttest1 + 1 week + posttest2. Similar training content each group.	(SpO ₂), RR interval with heart rate monitor, VO ₂ at the second ventilatory threshold.	10.02 vs N = 47.11 ± 6.21bpm). SpO ₂ : inter-groups significant differences (H-HRV = 90.4 ± 1.3 vs N = 94.2 ± 0.8%). R-R interval: no significant differences inter-groups (H-HRV = 9561.10 ± 9436.02ms ² ; H = 12199.41 ± 1293.14ms ² ; N = 7441.2 ± 4954.16ms ²).	Reporting	High	Not all of the study's pre-specified primary outcomes have been reported. The study appears to be free of other sources of bias
Location: French National Ski-Nordic Centre. Recruitment: members of the cross-country ski and Nordic combined French. Inclusion criteria: elite Nordic-skiers. Exclusion criteria: history of altitude related sickness and health risks that could compromise the subject's safety during training and/or hypoxic exposure.	H-HRV group: sleeping normobaric in hypoxia (simulated altitude of 2700 m) with HRV-guided training; daily hypoxic dose was similar between H-HRV and H; Night SpO ₂ was similar between H-HRV and H, but lower than in N. H: traditional training sleeping in hypoxia (simulated altitude of 2700 m). N: traditional training sleeping in normoxia. Follow-up (post test21 after 3 weeks end posttest1)	Others: duration of hypoxic exposure, HR, blood parameters (erythrocyte concentration, haemoglobin, haematocrit, ferritin), questionnaire of overtraining.	VO ₂ second VT: intra-groups significant changes for H-HRV (6.7 ± 6.1%).	Other	Low	

Vesterinen_2016	Randomized controlled trial	Recreational endurance runners (men = 20; women = 20) N = 40: EXP = 20 + TRAD = 20 Location: not specified. Recruitment: advertisement and social media Inclusion criteria: 2 years regular endurance running training. Exclusion criteria: disease or regular medication for chronic or long term diseases.	12 weeks (4 weeks of preparation + 8 weeks of training). The same volume as before de study for PREP and the same volume as for PREP for INT. EXP: Training MICT and HIIT according to HRV. TRAD: 50% sessions at low intensity and 50% sessions at moderate/high intensity. Week periodization 3:1. No follow-up periods. 9 withdrawals: sicknesses (n=2), injuries (n=2), lack of adherence (n=5)	Primary: VO _{2max} (maximal treadmill test-direct measurement). Secondary: Speed in Lactate 1, speed in Lactate 2, mean speed in 3000 test, time in 3000 test.	VO _{2max} : significant intra-group improvements (EXP = 3.7 ± 4.6%, TRAD = 5.0 ± 5.2%). Speed in L1 significant intra-groups improvement in EXP (2,8 ± 3,7 %). Speed in L2 significant intra-groups improvement in EXP (2,6 ± 3,3 %) and TRAD (1.9 ± 2.2%). Time in 3000 test: significant intra-group improvement in EXP (-14.3 ± 14.1s)	Selection Performance Detection Attrition Reporting Other	High High High High Low	Allocation based on the results of a laboratory test or a series of tests No blinding of participants and personnel The study did not address this outcome High rates of loss to follow up. Not all of the study's pre-specified primary outcomes have been reported. The study appears to be free of other sources of bias
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Excluded studies

As indicated in Figure 1, four studies were excluded from the qualitative analysis. Three studies were excluded because the VO_{2max} was not considered as an outcome [24,25,34], and one study was excluded because it was not an RCT [35].

Risk of bias in the included studies

The risk of bias in the included studies is summarized in Table 2. This assessment has been made following Cochrane Collaboration guidelines [30]. In addition, publication bias was assessed using a funnel plot (Figure 2). The Egger test provided statistical evidence of funnel plot symmetry, suggesting the absence of a significant publication bias.

Table 2. Risk of bias in included studies

Study	Risk of Bias Domains						Overall risk of bias
	Selection	Performance	Detection	Attrition	Reporting	Other	
Javaloyes_2019	Unclear	High	High	Low	Unclear	Low	Unclear
Kiviniemi_2010	Unclear	High	High	High	Unclear	Low	High
Nuuttila_2017	High	High	High	High	Unclear	Low	High
Schmitt_2018	High	High	High	Low	High	Low	High
Vesterinen_2016	High	High	High	High	High	Low	High

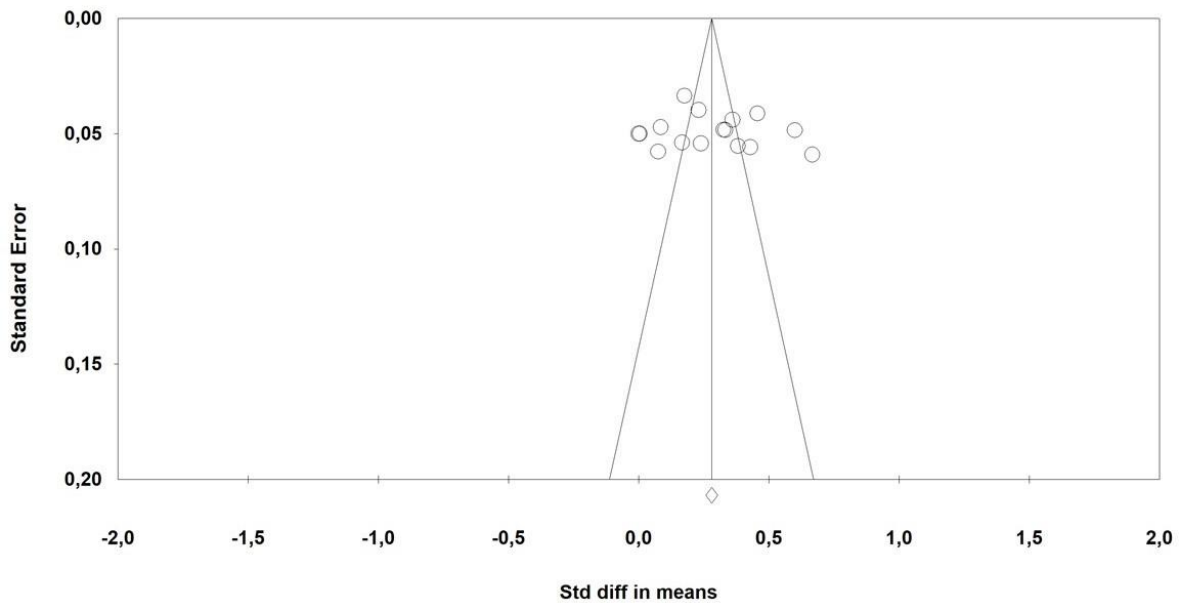


Figure 2. Funnel plot of standard error by standard differences in means.

- *Selection bias*

In Javaloyes_2019 and Kiviniemi_2010, neither the random component in the sequence generation nor the allocation concealment were described; therefore, the risk-of-bias selection was considered unclear. In Nuuttila_2017, Schmitt_2018 and Vesterinen_2016, the risk of bias was considered high because the randomization sequence was, in the first stage, based on the results of certain physical condition tests, the sport discipline, the age or the gender. Furthermore, in the second stage, the random component or the allocation concealment was not described.

- *Performance and detection bias*

Performance bias was universally high due to the nature of the interventions; this was because it was impossible to blind the participants from their treatment group allocation.

The detection bias was considered unclear in all of the included studies because they did not address this outcome.

- *Attrition bias*

In Javaloyes_2019 and Schmitt_2018, the attrition bias was considered low because there were no missing outcome data. On the other hand, Kiviniemi_2010, Nuuttila_2017, and Vesterinen_2016 presented high rates of follow-up loss for different reasons; these might be relevant in the ES observed. Moreover, no statistical procedure, such as intention to treat, was used to minimize this risk of bias. Therefore, they were considered as having a high risk of attrition bias.

- *Reporting bias*

The study protocols for the included studies were not available. Accordingly, Javaloyes_2019, Kiviniemi_2010, and Nuuttila_2017 were considered as having an unclear reporting bias due to the fact that all the pre-specified outcomes were reported. For their part, Schmitt_2018 and Vesterinen_2016 did not report every outcome result and were thus considered as having a high risk of reporting bias.

- *Other biases*

The included studies appear to be free from other sources of bias.

Synthesis of results

The Kiviniemi_2010 and Schmitt_2017 studies were segmented for quantitative analysis according to their intervention groups. The comparisons were: Kiviniemi_2010 a, HRV-1 (male subgroup, HRV-guided training) vs ST (standard training); Kiviniemi_2010 b, HRV-1 (male subgroup, HRV-guided training) vs CG (control group); Kiviniemi_2010 c, HRV-I (female subgroup, HRV-guided training) vs ST (standard training); Kiviniemi_2010 d, HRV-I (female subgroup, HRV-guided training) vs CG (control group); Kiviniemi_2010 e, HRV-II (female subgroup, HRV-guided training tailored for women) vs CG (control group); Kiviniemi_2010 g, HRV-II (female subgroup, HRV-guided training tailored for women) vs HRV-I (female subgroup, HRV-guided training); Schmitt_2017 a HRV (HRV-guided training) vs N (traditional training and normoxia sleeping); Schmitt_2017 b HRV (HRV-guided training) vs H (traditional training and

hypoxia sleeping). Therefore, the total number of individual studies analysed were 17 (k = 7 for the experimental group; k = 10 for the control group).

- *Primary outcome Measures*

There were four studies (Kiviniemi_2010, Nuutila_2017, Schmitt_2017 and Vesterinen_2016) with significant intra-group VO_{2max} improvements in the HRV-guided training group (n = 85), while no significant changes were found in Javaloyes_2019 (n = 9). On the other hand, in three studies (Kiviniemi_2010, Nuutila_2017 and Vesterinen_2016), there were also significant intra-group VO_{2max} improvements in the control group (n = 47). The overall risk of bias was considered high in every study but for Javaloyes_2019, which was considered unclear. A random effects meta-analysis of the five studies revealed a statistically significant ($p < 0.0001$) treatment effect for VO_{2max} in the HRV-guided training intervention (ES = 0.391; 95%CI = 0.265,0.517). Moreover, the other or no training intervention was also statistically beneficial ($p < 0.0001$) for VO_{2max} improvements in the control group (ES = 0.216; 95% CI = 0.110,0.321). However, the ES for the VO_{2max} was significantly higher ($p < 0.0001$) in the HRV-guided training group. The heterogeneity observed in the meta-analysis was significant and high in the overall analysis ($p < 0.0001$; $I^2 = 93.07\%$) and for the experimental ($p < 0.0001$; $I^2 = 94.08\%$) and the control group ($p < 0.0001$; $I^2 = 89.30\%$) (Figure 3).

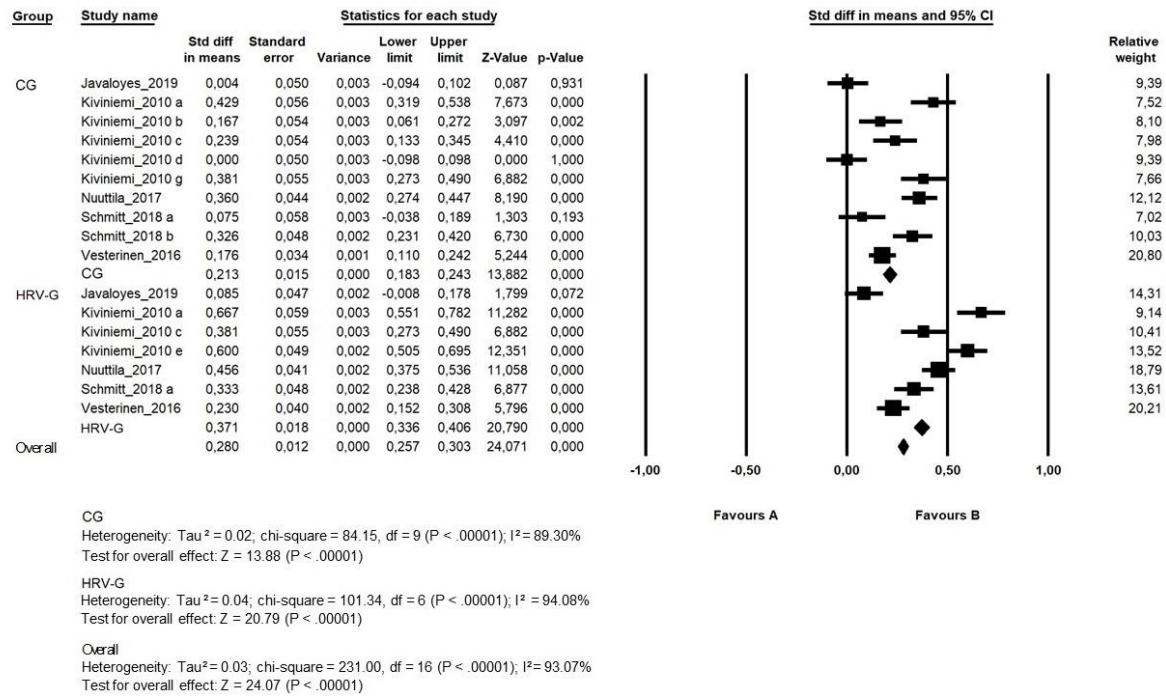


Figure 3. Standard differences in means (SDM) between post and pre measures for VO_{2max} in included studies, segmented by control group (CG) and heart rate variability guided training group (HRV-G). Squares represent the SDM for each trial; the diamond represents the pooled SDM across trials; weight determines how much each individual study contributes to the pooled estimate; 95%CI, confidence interval.

- *Moderator analyses*

Owing to the high heterogeneity observed in the meta-analysis, the potential moderating effect of the following were considered to be of interest: a. the athletes' level (elite vs amateur) and b. the sex of the participants ('men vs women' vs 'men and women'). We had originally planned to take into account the intervention duration but it was not finally included as a subgroup owing to there being only one study that considered an intervention period of 15 days (Schmitt_2017) while the others conducted an 8-week intervention. The sample size was used

for the meta-regression. Following the moderating variables (Table 3), the athletes' level (elite vs amateur) brought about statistically significant improvements ($p < 0.0001$) in both subgroups, while there were statistically significant differences between the subgroups ($p < 0.0001$) in favour of the non-professional subgroup (elite, ES = 0.17; amateur, ES = 0.32). According to the sex subgroups ('men vs women' vs 'men and women'), there were statistically significant improvements ($p < 0.0001$) in the three subgroups, and statistically significant differences ($p = 0.002$) between the three subgroups in favour of the women (men, ES = 0.30; women, ES = 0.32; men and women, ES = 0.23). The meta-regression findings (Figure 4) revealed that the sample size of the studies was not directly related to the ES magnitude (Regression coefficient = 0.003; Standard error = 0.003; Lower limit = 0.010; Upper Limit = 0.004; Z-value = -0.84; $p = 0.403$).

Table 3. Subgroup analyses for measuring their impact on VO_{2max} .

Group	Research Studies		Variable: VO_{2max}			
	N ^o Studies	References	SMD (95%CI)	I ²	p	p-difference
<i>Athlete level</i>						
Elite	3	Javaloyes_2019; Schmitt_2018 a; Schmitt_2018 b	0.17 (0.03; 0.30)	89.63	0.000	0.000
Amateur	8	Kiviniemi_2010 a; Kiviniemi_2010 b; Kiviniemi_2010 c; Kiviniemi_2010 d; Kiviniemi_2010 e; Kiviniemi_2010 g; Nuuttila_2017; Vesterinen_2016	0.34 (0.24; 0.44)	93.14	0.000	
<i>Sex</i>						
Women	4	Kiviniemi_2010 c; Kiviniemi_2010 d; Kiviniemi_2010 e; Kiviniemi_2010 g	0.32 (0.12; 0.53)	94.93	0.000	0.002
Men	4	Javaloyes_2019; Kiviniemi_2010 a; Kiviniemi_2010 b; Nuuttila_2017	0.31 (0.14; 0.47)	95.07	0.000	
Men and women	3	Schmitt_2017 a; Schmitt_2017 b; Vesterinen_2016	0.23 (0.15; 0.31)	77.93	0.001	

Note: SMD, standard mean difference; CI, confidence interval; VO_{2max} , maximal oxygen uptake; I^2 = I-squared.

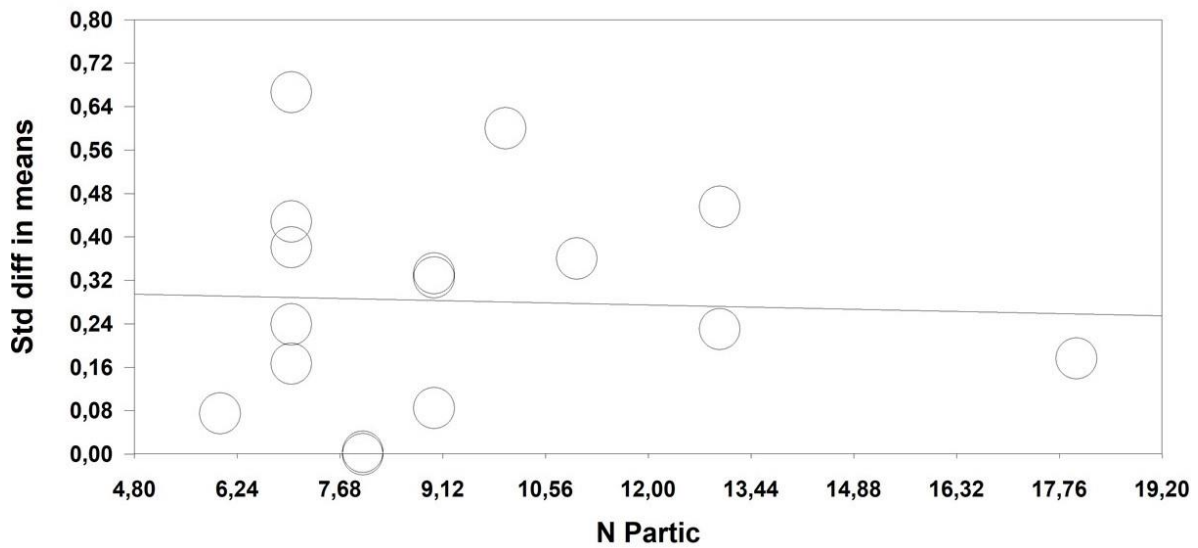


Figure 4. Meta-regression of number of participants (N Partic) on standard differences in means (Std diff in means).

Discussion

Summary of main results

Five RCT studies evaluating the effects of an HRV-guided training intervention on endurance athletes were included in this review. The results of the meta-analyses provide some evidence that either HRV-guided training or traditional training may improve their performance in terms of VO_{2max} (HRV-G: ES = 0.391, $p < 0.0001$; CG: ES = 0.216, $p < 0.0001$). However, more favourable outcomes ($p < 0.0001$) for the experimental groups compared to the control groups were recorded across the studies. Moderators indicated larger effect sizes for interventions involving amateur endurance athletes (ES = 0.32, $p < 0.0001$) and women (ES = 0.32, $p < 0.0001$). On the other hand, the sample size of the studies was not directly related to the ES magnitude ($p = 0.403$).

Overall completeness and applicability of the evidence

The total sample size of the studies meeting our original inclusion criteria was sufficiently large to warrant restricting the results to a meta-analysis of the RCTs. Data on the

primary outcome (VO_{2max}) were measured directly using a gas exchange analysis system and a maximal test in each study; this is the most accurate way to obtain cardiorespiratory data. However, some studies implemented this test using a treadmill (Nuutila_2017, Schmitt_2017 and Vesterinen_2016) and others, using a cycle ergometer (Javaloyes_2019 and Kiviniemi_2010). In the first case, training was based on running (Nuutila_2017 and Vesterinen_2016) and skiing (Schmitt_2017), which implies similar technical execution in the test. In the second case, the Javaloyes_2019 study was carried out on cyclists, whereas the Kiviniemi_2010 study sample was composed of runners. Despite statistical improvements regarding VO_{2max} in the Kiviniemi_2010 study, the specificity of the test may be a source of variability, and of potential imprecision, in the results. Following the training specificity principle [36], the body's physiological and metabolic responses and training adaptations are specific to the type of exercise and the muscle groups involved. Thus, the evaluation method should be as similar as possible to the training in order to obtain the most reliable results. This needs to be taken into account when interpreting the results.

Despite the intervention durations being quite homogeneous in the included studies (eight weeks for each study apart from Schmitt_2017), the total duration of the training process, the preparation weeks included, the endurance sport modality and the training intensities used for the control group (standard training) were different. There was also a marked heterogeneity in the sample of the included studies: elite (Javaloyes_2019 and Schmitt_2017) and amateur (Kiviniemi_2010, Nuutila_2017 and Vesterinen_2016) participants, or samples comprising only men (Javaloyes_2019, Kiviniemi_2010 and Nuutila_2017), only women (Kiviniemi_2010), or men and women (Schmitt_2017 and Vesterinen_2016). A standardized training protocol should be recommended to ensure the optimal benefits regarding VO_{2max} .

Quality of the evidence

The quality of the evidence from the included studies can be considered high. Despite each study being a randomized controlled trial, the sequence generation or the allocation concealment was considered skewed in most of them. The performance and detection biases were high in all the studies because incomplete blinding was considered. Attrition was high in most of studies because of the high follow-up rates. In addition, the reporting bias was generally unclear due to the fact that all the pre-specified outcomes were reported in most of the studies.

Potential biases in the review process

Although the systematic nature of the review process followed here decreases the potential for bias, the risk of bias in the review process remains. The greatest risk of bias present in this review was the study selection, specifically, the decision to limit the inclusion criteria to individual endurance sports, thus reducing the number of studies included and causing a potential limitation in the results.

Agreements and disagreements with other studies or reviews

Based on the results from this systematic review with meta-analysis, and in response to Research Question 1, it is not surprising that the meta-analysed results regarding improvements in athletes' VO_{2max} was associated to both training methodologies. According to Bartlett, O'Connor, Pitchford, Torres-Ronda, and Robertson [37] and Heyward [36], adequate prescribed training should maximize athletic performance when the specificity, overload, progression, initial level, individualization, diminishing return and reversibility principles are followed. However, it was also found that the individual training adaptation according to the endurance athletes' daily HRV scores produced better VO_{2max} results than the standardized prescribed training - which answers Research Question 2. As pointed out by Vesterinen et al. [4,9], not every athlete improves their VO_{2max} after standardized group training. Similarly, Gallo,

Cormack, Gabbett, Williams, and Lorenzen [38] reported that, in footballers, the internal load (perceived effort) of each athlete was different for a given external load; this definitely affects their individual performance during training and will be reflected in their individual performance improvements. Thus, daily individual HRV monitoring and training guidance balancing the sympathetic and parasympathetic autonomic nervous system leads to greater athletic performance in endurance athletes compared to standardized prescribed training. This is relevant if training optimization is the objective, supporting the idea that training should be prescribed appropriately to avoid overtraining and/or injury [37]. In the same vein, it is also interesting to point out that, according to studies such as Hulin, Gabbett, Lawson, Caputi, and Sampson [39] and Williams et al. [16], training individualization is also related to minimising overuse and reducing the injury risk, which may be a correlative benefit in the pursuit of endurance athlete training optimization.

On the other hand, the meta-analysed results show that VO_{2max} improvements were greater when the sample comprised amateur endurance athletes - this answers Research Question 3. According to the initial training level principle [36], individuals with a low initial level of physical fitness should achieve more significant relative increases than those of average or high levels. This is in accordance with the results of Sanchez-Sanchez et al. [40], where greater performance improvements were obtained in lower-level football players compared to the higher-level players, concluding that the lower the athlete's initial fitness level, the higher the available window of adaptability. Conversely, in the systematic review with meta-analysis by Hammami, Gabbett, Slimani, and Bouhlef [41], the athlete's level was not a determinant variable in terms of VO_{2max} enhancement, since it improved whether they were elite or amateur players. It should be noted that this review was conducted on football players, and that randomized and non-randomized controlled trials were included.

According to our meta-analysed results, and in response to Research Question 4, there

were higher effect sizes regarding VO_{2max} improvements when the sample was not mixed, especially in the case of women. There is controversy concerning the influence of sex in sport performance. Recent studies conducted on endurance athletes concluded that either sex was not a predictor variable of performance [42] or that performance between men and women was different in swimming, cycling and running [43]. In the case of the present systematic review with meta-analyses, we consider that the initial level of the sample influenced the result, given that, in the Kiviniemi_2010 study, where female samples were analysed, the participants were amateur level athletes. Thus, a higher relative performance increment is predictable based on the athletes' level.

Authors' Conclusions

Practical implications

Training optimization to enhance performance in endurance athletes is a goal that is undergoing a constant process of improvement. Finding a procedure to objectively individualize the training would be ideal for achieving this goal. The meta-analyses results considered in this review suggest that HRV is a good indicator of physiological responses to training in endurance athletes. Consequently, using daily HRV scores for training individualization and prescription is an effective method for optimizing performance in endurance athletes. This is reflected in the improved VO_{2max} results when the training is guided by HRV, considering VO_{2max} as one of the main performance indicators. In addition, it should be taken into account that a lower initial athlete fitness level will be relevant in achieving greater VO_{2max} improvement. Although gender may be a variable that influences the performance gains, in our opinion, this result is primarily conditioned by the level of the athletes included in the analysed studies. Therefore, we do not consider it to be a variable that clearly affects VO_{2max} improvements.

Research implications

The results from this review suggest that, while there is evidence that HRV-guided training is effective at improving VO_{2max} in endurance athletes, there is still work to be done in terms of identifying the characteristics of the interventions that contribute to this effectiveness and to identify the characteristics of participants who are more likely to respond to such interventions. The most important point is that more research is required since only five studies were included in this review. Moreover, only two of the studies used samples composed of elite endurance athletes, and these gave different results regarding VO_{2max} improvement. Consequently, the research should be extended to the professional field in order to clarify the effect of guiding training on VO_{2max} . This would also help to clarify whether the endurance sport modality is determinative of the VO_{2max} enhancement when following this training methodology.

Using daily HRV scores to control the training load and intensity over eight weeks is enough to improve VO_{2max} in endurance athletes. Nonetheless, the training protocol should be further standardized in terms of adjusting the number of preparation weeks or considering the measurement weeks within or around the training period - factors that determine the training duration. Moreover, the standardized training protocol used in the control groups varied between the studies, which considered low, moderate or high training intensities, as well as different numbers of sessions per week and session durations. This might very well have influenced the VO_{2max} results. Therefore, it is necessary to reach a consensus regarding a standardized training protocol to use in future studies. Similarly, although each study in this review used the most accurate method available to obtain the cardiorespiratory data, in future we should consider using a measuring instrument that allows us to implement the most specific sport technique in order to minimize result variability and imprecision.

Regarding the quality of the studies, authors should consider: improving the sequence

generation or allocation concealment, the blinding of the participants, personnel and outcome assessors, the rates of follow-up loss, using statistical procedures such as intention-to-treat to minimize attrition bias, and registering their protocols before starting the randomized controlled trial.

Lastly, to reinforce knowledge regarding performance optimization in endurance athletes, a good way to supplement the effect of HRV-guided training might be to register the risk of injuries associated to overuse using tools such as the Oslo Sports Trauma Research Center Overuse Injury Questionnaire, since this considers additional aspects affecting the execution of athletes' training.

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Publicación 2:

**“HRV-Guided Training for
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Protocol for a Cluster-Randomized
Controlled Trial”**

Estudio 2

HRV-Guided Training for Professional Endurance Athletes: A Protocol for a Cluster-Randomized Controlled Trial

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HRV-guided training for professional endurance Athletes: A protocol for a cluster-randomized controlled trial

Abstract

Physiological training responses depend on sympathetic (SNS) and parasympathetic nervous system (PNS) balance. This activity can be measured using heart rate variability (HRV). Such a measurement method can favor individualized training planning to improve athletes' performance. Recently, HRV-guided training has been implemented both on professional and amateur sportsmen and sportswomen with varied results. There is a dearth of studies involving professional endurance athletes following a defined HRV-guided training protocol. The objectives of the proposed protocol are: (i) to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period and (ii) to determine differences in the athletes' performance after following both training protocols. This will be a 12-week Cluster-Randomized controlled protocol in which professional athletes will be assigned to an HRV-based training group (HRV-G) or a traditional-based training group (TRAD-G). TRAD-G will train according to a predefined training program. HRV-G training will depend on the athletes' daily HRV. The maximal oxygen uptake (VO_{2max}) attained in an incremental treadmill test will be considered as the primary outcome. It is expected that this HRV-guided training protocol will improve functional performance in the high-level athletes, achieving better results than a traditional training method, and thus providing a good strategy for coaches of high-level athletes

Keywords: HRV; endurance training; training performance; high level athletes; VO_{2max} ; running.

Introduction

It is known that training is essential for improving physical performance [1] and that optimizing training for performance improvement in athletes is an important area of research within exercise physiology and sports medicine [2–4]. In this regard, different training methods for performance improvement have been tried and tested, such as intensified training [2,5,6] and submaximal tests [7]. However, it is also recognized that using the same standardized training program for a group of athletes can provoke a wide range of reactions in terms of performance and physiological adaptations [8,9].

As stated by Schmitt, Willis, Fardel, Coulmy, and Millet [10], an important component of the interindividual variability in physiological responses to standardized training is related to the balance between the parasympathetic (PNS) and sympathetic (SNS) activity of the autonomic nervous system (ANS) [11,12]. Heart rate variability (HRV) is one of the indicators that allows the noninvasive study of autonomic nervous system activity in its sympathetic and parasympathetic branches [13,14]. HRV is understood as the variation in the time interval between two consecutive heartbeats. It is obtained by calculating the time interval that elapses between two consecutive R waves (i.e., RR interval fluctuation) on an electrocardiogram (ECG) [15]. The period between beats is not constant; consequently, high HRV values are associated with an efficient ANS, which promotes behavioral adaptation and cognitive flexibility during stress [16], whilst low HRV values are indicative of an inefficient ANS, resulting in maladaptive responses to stress and perceived threats [15]. Furthermore, HRV is considered to be an indicator of cardiovascular health level [17].

Given that the SNS is responsible for changes in heart rate (HR) due to stress, and that the HR is one of the first parameters used to control the body's functional capacity [18], HRV analysis has been established as a useful method for assessing the heart's ability to adapt to both endogenous and exogenous loads [19], and can be used for the individual assessment of

responses to training loads. Indeed, in recent years, HRV has been used to analyze these imbalances between SNS and PNS in athletes [20] and to evaluate different aspects related to training [21] such as exercise intensity and duration [22], recovery and overtraining [23], training load [24] or psychophysiological profiles [25].

The control of training based on HRV, as an indicator of the precompetitive physical and psychological state in athletes, enables coaches and scientists to use these HRV records to adapt the recovery and training loads to each athlete in search of a better sports result. As indicated by Ortigosa-Márquez, Reigal, Carranque, and Hernández-Mendo [26], high HRV values indicate more parasympathetic than sympathetic activation in an athlete and, therefore, better recovery and preparation for dealing with high-intensity training sessions.

Traditionally, HRV has been measured with ECG [14]. One of the ways of quantifying HRV is through rMSSD (the root mean square of successive differences between adjacent RR intervals) [26] since it is a temporal statistical parameter that reports those variations occurring over the short term between RR intervals [27] and it is used to observe the influence of the SNP on the cardiovascular system [18]. Currently, there are other validated tools for determining HRV that facilitate measurement, such as the Kubios HRV, Elite HRV, Mobile Lab and HRV4Training applications (APPs).

In recent years, experimental studies have been carried out evaluating HRV-guided training in endurance athletes. These studies have been conducted both on elite athletes in sports such as cycling [24] and skiing [10], and with amateur endurance athletes [28–31]. One should take into account the scarcity of studies that have been published to date, especially on elite-level endurance athletes, as well as the absence of a common protocol to follow in this type of research.

The protocol proposed in the present study contributes to the scientific literature in this field in several ways: (i) it proposes research focused on elite athletes, a sample population for

which only two experimental studies have been carried out to date; (ii) the protocol is intended to research endurance runners, for which we are not aware of any research having been carried out on this type of sample and with these characteristics at the international level. Therefore, it is a novel study aiming to provide empirical support for HRV-guided training in long-distance runners, adapting daily training to the physiological responses of each individual athlete. The performance of these athletes could be compared to that of another group of long-distance runners who carry out traditional training over the same period of time.

Until just a few years ago, conducting research of this type, involving daily HRV measurements on each athlete and then adapting training based on these data, was only possible with the collaboration of high-cost laboratories. This study tests the use of noninvasive, commercial, low-cost and publicly accessible technology to evaluate the physiological responses obtained by adapting training to HRV.

Based on everything described above, we hypothesize that HRV-guided training will: (i) improve functional performance in high-level athletes and (ii) produce better performance results than a traditional training method. The objectives of the proposed protocol are: (i) to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period, and (ii) to determine differences in the athletes' performance after following both training protocols.

This will be a 12-week cluster-randomized controlled trial protocol in which professional athletes are assigned to either an HRV-based training group (HRV-G) or a traditional-based training group (TRAD-G). A block randomization method will be chosen to randomly assign participants to interventions in equally sized sample groups. This protocol has been designed following the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) Statement [32]. To describe the intervention, the TIDieR (Template for Intervention Description and Replication) checklist by Hoffmann et al. [33] has been used.

Materials and Methods

Study Setting

To detect an intervention-related effect in professional athletes, other studies with similar protocols [24,31] compared athletes from two clubs or associations. Similarly, our sample will comprise athletes from two sport institutions in Almería (Spain): the C.D. Atletas de Almería, based in the city of Almería (Spain) and the Asociación Espeleológica Velezana, based in Vélez Rubio (Spain).

Elegibility Criteria

The inclusion criteria for participating in the program will be: (i) to belong to the Spanish Athletics Federation; (ii) to have been training and competing in Spanish Athletics Federation competitions for at least two years; and (iii) to be in the first third of the classification for the last five races of the previous season. Regarding the exclusion criteria, the following will be taken into account: (i) having cardiovascular pathologies, abnormal blood pressure parameters or diagnosed respiratory problems; (ii) being treated for psychological problems, or regularly taking a drug(s) that has a direct or indirect effect on the nervous system (e.g., anxiolytics, antidepressants or neuroleptics); (iii) substance use that is not permitted by the International Association of Athletics Federations (IAAF); (iv) occasional consumption of medication for a disease related to the cardiorespiratory system (e.g., influenza) that might alter performance and (v) not performing at least 90% of the workouts during the intervention.

The trial steering members will be responsible for checking that the subjects interviewed meet the inclusion criteria. The Spanish Athletics Federation's medical team will certify that the subjects do not meet any of the exclusion criteria. After being informed of the study design and potential risks, all athletes will sign a written informed consent document. The model consent form is shown in Appendix A.

Interventions

Based on the methodology used by Javaloyes et al. [24] and Vesterinen et al. [31], the intervention will be divided into two training periods for both study groups (HRV-G and TRAD-G): a four-week preparation period (PR) and an eight-week training period (TR). Both will maintain the weekly training volume. The training carried out will mainly be running. The PR period will be common to both groups and will be a familiarization phase for the training sessions and their intensities. During this period, the training intensity will gradually increase for the first three weeks and then decrease in the fourth week. This will mean three weeks of overloading and one week of recovery (3:1). The training to be carried out by the athletes is presented in more detail in Table 1. In the TR period, each group will carry out the corresponding intervention. The TRAD-G group will train according to a predefined training program, which will include sessions carried out at low intensity (approximately 50% of the total), and other sessions of moderate and high intensity, with a structure similar to that carried out during the PR period (Table 2). The training prescribed to the HRV-G group will depend on the subjects' HRV, in accordance with authors such as Javaloyes et al. [24], Kiviniemi, Hautala, Kinnunen, and Tulppo [34], and Lamberts, Swart, Noakes, and Lambert [35].

Table 1. Periodization and training distribution for the heart rate variability group (HRV-G) and traditional-based training group (TRAD-G) during the preparation period (PR).

Weeks	High Intensity	Moderate Intensity	Low Intensity
1		90 min between VT1 and VT2	3–4 sessions between 30 and 35 min below VT1
2	4x 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1

3	50 min at VT2	4 x 12min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
4				3–4 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions will be performed with a 15- to 20-min warm up and 20 min of cooling down.

Table 2. Periodization and training distribution for TRAD-G during training period (TR).

Weeks	High Intensity	Moderate Intensity	Low Intensity
5	50 min at VT2		3–4 sessions between 30 and 35 min below VT1
6		4 x 12 min > VT2/3-min rest	90 min between VT1 and VT2
7	50 min at VT2	4 x 12 min > VT2/3-min rest	90 min between VT1 and VT2
8			2–3 sessions between 30 and 35 min below VT1
9	50 min at VT2		90 min between VT1 and VT2
10			2–3 sessions between 30 and 35 min below VT1
11	50 min at VT2	4 x 12 min > VT2/3-min rest	90 min between VT1 and VT2
12			2–3 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions will be performed with a 15- to 20-min warm up and 20 min of cooling down. Approximately 50% of the total sessions will be at low intensity.

To quantify the HRV, a Smartphone application known as “HRV4Training” (see <http://www.hrv4training.com/>) will be used. This tool has been validated by Plews et al. [36], showing a low typical estimate error (CV% (90% CI) = 3.8 (3.1; 5.0)) and a clear electrocardiographical correlation ($r = 1.00$ (1.00; 1.00)). It provides the root mean sum of the successive differences between R – R intervals (rMSSD) data using photoplethysmography. rMSSD is more suitable and reliable than other indexes [13,37]; nonetheless, the HRV data will be transformed by taking the natural logarithm, thus allowing parametric statistical comparisons that assume a normal distribution. In this way, a 7-day rolling average will be calculated (LnrMSSD7-d). The PR period will be used as a standardized phase to obtain the baseline LnrMSSD7-d and its range of normality (upper and lower limits). Following the indications of Plews, Laursen, Kilding, and Buchheit [38], this will be calculated as the mean $\pm 0.5 \times$ SD. During the TR period, the LnrMSSD7-d will be calculated daily in order to adapt the training prescribed to the HRV-G athletes. Moreover, the range of normality will be updated weekly. If the LnrMSSD7-d is within the range of normality, the athletes will perform a moderate or high-intensity session. If the weekly LnrMSSD7-d average falls below the normal range, a low intensity workout or rest will be undertaken. Athletes will perform a maximum of two consecutive sessions of moderate or high intensity; likewise, they will not accumulate more than two consecutive rest sessions. The modified scheme of Kiviniemi et al. [34] presented in Figure 1 will be followed.

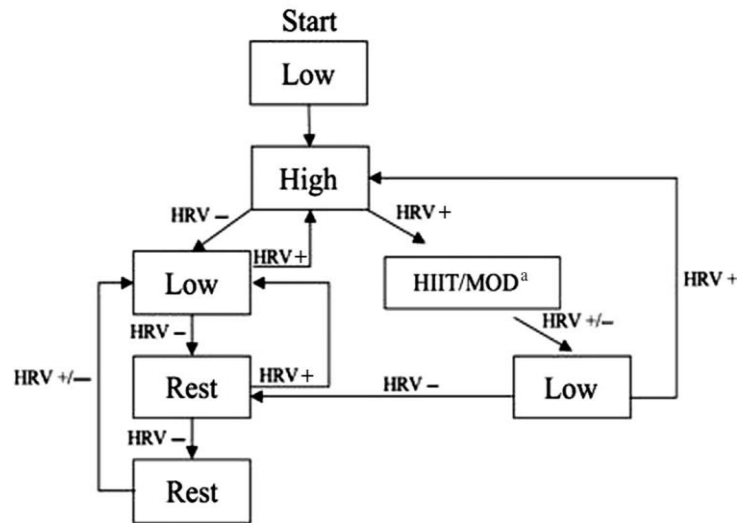


Figure 1. HRV-guided training schema. Modified from Kiviniemi et al. [34]. Note: When LnrMSSD7-d remained inside their normal range, high-intensity or moderate-intensity training sessions were prescribed. If LnrMSSD7-d fell outside their normal range (below), low intensity or rest were prescribed. HIIT/MOD = high/moderate-intensity interval training; HRV = heart rate variability; LnrMSSD7-d = 7-day rolling average of the natural logarithm of the root-mean-squared differences of successive RR intervals.

In accordance with Javaloyes et al. [24], all participants will be instructed to measure their HRV data at home each morning after waking up and emptying their bladders. They will be instructed to lie in a supine position and not perform any further activity during the recordings. Data will be recorded over a 60-s period [e.g., 36]. The daily control and recording of the rMSSD, as well as the LnrMSSD7-d calculation used to prescribe the training of the HRV-G athletes, will always be carried out by the trial steering members. These members will receive the information from each athlete via phone or email and, in turn, will inform the HRV-G coach of the training intensity corresponding to each athlete. This procedure will also serve as a strategy for maintaining and monitoring the athletes' adherence to the training programs. Concomitant care, or any other intervention, will not be allowed during the trial for either the

HRV-G or the TRAD-G. Athletes from both groups will carry out the training in their usual location.

Outcomes

The primary outcome of this study will be the maximal oxygen uptake (VO_{2max}) obtained in an incremental treadmill test. The secondary outcomes will be: the maximal speed in m/s, maximal heart rate, respiratory exchange ratio, ventilatory thresholds (VT1 and VT2) and their derived speed, heart rate, respiratory exchange ratio and VO_2 obtained in the incremental treadmill test. Other measurements considered as secondary outcomes will be: the time, speed, heart rate, rating of perceived exertion (RPE) and lactate in the 3000m running test. Body composition and rMSSD will be considered as other variables.

Measurements will be taken before and after the training period, which will correspond to weeks 5 (pretest) and 12 (post-test). Over the assessment weeks, care will be taken that participants do not carry out any high-intensity training sessions. Each assessment week will consist of two testing sessions with a 48-h recovery period. The first testing session will include maximal graded exercise test and body composition measurements. In the second testing session, athletes will perform a 3000m running test. The rMSSD will be measured daily, as explained in the intervention section.

The incremental treadmill test will be performed by the Physical Exercise and Human Performance Research Group at the University of Murcia (Spain). This is a more objective way of determining physical fitness and represents the maximal performance capacity of an individual [39]. First, with the athlete in the supine position, a cardiovascular examination will be carried out at rest by means of cardiac auscultation, blood pressure and an electrocardiogram (ECG). The electrodes for recording the ECG and heart rate will be kept in place throughout the test. The Cardioline Cube® electrocardiograph will be used. To perform the incremental treadmill test, the Runner srl (Cavezzo Italy) treadmill will be used, as it was in

other studies such as Ballesta-García, Martínez-González-Moro, Ramos-Campo, and Carrasco-Poyatos [40]. Similar to other studies, such as Nuutila et al. [30] or Vesterinen et al. [31], a prior 2-min aerobic warm up will be performed at 6 km/h. The test itself will start at a velocity of 7 km/h. The speed will be increased by 0.1 km/h every 6 s. The incline will remain at 1% throughout the test. The athletes will be encouraged to perform at maximum effort. The test will end when the subject can no longer run; the subject will indicate this with a hand gesture. The recovery phase will then begin at 4km/h for 3 min followed by rest for a further 2 min. The tests will be considered maximal and valid when the theoretical heart rate ($220 - \text{age}$) exceeds 85% and the respiratory exchange ratio (RER) is greater than 1.15 [41]. During the stress test, the subjects will breathe through a mask connected to a gas analyzer (Metalyzer 3b®, Cortex, Leipzig, Germany). All gas exchange parameters will be measured breath-by-breath and averaged every 30 s. The $\text{VO}_{2\text{max}}$ will be defined as the oxygen consumption plateau [42]. The aerobic (VT1) and anaerobic (VT2) thresholds will be determined. Before each test, the gas analyzer will be manually calibrated. The test's maximal speed (V_{max}), maximal heart rate (HR_{max}), and respiratory exchange ratio (RER) will be recorded. The V_{max} or HR_{max} will be defined as the highest speed, or heart rate, reached for a finished stage. The speed, heart rate, respiratory exchange ratio and VO_2 at each ventilatory threshold will also be recorded as VVT_1 , VVT_2 , HRVT_1 , HRVT_2 , RERVT_1 , RERVT_2 , VO_2VT_1 , and VO_2VT_2 , respectively. All tests will be carried out under similar environmental conditions (an ambient temperature of 20–22 degrees).

As in other studies [30,43], the 3000 m running test will be conducted individually on a 400 m outdoor running track. Participants will be instructed to run at their maximum speed. Before the test, a 15 min standardized aerobic warm-up will be performed, consisting of running at a low to moderate intensity. Capillary blood samples (5 μL) for blood lactate concentration analysis will be taken from the fingertip using a Scout+ analyzer (SensLab GmbH, Leipzig,

Germany). Lactate is considered a useful indicator for measuring the metabolic cost and intensity of effort in aerobic-anaerobic sports [44]. Following Ribas [45], it will be considered in this test to relate it to running intensity. Lactate samples will be taken at four different points in time, in accordance with Rodríguez and Valero [46]: (i) just before the test (Lactatepre), (ii) just after the test (Lactatepost), (iii) 3 min after the test (Lactatepost3) and iv) 5 min after the test (Lactatepost5). Other variables, such as heart rate, time, speed, and rated perceived exertion (RPE), will also be measured. Heart rate will be recorded at five different points in time: (i) just before the test (HRpre), (ii) just after the test (HRpost), (iii) 1 min after the test (HRpost1), (iv) 3 min after the test (HRpost3) and v) 5 min after the test (HRpost5). The time will be recorded every 1000m at three different points in time: (i) after running 1000 m, (ii) after running 2000 m and (iii) after running 3000 m (right at the end of the test). The speed will be calculated from these three points using the formula: speed = distance in m / time in seconds. The RPE will be measured at the end of the test using the modified Börg CR-10 scale of perceived exertion [47]. According to the authors, a 0 rating corresponds to rest; a 3 rating to moderate intensity; a 5 rating to hard intensity; a 7 rating to very hard intensity; and a 10 rating to maximal intensity. This tool has recently been determined as a stand-alone method for training load monitoring purposes in several sports and physical activities with men and women in different age categories (children, adolescents and adults) at various expertise levels [48].

Body composition will be analyzed just before the treadmill test using the InBody120 analyzer (Biospace Co. Ltd., Seoul, South Korea). Height will be measured using a measuring rod (Seca 213), and the body mass index (BMI) will be calculated according to the formula: $BMI = kg/m^2$.

The time schedule for enrolment, interventions and assessments is shown in Figure 2.

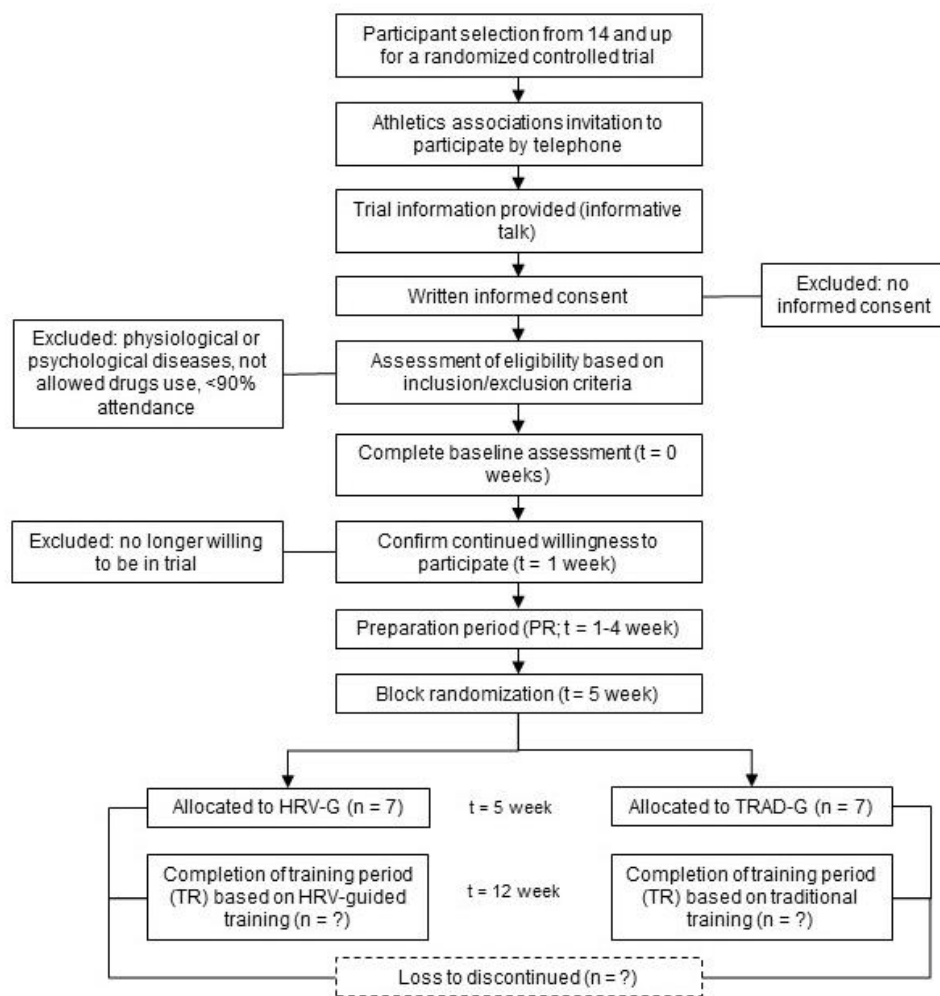


Figure 2. Schedule of enrolment, interventions and assessment. Note: HRV-G = heart rate variability-based training group; TRAD-G = traditional based training group; PR = preparation period; TR = training period.

Sample Size and Power

Calculations to establish the sample size will be performed using RStudio 3.15.0 software. The significance level will be set at $p \leq 0.05$. According to the mean standard deviation established for VO_{2max} in a previous study [31] ($SD = 1.5 \text{ mL/kg/min}$) and an estimated error (d) of 1.1, a valid sample size providing a 95% confidence interval (CI) in each group will

be 7 ($n = CI2 * d2/SD2$). Thus, a final sample size of 7 for each group will provide a power of 93% if between and within a variance of 2.

Recruitment

Each club or association involved in athletics in Almería (Spain) will be screened to identify the percentage of high-level or professionally federated athletes. When there are at least 7 high-level or professionally federated athletes, the club/association officers will be contacted by telephone to inform them of the study objective. Once they agree, an informative talk will be carried out with the athletes and the coach to inform them of the study objective, the time period in which it will take place, and the required commitment by the athletes to measure their daily HRV according to the established protocol, to attend the pre and post-test sessions and to attend at least 90% of the training sessions. The coaches will be informed of the required commitment to adapt each athlete's training session to the daily HRV score if their club is randomized into the HRV-G group. If they agree to participate, then they will have to sign the written consent and meet the eligibility criteria necessary to be recruited into the study. The recruitment process will be conducted by the trial steering members.

Allocation and Blinding

A block randomization method will be used to allocate participants to the groups, which will contain equal sample sizes. The block size will be determined by the data monitoring committee according to the statistical power provided. Blocks will be chosen randomly by tossing a coin to determine the participants' assignment into the groups. This procedure will be carried out by the data monitoring committee. The athletes and the data monitoring committee will be blinded to the exercise group assignment.

Data analysis

Data will be analyzed using Jamovi (Jamovi Project 2018, version 0.9.1.7, Sydney, Australia) and RStudio 3.15.0 software (PBC, Boston, USA). Prior to data analysis, the Shapiro

Wilk test and the Levene test will be performed to determine the normal distribution of the variables and the homogeneity of variance. Descriptive data will be reported as mean \pm SD and range. All the data will be analyzed based on the intention-to-treat principle (last observation carried forward). If the sample is normally distributed, Student's t-test will be calculated to compare variables before and after the intervention. For a variable to be considered as having a normal distribution, 95% of values will have to be within two standard deviations of the mean. If the sample is nonparametric, the U-Mann Whitney test will be used to compare variables before and after the intervention. The standardized mean differences (Cohen's effect size) will be calculated together with the 95% confidence intervals [49]. The effect sizes (ES) will be calculated using Cohen's d [49]. The relationship between variables will be assessed using the Pearson r correlation coefficient. If r is higher than 0.7, the determination coefficient (r^2) will be used to determine the percentage of Y variation with regard to the X variation. Significance will be accepted at $p \leq 0.05$.

Monitoring

A data monitoring committee will be set up during the study recruitment period. Interim analyses will be supplied to the committee in strict confidence, together with any other analyses that the committee may request. Based on the data monitoring committee's advice, the trial steering members will decide whether or not to modify the trial intake.

In our study, an adverse event will be defined as any untoward medical occurrence in a subject without regard to the possibility of a causal relationship. Adverse events will be collected after the subject has provided consent and enrolled in the study. If a subject experiences an adverse event after the informed consent document is signed (entry) but the subject has not started to receive study intervention, the event will be reported as not being related to the study's exercise program. For this study, the following will be considered serious adverse events: severe or permanent disability, use of prohibited substances and any other significant

hazard as determined by the study members. Serious adverse events occurring after a subject has stopped participating in the study will not be reported unless the researchers feel that the event may have been caused by the study protocol procedure.

Ethics and Dissemination

This protocol, the informed consent template contained in Appendix A and other requested documents (if any) will be reviewed and approved by the Bioethical Committee at the University of Almería with respect to the scientific content and compliance with applicable research and human subject regulations. Following initial review and approval, this protocol will be reviewed by the researcher at least once a year at Clinicaltrials.org, where it is registered with the ID: NCT04150952.

Any protocol modifications which might have an impact on conducting the study, potentially benefit a subject or affect a subject's safety, or change the study objectives, study design, subject population, sample sizes, study procedures, along with significant administrative issues, will require a formal amendment to the protocol. Such an amendment will be agreed upon by the Bioethical Committee at the University of Almería prior to implementation, and the clubs/associations enrolled will be notified. Administrative changes to the protocol that are minor corrections, and/or clarifications having no effect on the way the study is to be conducted, will be agreed upon by the researchers and documented in a memorandum. The Bioethical Committee at the University of Almería may be notified of the administrative changes.

All study-related information will be stored securely at the study site. All records that contain names or other personal identifiers, such as locator forms and informed consent forms, will be stored separately from the study records and identified by a code number. Forms, lists, logbooks, appointment books and any other listings that link participant ID numbers to other identifying information will be stored in a separate, locked file.

Discussion

This protocol describes the rationale, design, and methods of an HRV-guided training design for professional endurance athletes. It will allow accomplishment of a randomized controlled intervention to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period. Moreover, the differences in the athletes' performance after following both training protocols will be determined. To design this protocol with professional endurance athletes, the guidelines described in Kiviniemi et al. [34] have been followed. This procedure has also been adapted in other professional sports such as cycling [24,50] and skiing [10], as well as to amateur endurance athletes [28–31].

This is the first time that this kind of protocol will be applied in endurance elite athletes. After its implementation, we expect that both high-level athletes groups (HRV-G and TRAD-G) improve: (i) VO_{2max} and other secondary outcomes measured in the treadmill test (the maximal speed in m/s, maximal heart rate, respiratory exchange ratio, or ventilatory thresholds), (ii) the time, speed, heart rate, rating of perceived exertion (RPE) and lactate in the 3000m running test. Additionally, HRV-G will be better regarding performance results than the TRAD-G. These findings will suggest that training guidance balancing the sympathetic and parasympathetic autonomic nervous system leads to greater athletic performance in endurance athletes compared to standardized prescribed training. This is relevant for training optimization and for minimizing overuse and reducing injury risk.

Conclusions

Experimental research conducted in recent years shows that improvements in variables related with athletes' performance (e.g., VO_{2max}) can be obtained through HRV-guided training. However, accordingly to these studies, results do not allow a consensus to be established regarding the performance benefits of HRV-guided training for endurance athletes.

From studies carried out until now, this article describes a novel protocol to conduct a randomized controlled trial with endurance athletes. So far, no other HRV-guided training research has been conducted with these types of professional athletes. Besides, this protocol proposes to use emergent technologies in the training and the research fields, such as smartphone applications; in this case, HRV4training, an app scientifically validated that allows calculation of the daily HRV measurement for each athlete. Although more research is needed, the implementation of the protocol described here will contribute to this scientific field of study.

Author Contributions: Conceptualization, M.C-P, A.G-Q and A.G-G; methodology, M.C-P, A.G-Q, A.G-G. and I.M-G-M; investigation, M.C-P, A.G-G. and A.G-Q.; writing—original draft preparation, M.C-P. and A.G-Q.; writing—review and editing, A.G-G. and I.M-G-M. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

Mr. Antonio Granero-Gallegos and Ms. María Carrasco-Poyatos, lead researchers on the project: Physiological and psychological effects from heart rate variability-based training in professional athletes have informed:

Mr/Ms ID., about the present study's general proceeding, its objectives, duration, purpose, inclusion and exclusion criteria, associated risks and benefits, as well as the possibility of leaving it without having to give reasons. In knowledge of all the above, and the measures that will be adopted to protect the participant's personal data, according to the current regulations,

I CONSENT to participate in the present research.

Signed: Mr/Ms ID.

Signed: Mr. Antonio Granero-Gallegos, and Ms. María-Carrasco-Poyatos

Project Leading Research.

Antonio Granero-Gallegos

María Carrasco-Poyatos

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Publicación 3:

“HRV-GUIDED TRAINING IN
PROFESSIONAL RUNNERS: EFFECTS
ON PERFORMANCE AND VAGAL
MODULATION”

Estudio 3

HRV-guided training in professional runners: effects on performance and vagal modulation

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HRV-Guided Training in Professional Runners: Effects on Performance and Vagal Modulation

Abstract

Purpose: to analyze the training structure following a heart rate variability (HRV) -guided training or traditional training protocol, determining their effects on the cardiovascular performance of professional endurance runners, and describing the vagal modulation interaction.

Methods: This was an 8-week Cluster-Randomized controlled trial. Twelve professional endurance runners were randomly assigned to an HRV-guided training group (HRV-G; n=6) or a traditional training group (TRAD-G; n=6). The training methodology followed by the HRV-G was determined by their daily HRV scores. Training intensities were recorded daily. HRV4Training was used to register the rMSSD every morning and during a 60-second period. Cardiovascular outcomes were obtained through an incremental treadmill test. The primary outcome was the maximal oxygen uptake (VO_{2max}).

Results: total training volume was significantly higher in TRAD-G, but moderate intensity training was significantly higher in HRV-G ($X \pm SDDif = 1.98 \pm 0.1$ %; $P = 0.006$; $d = 1.22$) and low intensity training in TRAD-G ($X \pm SDDif = 2.03 \pm 0.74$ %; $P = 0.004$; $d = 1.36$). The maximal velocity increased significantly in HRV-G ($P = 0.027$, $d = 0.66$), while the respiratory exchange ratio increased in TRAD-G ($P = 0.017$, $d = 1$). There was a small effect on the LnRMSSD increment ($P = 0.365$, $d = 0.4$) in HRV-G. There were statistical inter-group differences in the Δ maximal heart rate when Δ LnRMSSD was considered as a covariable ($F = 7.58$; $P = 0.025$; $d = 0.487$). There were significant and indirect correlations of LnRMSSDTEST with VO_{2max} ($r = -0.656$, $P = 0.02$), Δ LnRMSSD with ΔVO_{2max} ($r = -0.606$, $P = 0.037$), and Δ LnRMSSDCV with Δ VENT ($r = -0.790$,

P=0.002).

Conclusions: higher HRV scores suggest better cardiovascular adaptations due to higher training intensities, favoring HRV as a measure to optimize individualized training in professional runners.

Keywords: endurance, professional, VO_{2max} , rMSSD, ventilatory threshold.

Introduction

The key to achieving optimal adaptations in endurance training is to find the most efficient stimulus [1]. The importance of macrocycle (weeks or months) training periodization is well-recognized in endurance training interventions. However, some experimental studies [2,3] highlight the relevance of microcycle (day-to-day) periodization in order to achieve better cardiovascular adaptations, especially when operating under the constraints of limited resources and time.

Monitoring the cardiac autonomic nervous system (ANS) is currently being used as a promising method to optimize the training prescription, as variability in physiological responses to standardized training is related to the balance between parasympathetic and sympathetic activity [4]. The parasympathetic branch of the ANS is of particular importance as it represents the body's integrative system for adaptively self-regulating and maintaining homeostatic balance [5]. In this regard, heart rate variability (HRV) is commonly used as an indicator of day-to-day autonomic nervous system activity. Higher parasympathetic activation is reflected in higher HRV values, which are indicative of better recovery [6]. Recently, validated smartphone tools have become available, such as HRV4Training, that have enabled day-to-day HRV recording, making it easier to adapt the training loads to each athlete for performance optimization.

According to Seiler et al. [7], the recovery of well-trained runners is better than that of novices since endurance training adaptations are associated with reduced sympathetic branch activation. Therefore, endurance training adaptations should induce higher HRV values. If the HRV is recorded daily, higher intensity sessions might be included in the training periodization, with the athletes benefiting from greater peripheral and central physiological adaptations associated to more polarized training. According to this assumption, a recent meta-

analysis 8 showed that the individual training adaptation, based on the endurance athletes' daily HRV scores, produced better VO_{2max} results than the standardized training prescribed, with the training level being a determinant factor.

However, when the sample is composed of runners, there is still a lack of consensus regarding the training design, when it is HRV-guided or traditionally prescribed [9,10] in terms of the total volume achieved and the proportion of training performed at high, moderate or low intensities. It is also unclear as to whether HRV-guided training induces better physiological performance, leading to a higher running velocity [9,2,10] or enhancing the oxygen consumption [2,3,9]. Moreover, HRV does not always improve statistically after an HRV-guided intervention [2,3,11,9] and its correlation to performance outcomes has only been analyzed in one study [10].

Therefore, the aims of the present study were: 1) to analyze the training structure (volume and intensity) performed by an HRV-guided training group and a traditional training group (HRV-G and TRAD-G), comprising professional endurance runners; 2) to determine the effect of the two training methods on the athletes' cardiovascular performance and vagal modulation; and 3) to describe the influence of HRV on the performance outcomes in both groups.

Materials and Methods

Design

This study was an 8-week Cluster-Randomized controlled trial in which professional endurance runners were assigned to an HRV-based training group (HRV-G; n=6) or a traditional training group (TRAD-G; n=6). The trial design followed the CONSORT guidelines [12]. The study was approved by the University of Almería's Bioethics Committee (UALBIO2019/026). The study was registered prospectively with ClinicalTrials.gov (NCT04150952).

Participants

The professional endurance runners were recruited from two sport institutions in Almería province (Spain): the C.D. Atletas de Almería, based in the city of Almería (Spain) and the Asociación Espeleológica Velezana, based in Vélez Rubio (Spain). The HRV data were collected in Almería whereas the incremental treadmill test data were collected at the University of Murcia (Spain). The inclusion and exclusion criteria are detailed elsewhere [13]. All participants signed a consent form before beginning the study.

Interventions

The training design followed in the preparation period (PR; the first four weeks) for both groups, or in the intervention period (TR; the following eight weeks) for the HRV-G and TRAD-G groups, are detailed in Tables 1-3. Moreover, the training prescription methodology followed by the HRV-G group, determined by their daily HRV scores and based on the Kiviniemi et al. [2] scheme modified by Javaloyes et al. [14], is detailed in the protocol of Carrasco et al. [13]. Figures 1 and 2 are an example of the HRV fluctuations during the TR period in HRV-G and TRAD-G, respectively.

Outcomes

The maximal oxygen uptake relative to weight (VO_{2max}) was considered as the study's primary outcome. The secondary outcomes recorded were: i) the maximal velocity scores (V_{max}), the heart rate (HR_{max}), the percentage of the heart rate relative to the maximum (HR_{rel}), the maximal oxygen uptake in liters per minute (VO_{2max}), the respiratory exchange ratio (RER), and the ventilatory rate (VENT). Other variables considered were the body mass index, the LnrMSSD, and the training volume at high (\geq Ventilatory Threshold 2, VT2, or Zone 3), moderate (between VT2 and VT1, or Zone 2) and low (\leq VT1 or Zone 1) intensities in the TR or PR periods.

The primary and secondary outcomes were obtained through an incremental treadmill

test. This test procedure and the instruments involved are explained in the Carrasco et al. protocol [13] together with the HRV and body mass index measurements. According to Piatrikova et al. [15], the weekly average for the LnrMSSD calculation was implemented with its coefficient of variation ($\text{LnrMSSD}_{\text{CV}} = [\text{LnrMSSD}_{\text{SD}}/\text{LnrMSSD}_{\text{MEAN}}] \times 100$) since it was also found to be a marker of day-to-day HRV variability and representative of training adaptation (a higher $\text{LnrMSSD}_{\text{CV}}$ was associated with a less optimal response). Moreover, the final test-day LnrMSSD was considered ($\text{LnrMSSD}_{\text{TEST}}$). The total training volume and the training volume at each intensity were recorded daily by the respective coaches.

The pre-test and post-test were conducted in weeks 5 and 12, coinciding with the first and the last weeks of the training period, in which low to moderate training sessions were carried out by the runners.

Sample size and power

According to the mean standard deviation established for the $\text{VO}_{2\text{max}}$ in our study ($\text{SD}=6.19$ ml/kg/min) and an estimated error (d) of 4.8, a total of six subjects was determined to be a valid sample size for each group, providing a 95% confidence interval (CI) ($n=\text{CI}2 \times d^2/\text{SD}^2$). Thus, a final sample size of 6 for each group provides a power of 88% if it is between and within a variance of 2. Calculations to establish the sample size were performed using RStudio 3.15.0 software. The significance level was set at $P \leq 0.05$.

Randomization and blinding

Participants were randomly allocated to each group using a block randomization method. The treatment was randomly assigned to the groups by coin tossing. The block size was determined according to the statistical power provided, making sure that the sample size was the same for each group (HRV-G, TRAD-G, $n = 6$). This process was implemented by the principal investigator. Participants and research staff were blinded. Only the coaches knew about the groups so that they could adapt, where appropriate, the training sessions to the daily

HRV recording.

Statistical methods

Prior to data analysis, the Shapiro-Wilk test was used to determine the normal distribution of the variables. Levene's test was also performed to determine the homogeneity of variance. Descriptive data are presented as mean \pm SD and range. The variables were normally distributed, so to test for differences between the groups at baseline, an unpaired two-tailed t-test was used. To compare the groups after the intervention, an analysis of covariance (ANCOVA) was used with the baseline values, while the final test-day LnRMSSD (LnRMSSD_{TEST}), the change in LnRMSSD (Δ LnRMSSD), and the change in its coefficient of variation (Δ LnRMSSD_{CV}), were included as co-variables in order to adjust for any potential influence in the dependent variables. To test for differences within groups, a paired two-tailed t-test was used. Owing to our small sample size, the standardized mean differences (Cohen's effect size) were calculated together with the 95% confidence intervals so as to describe if the treatment effect had a relevant magnitude. An effect size (ES) value of 0.20 indicates a small effect, 0.50 a moderate effect, and 0.8 a large effect [16]. A bivariate Pearson correlation was utilized to assess the relationships between the LnRMSSD and the physiological outcomes. The correlation thresholds were 0.1, small; 0.3, moderate; 0.5, large; 0.7, very large; and 0.9, nearly perfect [17]. The level of significance was set at $P \leq 0.05$. The statistical analyses were conducted using the IBM SPSS Statistics V.24 program for Windows (SPSS Inc., IL, USA), Microsoft Excel 2010 (Microsoft Corporation, WA, USA) and Rstudio 3.15.0 software.

Results

The participant flow as this randomized trial progressed is detailed in Figure 3. There were no losses or exclusions after the randomization process, thus 12 participants completed the study (HRV-G, $n = 6$; TRAD-G, $n = 6$). Recruitment was carried out in September 2019 and the intervention was conducted in the months of October and November 2019. There were no

special harm events that forced the intervention to stop. The baseline characteristics of the two groups are shown in Table 4.

Results for Objective 1: Regarding the training volume and intensity in the TR period, the HRV-G group accomplished a total of 46 hours, 17 minutes, and 24 seconds (46h 17' 24") distributed as 7h 43' 12" at \geq VT2, 17h 29' 24" between VT2 and VT1, and 21h 4' 48" at \leq VT1, which results in 16.68%, 37.77%, and 45.54% of the training time at the respective intensities. For its part, the TRAD-G group trained for 56h 46' 12" distributed as 9h 26' 24", 20h 19' 48", and 27h for the \geq VT2, between VT2 and VT1, and \leq VT1 intensities, respectively, which results in 16.63%, 35.81%, and 47.56% of the respective intensities. There were significant differences favoring TRAD-G regarding the total training volume ($X \pm SD_{Dif} = 629 \pm 12.96$ min; $P = 4.78 \cdot 10^{-7}$; $d = 6.57$ [3.71, 9.44]) and the training minutes at \geq VT2 ($X \pm SD_{Dif} = 103.17 \pm 7.05$ min; $P = 2 \cdot 10^{-6}$; $d = 5.63$ [3.1, 8.15]), between VT2 and VT1 ($X \pm SD_{Dif} = 170.83 \pm 19.36$ min; $P = 6 \cdot 10^{-6}$; $d = 4.98$ [2.69, 7.28]), and at \leq VT1 ($X \pm SD_{Dif} = 355 \pm 23.21$ min; $P = 3 \cdot 10^{-6}$; $d = 5.38$ [2.95, 7.82]) (Figure 4). On the other hand, the training time in Zone 2 was significantly higher in HRV-G ($X \pm SD_{Dif} = 1.98 \pm 0.1$ %; $P = 0.006$; $d = 1.22$ [-0.009, 2.46]), while TRAD-G spent significantly more time training in Zone 1 ($X \pm SD_{Dif} = 2.03 \pm 0.74$ %; $P = 0.004$; $d = 1.36$ [0.11, 2.62]) (Figure 5). There were also statistical differences between groups regarding the training time at moderate-to-high intensity (Zones 2 + 3) favoring HRV-G ($X \pm SD_{Dif} = 2.03 \pm 0.74$ %; $P = 0.004$; $d = 1.36$ [0.11, 2.62]).

Results for Objective 2: At the end of the intervention, there were no significant inter-group differences in the primary or secondary outcomes. In contrast, the V_{max} increased significantly in HRV-G ($P = 0.027$, $d = 0.66$), and the RER increased significantly in TRAD-G ($P = 0.017$, $d = 1$). Moreover, in HRV-G, there was a large effect in the VENT reduction ($P = 0.11$, $d = 1.01$) and a small effect in the VO_{2max} reduction ($P = 0.267$, $d = 0.32$) as well as an increment in the LnRMSSD ($P = 0.365$, $d = 0.4$). These results are shown in Table 5.

Results for Objective 3: The ANCOVA results showed that there were significant differences between groups in ΔHR_{max} ($F=7.58$; $P=0.025$; $d=0.487$) and ΔHR_{rel} ($F=8.47$, $P=0.02$, $d=0.514$) when the covariable was $\Delta LnrMSSD$, but also when the covariable was the baseline result (ΔHR_{max} : $F=5.646$, $P=0.045$, $d=0.414$; ΔHR_{rel} : $F=7.18$, $P=0.028$, $d=0.473$) (Table 6). On the other hand, a significant interaction of the $LnrMSSD_{TEST}$ with the post-test HR_{max} ($F=5.305$, $P=0.05$, $d=0.399$) and the VO_{2max} ($F=10.653$, $P=0.011$, $d=0.571$) was found, along with a significant interaction of $\Delta LnrMSSD_{CV}$ with $\Delta VENT$ ($F=11.663$, $P=0.009$, $d=0.593$), but these were not statistically different. Moreover, there were significant and indirect correlations of the $LnrMSSD_{TEST}$ with the post-test VO_{2max} ($r=-0.656$, $P=0.02$), the $\Delta LnrMSSD$ with the ΔVO_{2max} ($r=-0.606$, $P=0.037$), and the $\Delta LnrMSSD_{CV}$ with the $\Delta VENT$ ($r=-0.790$, $P=0.002$).

Discussion

The objectives of the present study were to analyze the training structure following an HRV-guided training or a traditional training methodology, determining their effect on performance in a sample of professional endurance runners, and describing the vagal modulation interaction. First of all, the total training volume and the time training in Zones 1, 2 and 3 were significantly higher in TRAD-G. Nevertheless, the proportion of training time at high intensity (Zone 3) was similar for both groups, but HRV-G spent significantly more time training at moderate intensity (Zone 2), and TRAD-G trained more in Zone 1. This training structure did not generate differences between groups but some performance aspects significantly changed in both groups, such as the maximal velocity or the respiratory exchange ratio, favoring the HRV-guided training. Moreover, there was a non-significant but small-to-high effect in the $LnrMSSD$ increment and in the VO_{2max} , and a decrease in the ventilation rate in HRV-G. These changes in $LnrMSSD$ had a significant impact on the maximal and relative heart rate, and were significantly correlated to the changes in VO_{2max} , meaning that an increase in cardiac vagal activation positively influences the endurance runners' cardiovascular performance.

Regarding the training structure, the HRV-G training was more efficient, resulting in less total training volume than in a traditional training methodology, as was reported by Vesterinen et al. [18]. In addition, higher proportions of moderate-to-high intensities (Zone 2 and Zone 2 + 3) were achieved by the HRV-G group while a higher proportion of low-intensity training (Zone 1) was achieved by the TRAD-G group. The distribution of volume and intensities in TRAD-G fitted with the traditional characteristics of elite endurance athletes' training [19]. On the other hand, the higher training intensities achieved by HRV-G show that their recovery was better than theoretically expected, as pointed out by Seiler et al. [7] and Hackney [20]. Thus, their LnrMSSD remained within the normal range more often, demonstrating the physiological adaptations associated to training, as can be seen in Figures 1 and 2. In this way, designing the training based on the daily HRV scores allows professional runners to train at higher intensities and lower volume than when following a traditional and theoretical training method. If intensity is the key to optimizing aerobic training [1], HRV-guided training may provide a practical tool for better adapting the training prescription to professional endurance runners, increasing the importance of microcycle (day-to-day) training periodization.

Regarding the training time at the different intensities, similar training proportions in Zones 1, 2 and 3 were found in the study by Javaloyes et al. [21] on professional cyclists. On the other hand, our results do not agree with those of Javaloyes et al. [14] since they found a higher proportion of low and high-intensity training in professional cyclists, and Vesterinen et al. [11] where similar proportion of times in the different training zones between groups were found in the recreational runners. This indicates that the focus of the training design determines the intensity of the training sessions when moderate or high-intensity sessions can be chosen. In this regard, a simpler schema to design the HRV-guided training could help to unify the interventions in professional athletes. In accordance with Seiler et al. [7], it seems that the recovery is essentially identical following a moderate-intensity training session or a session

over the VT 2 in highly trained endurance runners, while the athlete's level does affect the recovery. Therefore, including only high or low-intensity training options could be a more optimal procedure when the sample is composed of professional athletes.

Regardless of this, the training structure followed by HRV-G reported a significant increase in maximal velocity ($\Delta V_{\max}=0.68\pm 3.08$ km/h, $P=0.027$, $d=0.66$) and a small-to-high effect in the $VO_{2\max}$ ($\Delta VO_{2\max}=0.67\pm 7.58$ ml/kg/min, $P=0.267$, $d=0.32$), with a decrease in the ventilation rate ($\Delta VENT=9.52\pm 12.68$ bpm, $P=0.11$, $d=1.01$). These outcomes fit with certain physiological effects expected when training in Zone 2 [12]. Similarly, the training structure followed by TRAD-G is reflected in the significant increment in the RER ($\Delta RER=0.03\pm 0.001$ l, $P=0.017$, $d=1$) and in the low effect associated to the change in the other physiological outcomes measured. This is in accordance with the physiological effects generated by training in Zone 1, as it is sufficient to build a solid aerobic base; however, when the objective is to improve endurance performance, higher training velocities are necessary to stimulate the neuromuscular system to maintain higher competition velocities [1]. Therefore, the higher proportion of moderate-intensity training in HRV-G resulted in better physiological performance after the intervention than the higher proportion of low-intensity training followed by TRAD-G. These results are in the line with other similar studies carried out on professional or amateur athletes [2,3,10,14,8,21,23,18,24,25]. However, when a block periodization was followed in these studies, significant changes were found in variables such as the $VO_{2\max}$ or peak power in their HRV-guided training groups, or the V_{\max} in their traditional training groups. Therefore, including a higher proportion of high-intensity training might improve the physiological results.

Continuing with the intervention effect, and in accordance with other studies [15,2,3], there were no significant inter or intra-group changes regarding the $\ln rMSSD$. However, the moderate tendency for increased cardiac vagal activation found in the HRV-G at the end of this study was considered relevant as this could be related to this group's higher proportion of

training in Zone 2. According to López-Chicharro et al. [1] or Hackney [20], endurance training between the ventilatory thresholds 1 and 2 is related to a reduction in sympathetic activation. This assumption was proven in several experimental studies, such as Iellamo et al. [26] and Nuutila et al. [10]. Additionally, it seems that training below VT 1 (Zone 1) had no effect on the autonomic nervous system balance in highly trained runners [7]. On the other hand, the autonomic cardiac vagal modulation is unclear when the training intensity is increased below the VT 2 [26,7,27,3,28,10]. Thus, a large proportion of training at moderate intensity (Zone 2) had a moderate effect on the professional endurance runners' cardiac vagal activation increment, which could be enhanced if the training volume (for example, the duration of the intervention) were increased. Nevertheless, the effects that the major total training volumes or higher proportions of training in Zone 3 have on the LnrMSSD should continue to be studied.

In this regard, the change in cardiac vagal activation found after the intervention (Δ LnrMSSD) had a significant impact on our sample's maximal and relative heart rate change, which was significantly lower in HRV-G than in TRAD-G (Δ HR_{max HRVG}= 0.17 ± 26.69 bpm, Δ HR_{max TRADG}= -1.67 ± 7.9 bpm, $F=7.58$, $P=0.025$, $d=0.487$; Δ HR_{relHRVG}= -0.3 ± 14.81 bpm, Δ HR_{relTRADG}= -0.87 ± 4.17 bpm, $F=8.47$, $P=0.02$, $d=0.514$). Moreover, the change in the Δ LnrMSSD_{CV} influenced the change in the ventilation rate, which decreased in HRV-G with a large effect size despite the absence of significant differences between groups. Furthermore, significant indirect correlations were found between Δ LnrMSSD and Δ VO_{2max} ($r=-0.606$, $P=0.037$), between Δ LnrMSSD_{CV} and Δ VENT ($r=-0.790$, $P=0.002$), and even between LnrMSSD_{TEST} and posttest VO_{2max} ($r=-0.656$, $P=0.02$). This association between LnrMSSD and the performance variables was also found in other studies carried out with professional swimmers [15] and recreational runners [11,29,10], but the performance-related measure was the maximal speed, which represents a submaximal intensity outcome. However, the relevance of our findings can be found in the performance measures that represent the runners' maximal capacity (maximal

heart rate or VO_{2max}). As is shown by these results, the cardiac vagal activation was conditioning our runners' performance - the higher the change in LnrMSSD, the lower the change in maximal capacity outcomes. These results are in accordance with the assumption that increased cardiac vagal activation allows the training demands to be met with more stable physiological responses [1,22], which entails more optimal physiological responses to maximal effort. On the other hand, the baseline scores also had a significant impact on our sample's maximal and relative heart rate changes. According to the initial level training principle [30], the initial level of the HRV-G runners was somewhat better than that of the TRAD-G runners, even though there were no significant differences between groups. Therefore, the cardiac vagal activation impacts positively on the professional runners' performance, although the baseline scores should also be taken into account.

Limitations and practical applications

A polarized training method, combining high and low intensities, and/or longer interventions (>8 weeks), should be considered in further research. The athletes' level (professional or amateur) should be taken into account to homogenize the performance results. However, using the HRV4Training device allows coaches and athletes to directly collect day-to-day recovery data without compromising the data's validity. HRV measurements can optimize the adjustments made to the training process planning, reducing the burden of repetitive performance testing and adapting the training individualization to maximize the professional athletes' potential. According to the previously reviewed research, HRV monitoring will also help coaches in deciding whether or not to increase the sessions' training intensity, thus reducing the risk of overtraining. Recording the training load and HRV can also assist coaches and athletes in controlling other factors that influence stress recovery rather than reducing the training itself.

Conclusions

Using HRV as a daily guide to professional runners' training demonstrates that their recovery is better than expected, allowing them to achieve higher intensities at a lower training volume. However, polarized training, which includes only low or high training intensities, is recommended to strengthen cardiovascular performance and cardiac vagal modulation. Nevertheless, the increment in HRV scores suggest better cardiovascular adaptations, favoring HRV as a measure for optimizing training individualization and cardiovascular performance.

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Conclusiones / Conclusions

Conclusiones/ Conclusions

In this doctoral thesis three conclusions should be established in order to respond to each of the proposed research's objectives:

- Paper 1:
 - The results of the meta-analyses suggest that HRV is a good indicator of the physiological responses to training in endurance athletes. Thus, using daily HRV scores for training individualization and prescription is a good method for performance optimization in endurance athletes. This is reflected in the better VO_{2max} results when training is guided by HRV, considering VO_{2max} as one of the main performance indicators. Moreover, it should be taken into account that a lower initial athlete fitness level will be of relevance in order to achieve higher scope of VO_{2max} improvement. On the other hand, although gender is a variable that may influence the performance gains, in our opinion this result is conditioned by the level of the athletes included in the analyzed studies. Therefore, we don't consider it as a variable that clearly affects VO_{2max} improvements.
- Paper 2:
 - It is expected that this HRV-guided training protocol will improve functional performance in the high-level athletes, achieving better results than a traditional training method, and thus providing a good strategy for coaches of high-level athletes.
- Paper 3:
 - The results of the RCT suggest that using HRV as a daily guide to professional runners' training demonstrates that their recovery is better

than expected, allowing them to achieve higher intensities at a lower training volume. However, polarized training, which includes only low or high training intensities, is recommended to strengthen cardiovascular performance and cardiac vagal modulation. Nevertheless, the increment in HRV scores suggest better cardiovascular adaptations, favoring HRV as a measure for optimizing training individualization and cardiovascular performance.

Apéndice

Documentos de consentimiento informado

FORMULARIO DE CONSENTIMIENTO INFORMADO

NOMBRE: _____

DNI: _____

D. Alberto González Quílez, doctorando de la Universidad de Almería (UAL), ha informado antes del comienzo de las mediciones los principales problemas que pueden surgir a la hora de medir los efectos fisiológicos y psicológicos del entrenamiento basado en la Variabilidad de la Frecuencia Cardíaca (HRV) en corredores de fondo, y de la necesidad de realizar con anterioridad un reconocimiento previo a la realización de las mediciones, debido a las características de éstas.

Por tanto, el sujeto cuyo nombre y firma figuran en el consentimiento insiste en su deseo de participar en el estudio y acudir a todas las sesiones de valoración, asumiendo, en caso de que sucedan, las consecuencias patológicas posibles.

Objetivo:

Realizar una serie de mediciones fisiológicas y psicológicas para analizar el efecto del entrenamiento basado en la variabilidad del HRV en corredores de fondo.

Procedimiento:

El estudio tendrá una duración de 3 meses. Durante este tiempo los participantes deberán realizar los siguientes test: test de esfuerzo máximo en tapiz rodante para el cálculo del VO₂max y test de resistencia de 3000m, en tres momentos del estudio. Las mediciones asociadas son: HRV pre test y HRV diario, electrocardiografía, lactato, esfuerzo percibido (Escala de Borg), frecuencia cardíaca máxima, composición corporal, escalas de ansiedad, estado de ánimo y estrés, analítica.

Riesgos:

Puede ocasionar sensación de mareo, caídas, hipoglucemias y otras causas derivadas de los test.

Confidencialidad:

Todos los datos personales obtenidos son estrictamente confidenciales y serán analizados anónimamente. Solo yo y el equipo investigador tendremos acceso a los mismos y estarán protegidos ante cualquier uso indebido atendiendo a la Ley 15/1999 de 13 de diciembre.

Derechos del participante:

En todo momento soy libre de dejar de colaborar si lo creo conveniente. No habrá contra mí ningún tipo de sanción o represalias.

Reconozco que participo libremente bajo mi propia responsabilidad.

Soy consciente de la información incluida en este formulario, comprendo los procedimientos y consiento libremente en realizar los test y en que se tomen fotografías de dicha ejecución.

En _____, a _____ de _____ de 20____

Firma del participante

FORMULARIO DE CONSENTIMIENTO INFORMADO PARA PRUEBA DE ESFUERZO

Usted tiene derecho a conocer el procedimiento de prueba de esfuerzo convencional (ergometría) a la que va a ser sometido y las complicaciones más frecuentes que pueden ocurrir. Este documento intenta explicarle todas esas cuestiones; léalo atentamente y consulte con el personal sanitario todas las dudas que se le planteen. Le recordamos que, por imperativo legal, tendrá que firmar el consentimiento informado para que podamos realizar dicha prueba.

La ergometría es una prueba no invasiva, salvo que se tomen muestras de sangre para análisis del ácido láctico. Permite comprobar la respuesta del sistema cardiovascular al ejercicio físico. Se utiliza para valorar la aparición de síntomas o alteraciones electrocardiográficas inducidas por el ejercicio, evaluar la capacidad funcional, la respuesta al ejercicio de algunas arritmias y de la tensión arterial; y para el diagnóstico de isquemia cardiaca que puede aparecer con el ejercicio.

Se realiza caminando y corriendo por una cinta rodante o pedaleando en bicicleta ergométrica. Durante la prueba la velocidad aumenta progresivamente y/o la pendiente, o el nivel de carga de la bicicleta. La duración suele ser de 8-12 minutos.

Durante su realización, se controla la presión arterial, la frecuencia cardiaca el electrocardiograma para analizar sus variaciones y los gases espirados. La prueba se suspende si aparecen signos o síntomas alarmantes, o si usted no desea continuar. Inicialmente la prueba de esfuerzo es máxima, por lo que suele terminarse por cansancio muscular, dolores en las piernas o fatiga general, que desaparecen o se alivian al cesar el esfuerzo.

La realización de esta prueba tiene unos riesgos tales como: palpitaciones, mareo, cansancio, calambres en las piernas y aumento o disminución de la tensión arterial. De forma infrecuente puede presentarse: dolor torácico anginoso, síncope y la presentación de arritmias ventriculares, que implican la terminación de la prueba. Si aparecen complicaciones, el personal médico y de enfermería están capacitados y disponen de los medios necesarios para tratar de resolverlas.

He leído qué es, cómo se realiza y para qué sirve la ergometría. También he entendido de los riesgos existentes, las posibles molestias y complicaciones. He comprendido todo lo anterior y doy voluntariamente mi autorización y consentimiento para que el personal docente de la Universidad de Murcia la realicen y utilicen los datos e información obtenida (incluidas fotos) con fines docentes y de investigación, respetando mi anonimato. Si durante el procedimiento, de manera imprevista, se necesitase para bien del paciente realizar algún tipo de maniobra o intervención no informada previamente, autorizo expresamente a que se lleve a cabo. Puedo retirar este consentimiento cuando lo desee.

Firmado:

Nombre y apellidos: _____ DNI: _____

Yo, D. _____ he informado a este paciente y/o a su representante legal, del propósito y naturaleza del procedimiento, así como de sus riesgos.

Firma y nº de colegiado del médico responsable: _____

Informe del Comité de Bioética

D. DIEGO LUIS VALERA MARTÍNEZ, presidente de la Comisión de Bioética de la Universidad de Almería

INFORMA QUE:

Tras estudiar el informe presentado por el Comité de Bioética de Investigación Humana, en la reunión de la Comisión de Bioética de 19 de septiembre de 2019, y que fue discutido, esta Comisión evalúa positivamente y emite **Informe Favorable** para el siguiente estudio:

Título del estudio	Investigador/a principal
Efectos fisiológicos y psicológicos del entrenamiento basado en la variabilidad de la frecuencia cardiaca en corredores de fondo	Antonio Granero Gallegos

Y a los efectos oportunos lo firmo en Almería a 19 de septiembre de 2019



Diego Luis Valera Martínez

Presidente de la Comisión de Bioética

Universidad de Almería



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Artículos publicados



Review

HRV-Based Training for Improving VO₂max in Endurance Athletes. A Systematic Review with Meta-Analysis

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Abstract: This review aimed to synthesize evidence regarding interventions based on heart rate variability (HRV)-guided training for VO₂max improvements in endurance athletes and address the issues that impact this performance enhancement. The Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE, CINAHL Complete, the Web of Science Core Collection, Global Health, Current Contents Connect, and the SciELO citation index were searched. Inclusion criteria were: randomized controlled trials; studies with trained athletes enrolled in any regular endurance training; studies that recruited men, women, and both sexes combined; studies on endurance training controlled by HRV; studies that measured performance with VO₂max. A random-effects meta-analysis calculating the effect size (ES) was used. Moderator analyses (according to the athlete's level and gender) and metaregression (according to the number of participants in each group) were undertaken to examine differences in ES. HRV-guided training and control training enhanced the athletes' VO₂max ($p < 0.0001$), but the ES for the HRV-guided training group was significantly higher ($p < 0.0001$; $ES_{HRVG-CG} = 0.187$). The amateur level and female subgroup reported better and significant results ($p < 0.0001$) for VO₂max. HRV-guided training had a small ($ES = 0.402$) but positive effect on endurance athlete performance (VO₂max), conditioned by the athlete's level and sex.

Keywords: performance; heart rate variability; high-level athletes; maximal oxygen uptake

1. Introduction

1.1. Description of the Condition

The key components in any training program are the volume (i.e., how much), intensity (i.e., how hard), and frequency (i.e., how often) of the exercise sessions, and the combination of these 'training impulses' determines the magnitude of adaptive responses that improve the physical condition of an athlete or increase fatigue [1]. Combining these key elements to optimize training in athletes for better performance represents a relevant area of research within exercise physiology and sports medicine [2]. It is recognized that a standard training program applied to a group of athletes

can induce diverse responses in terms of performance and physiological adaptations [3,4]. Therefore, individualization is recognized as a training principle [1] as well as the need to adjust training stimuli to the psychophysical load capacity and individual tolerance of each athlete, if individual responses to training and recovery loads are intended for optimal performance [5]. The maximal oxygen uptake (VO_{2max}) is considered one of the main indicators for measuring an athlete's performance and cardiovascular adaptation to training loads [6]. The VO_{2max} is defined as the largest volume of oxygen that the body can capture, use, and transport during intense exercise [7] and is a determining factor of endurance performance [7,8]. As Vesterinen et al. [4,9] state, although some athletes show great endurance performance improvements after standardized group training (even up to 40% in VO_{2max}), other athletes show no changes or benefits, and sometimes even show a decrease in endurance performance. In recent years, research has looked at whether heart rate variability (HRV)-guided training has positive effects on athletic performance, given that this type of training allows daily adjustment of the training and recovery stimuli, individually based on HRV records [4,5,10].

1.2. Description of the Intervention

HRV is an indicator that enables the noninvasive analysis of autonomic nervous system activity in both its sympathetic and parasympathetic branches [11]. This is relevant if we consider that an important component of the interindividual variability in physiological responses to training is related to the balance between the parasympathetic (PNS) and sympathetic (SNS) activity of the autonomic nervous system (ANS) [12]. According to Huang et al. [13], HRV is considered the variation in the time interval between two consecutive heartbeats and obtained by calculating the time interval between two consecutive R waves (i.e., RR interval fluctuation) in the electrocardiogram (ECG). Since the elapsed time between beats is not constant, high vagally related HRV values are associated with efficient ANS, promoting behavioral adaptation and cognitive flexibility during stress [14], while low HRV is indicative of an inefficient ANS, resulting in maladaptive responses to stress and perceived threats [13]. HRV analysis is considered a useful method for measuring the heart's ability to adapt to endogenous and exogenous loads [15]; therefore, it can be used for the individual assessment of responses to training loads and recovery adaptation [4,16]. High HRV measurements indicate more parasympathetic than sympathetic activation, which is indicative of better recovery and preparedness for facing high-intensity training sessions [17]. HRV-guided training starts with a preparation period of about four weeks, which serves as a standardized data collection phase to obtain the baseline HRV values (e.g., $\ln rMSSD$; the natural logarithm of the square root of the mean value of the sum of the squares of the differences between the adjacent RR intervals) and their normal range (upper and lower limits) for each athlete [9,18]. Once the normal range of HRV measurements has been established, the training prescribed (moderate- or high-intensity session) is based on this calculation, which is normally updated weekly [19]. Traditionally, the vagally related HRV index has been measured with ECG [20], and quantified by means of $rMSSD$ [17]. Currently, the development and validation of new applications (i.e., smartphone applications: Kubios-HRV, Elite-HRV, Mobile Lab, or HRV4Training) facilitate daily HRV measurements and their quantification and, thus, the individual adaptation of training loads and recovery.

1.3. How the Intervention Might Work

Bellenger et al. [21], in a recent systematic review with meta-analysis, highlighted the need to use monitoring systems that accurately reflect the athletes' adaptations to the training stimulus. Although there have been numerous research studies using the HRV measure to check wellness and training adaptation in athletes [22,23], these have not focused on performance improvement based on HRV-guided training but have followed training interventions based on a traditional and nonindividualized methodology.

In contrast, evidence exists supporting the use of HRV-guided training for improved performance in endurance athletes. With this type of training monitoring, some studies have found significant VO_{2max} improvements in athletes who have developed individualized endurance training programs based on daily HRV values. These studies alternated moderate-intensity sessions with

high-intensity sessions [4,10] or even rest sessions, vigorous-intensity training, and moderate-intensity exercise [5]. However, Javaloyes et al. [18], in a program with similar characteristics developed with professional cyclists, found no significant improvements in VO_{2max} . Likewise, significant improvements have been found among athletes following HRV-guided training in other variables; for example, for lactate in maximal test [10], speed in maximal test [4], time in maximal test [4,10], or muscle strength [24]. At the level of perceived recovery, significant improvements have also been found in variables such as general stress, emotional stress, lack of energy, and even overall mood disturbance [25]. HRV-guided training may, therefore, function as an alternative method for improving performance in resistance athletes.

1.4. Why Is This Review Important?

In the search to improve athletic performance, different training methods have been tried and studied, such as intensified training [2] or submaximal tests [26]. However, it has also been recognized that the same training program followed by a group of athletes can provoke a wide range of reactions in terms of performance and physiological adaptations [3]. Overuse injuries occur due to repetitive submaximal loading of the musculoskeletal system when there is inadequate rest to allow for structural adaptation to take place [27]. In recent years, HRV-guided training has shown itself to be a promising method for improving different performance variables (e.g., VO_{2max}) compared to predefined training (traditional training) through the monitoring and individualization of endurance athletes' training [4,28]. HRV-guided training has been investigated in randomized trials on samples from different endurance sports, such as skiers [28], runners [4,25], and cyclists [18]), as well as athletes of different ages and levels: elite [18,28] and recreational endurance athletes [5,24,25]. Therefore, it is important to carry out a systematic review and meta-analysis of the different experimental studies conducted so far on endurance athletes in order to assess whether HRV-guided training is an effective method for performance improvement.

2. Objectives

As mentioned above, this review aimed to analyze the effect of HRV-guided training on VO_{2max} in endurance athletes.

We asked the following research questions regarding HRV-guided training in endurance athletes:

Research Question 1: *Does HRV-based training have an effect on VO_{2max} ?*

Research Question 2: *Is the effect of this type of training superior to that of traditional training?*

Research Question 3: *Is the level of the athletes decisive in obtaining an effect on the VO_{2max} ?*

Research Question 4: *Does the effect of HRV-guided training determine VO_{2max} scores according to the gender of the athlete?*

3. Methods

The methods detailed below are reported in accordance with the Campbell Collaboration policies and guidelines for systematic reviews [29].

3.1. Criteria for Considering Studies for This Review (Eligibility Criteria)

3.1.1. Types of Studies

We included randomized controlled trials (RCTs) and the first period of cross-over RCTs and experimental studies using a random method for the treatment assignment in order to reduce the risk of allocation bias. We restricted study eligibility by language. We did not restrict study eligibility by publication status.

3.1.2. Types of Participants

We included studies with trained athletes enrolled in any form of regular endurance training (e.g., runners, triathletes, skiers, and cyclists). We included studies that recruited both men and women, or men and women separately.

3.1.3. Types of Interventions

We included studies on endurance training controlled by heart rate variability to improve the athletes' performance. We considered designs comprising any dose, frequency, and duration. We also considered studies with the following types of comparisons:

- Endurance training controlled by HRV versus no specific training intervention (e.g., habitual physical activity).
- Endurance training controlled by HRV versus another training intervention (e.g., traditional endurance training or another type of traditional training).
- Endurance training controlled by HRV versus another training intervention (i) versus a further training intervention (ii).
- Endurance training controlled by HRV (i) versus endurance training controlled by HRV (ii) versus another training intervention versus no specific training intervention.

3.1.4. Types of Outcome Measures

- Primary
 - Maximal oxygen consumption (VO_{2max})

3.2. Search Methods to Identify the Studies

3.2.1. Electronic Searches

The register contains studies identified from the Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE, EMBASE, CINAHL Complete, the Web of Science Core Collection, Global Health, Current Contents Connect, and the SciELO citation index.

The search is up to date as of 15 June 2020. The language was restricted, considering only English or Spanish. The terms used to search the databases were: (*amateur OR elite OR train**) AND (*HRV-guided OR "heart-rate variability guided"*).

3.2.2. Searching Other Resources

We checked the reference lists of all the included studies and systematic reviews for additional references. We contacted experts in the field and the authors of the included studies to identify additional unpublished studies. We also checked the results of completed trials registered on the US National Institutes of Health Ongoing Trials Register, ClinicalTrials.gov, the World Health Organization International Clinical Trials Registry Platform (WHO ICTRP), and proceedings of conferences for relevant research.

3.3. Data Collection and Analysis

We conducted the following data collection and analysis in accordance with the recommendations in the Cochrane Handbook for Systematic Reviews of Interventions [30].

3.3.1. Selection of Studies

Two review authors independently screened the titles and abstracts of all the retrieved references in Microsoft Excel 2018 (Microsoft, New York, United States) for Windows. The full-text study reports were retrieved for all the citations that at least one review author considered potentially relevant. Two review authors independently screened the full-text articles and identified studies for inclusion; they also identified and recorded the reasons for excluding studies in the excluded studies

characteristics. Any disagreements were resolved through discussion. The selection process is detailed in a PRISMA flow diagram [31].

3.3.2. Data Extraction and Management

We used a standardized piloted data collection form in Microsoft Excel 2018 for Windows and extracted the following study characteristics and outcome data: (i) Methods: study design; (ii) Participants: randomized number, study participants' mean age or age range, study location and setting, recruitment methods, inclusion and exclusion criteria, and type of endurance sport; (iii) Interventions: a description of the training intervention characteristics, the dose and duration of the training intervention, a description of the comparison intervention characteristics, the length of follow-up, the number of withdrawals, and the reasons for withdrawal; (iv) Outcomes: a description of the primary and secondary outcomes in the review that were reported in the trial and a listing of other outcomes collected in the trial; (v) Notes: the trial funding and notable conflicts of interest of the trial authors; (vi) a 'risk of bias' assessment. Two review authors independently extracted the outcome data from the included studies into Microsoft Excel 2018 spreadsheets and compared the data to identify any discrepancies in the data entries. Any disagreements were resolved by consensus. In the Characteristics of Included Studies section, we noted down if a trial did not report outcome data in a usable way. We then transferred all the outcome data into the Comprehensive Meta-Analysis software version 2.2.064 (Biostat, Englewood, United States) [32].

3.3.3. Risk-of-Bias Assessment in the Included Studies

Two review authors (M.C.P., A.G.G.) independently assessed the risk of bias for each included trial using the Cochrane risk-of-bias tool [30]. Any disagreements were resolved by discussion. The risk of biases were assessed for the following domains: random sequence generation (selection bias), allocation concealment (selection bias), blinding of participants and personnel (performance bias), blinding of outcome assessment for each outcome (detection bias), incomplete outcome data (attrition bias), selective outcome reporting (reporting bias), and other biases (such as the validity of outcome measure and baseline comparability). Each potential source of bias was assessed as either high, low, or unclear, and a quotation from the study report was provided together with a justification for the judgment in the 'risk of bias' tables. The judgments across the different studies were summarized for each of the domains listed.

3.3.4. Treatment Effect Measures

The outcome data for each study were uploaded into the data tables of the Comprehensive Meta-Analysis software to calculate the treatment effects. We used the mean difference (MD) for continuous outcomes reported on the same scale, and the standardized mean difference (SMD) for continuous outcomes measured on different scales in different trials ($SMD = \frac{M_{HRV \text{ guided training}} - M_{\text{control group}}}{\text{Standard deviation}}$) [33]. Uncertainty was expressed with 95% confidence intervals (CIs) for all the effect estimates.

3.3.5. Assessment of Heterogeneity and Reporting Bias

Heterogeneity was assessed qualitatively between studies in three ways: a visual examination of the forest plots, the Chi² test ($p \leq 0.10$) for heterogeneity, and the I² statistic. The implications of the observed I² statistic value were considered as follows: 0% to 40%—might not be important; 30% to 60%—may represent moderate heterogeneity; 50% to 90%—may represent substantial heterogeneity; 75% to 100%—considerable heterogeneity [30]. Publication bias was assessed by examining the asymmetry of a funnel plot using Egger's test. If studies were distributed symmetrically around the mean effect size (ES), there was an absence of publication bias [33]. Subgroup analysis was carried out using the outcome for athlete level (elite vs. amateur) and sex (men, women, and both sexes combined). Metaregression was used to assess the relationship between the studies and the variable sample size.

3.3.6. Sensitivity Analysis

A sensitivity analysis was carried out to check whether the results varied according to the endpoint data.

4. Results

4.1. Description of the Studies

4.1.1. Search Results

The search produced a total of 36 studies, with 222 additional records identified through other sources. The removal of duplicates resulted in eleven studies, which were screened by the two authors based on the title and abstract. Three studies were excluded. Eight full-text articles were assessed for eligibility. Two more studies were excluded, and six studies were included either in the qualitative analysis or in the quantitative metasynthesis. The PRISMA flow chart illustrates the search and selection process (Figure 1).

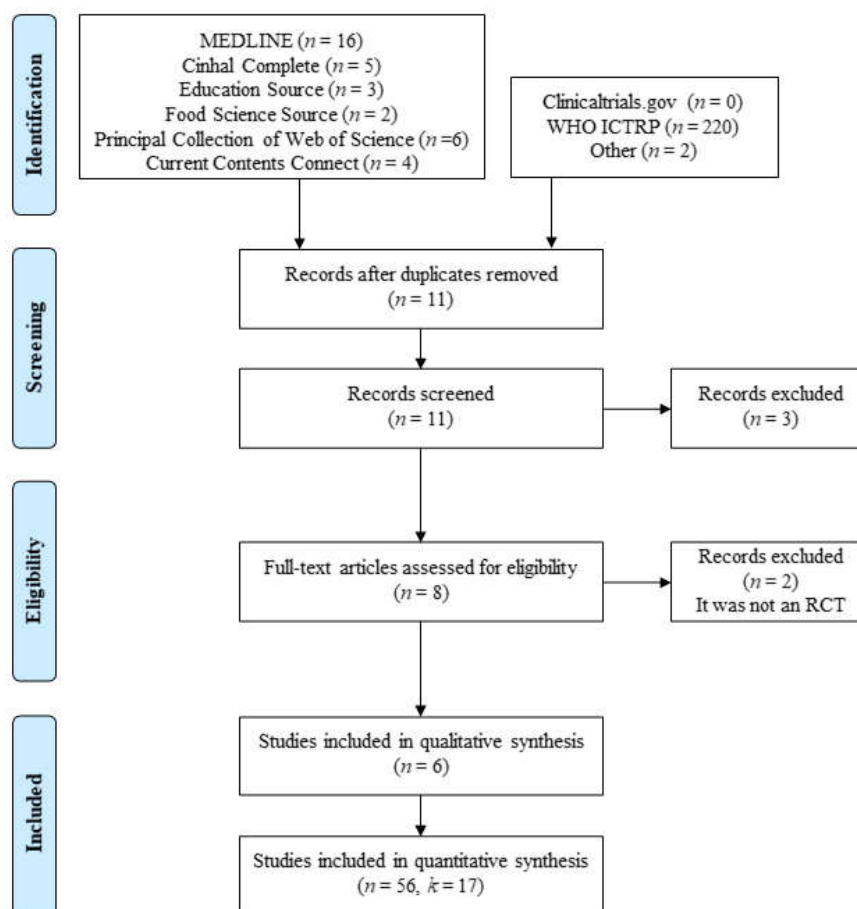


Figure 1. Study flow diagram following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses Guidelines [31], where n is the number of papers and k is the number of individual studies.

4.1.2. Included Studies

Six studies carrying out HRV-guided training with elite or amateur athletes were included in this review [4,5,10,18,19,28], which were identified by the first author and publication date: Javaloyes_2019, Kiviniemi_2007, Kiviniemi_2010, Nuuttila_2017, Schmitt_2018, and Vesterinen_2016.

- Study location

Schmitt_2018 conducted their study at the French National Ski-Nordic Center, while the locations for the other five studies were not specified.

- Study design

Every study included in this review was a randomized controlled trial.

- Participants

A total of 195 participants (134 men and 61 women) were included in these studies. Kiviniemi_2007, Javaloyes_2019, and Nuuttila_2017 considered only male samples of 30, 17, and 32 participants, respectively. In the rest of the studies, the samples were composed of men and women: Kiviniemi_2010 included 24 men and 36 women, Schmitt_2018 incorporated 19 men and 5 women, and Vesterinen_2016 assessed 20 men and 20 women. In the studies by Javaloyes_2019 and Schmitt_2018, the samples were composed of professional athletes (cyclists and Nordic skiers, respectively) while in the other four studies, the samples were of a nonprofessional level.

- Interventions

According to the types of comparisons contemplated in the present systematic review ((a) endurance training controlled by HRV versus no specific training intervention; (b) endurance training controlled by HRV versus other training intervention; (c) endurance training controlled by HRV (i) versus another training intervention (ii) versus another training intervention; (d) endurance training controlled by HRV (i) versus endurance training controlled by HRV (ii) versus other training intervention versus no specific training intervention. Kiviniemi_2007, Javaloyes_2019, Nuuttila_2017, and Vesterinen_2016 were classified in Comparison B, Schmitt_2018 in Comparison C, and Kiviniemi_2010 in Comparison D.

The interventions in the included studies focused on running (Kiviniemi_2007, Kiviniemi_2010, Nuuttila_2017, and Vesterinen_2016), skiing (Schmitt_2018), and cycling (Javaloyes_2019). They were from 6 to 15 weeks long. In most of the studies, three (Nuuttila_2017) or four (Javaloyes_2019, Schmitt_2018, and Vesterinen_2016) low-intensity preparation weeks were followed either by the experimental or control groups (standard training) before the intervention. An eight-week intervention was carried out in Javaloyes_2019, Kiviniemi_2010, Nuuttila_2017, and Vesterinen_2016, whereas Kiviniemi_2007 considered four weeks of training and Schmitt_2018 15 days. The assessment weeks were treated separately from the intervention period in Javaloyes_2019, Kiviniemi_2007, and Schmitt_2018.

In every study, the experimental groups trained at moderate or high intensities according to their daily HRV scores. The control groups (standard training) followed a predefined training design at high, moderate, and low intensities (Javaloyes_2019), high and moderate intensities (Kiviniemi_2010 and Nuuttila_2017), high and low intensities (Kiviniemi_2007) or moderate and low intensities (Vesterinen_2016). The control group (standard training) design was not explained in Schmitt_2018.

- Outcomes

The primary outcome analyzed in the included studies was VO_{2max} . The secondary outcomes were: ventilatory thresholds (Javaloyes_2019, Kiviniemi_2007) and power in the cycling test (Javaloyes_2019); rMSSD or RR interval (Javaloyes_2019, Kiviniemi_2007, and Schmitt_2018); basal heart rate (Nuuttila_2017, Kiviniemi_2010, and Schmitt_2018); maximal heart rate in the ergometer test (Nuuttila_2017); speed in the treadmill test (Kiviniemi_2007, Nuuttila_2017, and Vesterinen_2016); maximal speed in the 10 m test (Nuuttila_2017); time and lactate in the 3000 m test (Nuuttila_2017); maximal load in the ergometer test (Kiviniemi_2007 and Kiviniemi_2010); and oxygen saturation and VO_2 at the second ventilatory threshold (Schmitt_2018).

Further details about participants, interventions, comparators, and outcomes are provided in Table 1.

Table 1. Overview of the studies included in the review.

Author, Year	Method	Participants	Intervention	Outcomes	Results	Risk of Bias		
						Bias	Author's Judgment	Support for the Judgment
Javaloyes_2019	Randomized controlled trial	Trained male cyclist, mean age of 38.42 years. N = 17: EG = 9 + CG = 8 Location: not specified. Recruited from local clubs Inclusion criteria: at least 2 years of experience in cycling. Exclusion criteria: not specified.	15 weeks (4 weeks of baseline period to capture baseline HRV + 8 weeks of training + 3 weeks of assessments); 4–7 sessions/week; time depended on the training intensity. EG: HRV-G-based training before each session; training MICT and HIIT according to HRV. CG: 4 high-intensity training sessions + 4 high-intensity interval training sessions + 6 moderate-intensity training sessions + 2–5 low-intensity training sessions/week. No follow-up periods. No withdrawals	Primary: VO _{2max} (maximal bicycle ergometer test, direct measurement). Secondary: ventilatory thresholds in the graded test, peak power output in the graded test, rMSSD with a heart rate monitor + kubios (LnrMSSD) in a supine position for 90 s, mean power output during a 40 min all-out cycling test.	VO _{2max} : no significant differences between intragroups and intergroups. Moderate training load: significant intergroup differences (EG = 24%; CG = 27%). VT2: significant improvements in EG (36.11 ± 3.73W). Peak power output: significant improvements in EG (17.45 ± 3.91W). LnrMSSD: significant differences between intergroups for the percentage of change (EG = 0.85 ± 3.21%, CG = -2.02 ± 5.21%). Mean power 40M: significant improvements in EG (17.67 ± 3.03W)	Selection	Unclear	Insufficient information about the sequence generation process and allocation to permit judgment of 'low risk' or 'high risk'.
						Performance	High	Incomplete blinding, and the outcome is likely to be influenced by lack of blinding.
						Detection	Unclear	The study did not address this outcome.
						Attrition	Low	No missing outcome data.
						Reporting	Unclear	Insufficient information to permit judgment of 'low risk' or 'high risk'.
						Other	Low	The study appears to be free of other sources of bias.
Kiviniemi_2007	Randomized controlled trial	30 healthy recreational male runners N = 30: TRA: predefined training group (n = 10) + HRV: HRV-guided training (n = 10) + CG: Control group (n = 10). Location: Not specified Recruitment: The candidates were interviewed with a standardized	6 weeks: 1-week baseline resting + pretest Intervention: 4-week training period (6 days per week) consisting of running sessions at either a low- or high-intensity level according to recommendations by the American College of Sports Medicine: low-intensity: 40 min of jogging at 65% of maximal HR; high-intensity exercise included 5 min warm-up and cool-down	Primary: VO _{2peak} (maximal treadmill ergometer test: direct measurement). Secondary: high-frequency power of RR interval with software while standing for 5 min, maximal load in the ergometer test, maximal running velocity in the ergometer test; ventilatory threshold (VT) from the relation of running velocity	VO _{2peak} : significant intragroup improvements in the HRV group (pretest = 56 ± 4; post-test = 60 ± 5 mL/kg/min). High-frequency power of RR interval: significant intragroup improvements in TRA (pretests = 4.7 ± 0.4; post-test = 5.5 ± 0.8 ln ms2), and HRV (pretests = 4.8 ± 0.6; post-test = 5.2 ± 0.8 ln ms2). Maximal load: significant intragroup improvements in TRA	Selection	Unclear	Insufficient information about the sequence generation process and allocation to permit judgment of 'low risk' or 'high risk'.
						Performance	Unclear	Insufficient information to permit judgment of 'low risk' or 'high risk'.
						Detection	Unclear	Insufficient information to permit judgment of 'low risk' or 'high risk'.
						Attrition	Unclear	The study did not address this outcome.
						Reporting	Unclear	Insufficient information to permit judgment of 'low risk' or 'high risk'.
						Other	Low	The study appears to be free of other sources of bias.

		<p>scheme to ascertain their medical history and levels of physical activity. Inclusion criteria: healthy men. Exclusion criteria: subjects who had done regular physical exercise training less than twice a week for the past 3 months, competing athletes, and subjects with diabetes mellitus, asthma, or cardiovascular disorders were excluded.</p>	<p>periods at 65% of the maximal HR before and after 30 min of running at 85% of maximal HR. The last week for the post-test. HRV: exercised at low- or high-intensity or rested based on their daily HRV measurements at home. If HRV increased or did not change, vigorous-intensity training on that day. If HRV decreased, moderate-intensity exercise or rest. TRA: weekly training started with low-intensity exercise followed by two sessions of high-intensity exercise on successive days. This 3-day period was repeated before a day of rest. CG: no intervention. No follow-up period. 4 withdrawals: TRA (2); HRV (1); CG (1).</p>	<p>and selected ventilatory parameters.</p>	<p>(pretest = 15.1 ± 1.3 km/h; post-test = 15.7 ± 1.2 km/h); significant intergroup differences between CG (post-test = 14.9 ± 1.5 km/h) and TRA (post-test = 15.7 ± 1.2 km/h), TRA and HRV (post-test = 16.4 ± 1.0 km/h), and between CG and HRV. VT: significant intragroup improvements in HRV (pretest = 12.2 ± 0.6 km/h; post-test = 16.4 ± 1.0 km/h)</p>			
Kiviniemi_2010	Randomized controlled trial	<p>Healthy men and women. Mean age of 34.57 years. N = 60. Men, n = 24; women, n = 36). ST: standard training (8 men + 8 women) + HRV-I: HRV-guided training for men and women (EG: 8 men + 8 women) + HRV-II: HRV-guided training tailored for</p>	<p>8 weeks of aerobic exercise sessions (40 min), vigorous-intensity level: HR between 85% of the HRpeak-5 bpm lower limit; moderate-intensity exercise was 70% of the HRpeak-5 bpm lower limit. HRV-I: if HRV increased or did not change, vigorous-intensity training on that day. If HRV decreased, moderate-</p>	<p>Primary: VO_{2max} (maximal bicycle ergometer test: direct measurement). Secondary: HR, RR interval with a heart rate monitor (SD1) while standing for 3 min, maximal load in the ergometer test.</p>	<p>VO_{2max}: significant intragroup improvements in ST (men subgroup) (pretest = 50 ± 7; post-test = 53 ± 7 mL/kg/min), ST (women subgroup) (pretest = 35 ± 5; post-test = 37 ± 4 mL/kg/min), HRV-I (men subgroup) (pretest = 50 ± 6; post-test = 54 ± 6 mL/kg/min), HRV-I (women subgroup)</p>	<p>Selection</p> <p>Performance</p> <p>Detection</p> <p>Attrition</p> <p>Reporting</p> <p>Other</p>	<p>High</p> <p>Unclear</p> <p>Unclear</p> <p>High</p> <p>Unclear</p> <p>Low</p>	<p>Allocation based on the results of a laboratory test or a series of tests.</p> <p>Insufficient information to permit judgment of 'low risk' or 'high risk'.</p> <p>The study did not address this outcome.</p> <p>High rates of loss to follow-up.</p> <p>Insufficient information to permit judgment of 'low risk' or 'high risk'.</p> <p>The study appears to be free of other sources of bias.</p>

		women (12) + CG (8 men + 8 women). Location: Not specified Recruitment: advertisement local newspaper Inclusion criteria: healthy men and women Exclusion criteria: smoker, BMI ≥ 30 kg/m ² ; regular physical exercise training more than twice a week for the last 3 months, competing athletes, mellitus, asthma, or cardiovascular disorders.	intensity exercise or rest. HRV-II: vigorous-intensity exercise only when HRV had increased. ST group: two moderate-intensity and three vigorous-intensity exercises weekly. CG: no intervention No follow-up period. 7 withdrawals: ST (1 man + 1 woman) + HRV-I (7 men + 7 women); CG (7 men + 8 women) + HRV-II (10); 4 because of illness or injury and 3 because of insufficient compliance.	(pretest = 36 ± 4; post-test = 39 ± 3 mL/kg/min), and in HRV-II (women subgroup) (pretest = 37 ± 5; post-test = 40 ± 5 mL/kg/min). HR: RR interval: significant intragroup improvements in HRV-I (men subgroup) (pretests = 13.7 ± 6.7; post-test = 16.9 ± 8.7 ms). Maximal load: significant intragroup improvements in ST (men subgroup) (pretest = 275 ± 28W; post-test = 293 ± 35W), ST (women subgroup) (pretest = 179 ± 32W; post-test = 198 ± 35W), HRV-I (men subgroup) (pretest = 270 ± 29W; post-test = 300 ± 25W), HRV-I (women subgroup) (pretest = 174 ± 28W; post-test = 189 ± 25W), and in HRV-II (women subgroup) (pretest = 177 ± 26W; post-test = 194 ± 23W)			
Nuuttila_2017	Randomized controlled trial	Males, 19–37 years. N = 24. EG = 13 and CG = 11. Location: not specified. Recruitment: not specified. Inclusion criteria: recreationally endurance training. Exclusion criteria: not specified	11 weeks (3 weeks of control + 8 weeks of training). EG: 2–5 sessions/week; CG: 6 sessions/week; time depended on the training intensity. EG: 4 moderate-intensity endurance training sessions + 20 high-interval intensity training sessions. Training MICT and HIIT according to HRV. CG: 22 moderate-intensity endurance	Primary: VO _{2max} (maximal treadmill test: direct measurement). Secondary: basal heart rate, maximal heart rate, lactate in the treadmill test, V _{max} in the treadmill test, V _{max} in the 10m test, time and lactate in the 3000 m test, rMSSD with a heart rate monitor + Firstbeat in a supine position for 3 min.	VO _{2max} : significant intragroup changes (EG = 3.1 ± 0.8 mL/kg/min; CG = 2.2 ± 0.6 mL/kg/min). Basal HR: significant intragroup decrease (EG = 4.4 ± 0.6bpm; CG = 3.6 ± 0.1bpm). V _{max} in the treadmill test: significant intragroup improvements (EG = 0.9 ± 0.1km/h; CG = 0.5 ± 0.1 km/h). V _{max} in 10 m: decreased significantly in CG	Selection: High Performance: Unclear Detection: Unclear Attrition: High Reporting: Unclear Other: Low	Allocation based on the results of a laboratory test or a series of tests. Insufficient information to permit judgment of 'low risk' or 'high risk'. The study did not address this outcome. High rates of loss to follow-up. Insufficient information to permit judgment of 'low risk' or 'high risk'. The study appears to be free of other sources of bias.

		training sessions + 20 high-interval intensity training sessions + 4 high-intensity strength training sessions No follow-up periods. 9 withdrawals: illness (<i>n</i> = 1), injuries (<i>n</i> = 2), personal reasons (<i>n</i> = 3), lack of adherence (<i>n</i> = 3).	Other: body weight, height in countermovement jump, strength in the concentric dynamic leg press, nocturnal heart rate variability, testosterone, and cortisol (blood samples), % of fat (InBody 720).	from pre to mead test (0.08 ± 0.04 m/s). Time in the 3000 m test: significant intragroup decrease (EG = 35 ± 2s; CG = 35 ± 6s). Lactate in the 3000 m test: significant intragroup improvements (EG = 12 ± 18.4%) from mead-to post-test; CG = 16–0 ± 23.5%) from pre-to post-test. rMSSD: significant improvements in EG (13 ± 3ms)				
Schmitt_2018	Randomized controlled trial	24 elite Nordic skiers (19 men, age 23.3 ± 3.6; 5 women, age 22.8 ± 4.1). N = 24; H-HRV, HRV-guided training normobaric hypoxic group (<i>n</i> = 9) + H, sleeping in normobaric hypoxia group (<i>n</i> = 9); N, normoxia group (<i>n</i> = 6). Location: French National Ski-Nordic Center. Recruitment: members of the cross-country ski and Nordic combined French. Inclusion criteria: elite Nordic skiers. Exclusion criteria: a history of altitude-related sickness and health risks that could compromise the	Prior to pretest: 3 low-intensity training weeks (base training) with progressive training volume + 1-week recovery; Intervention: pretest + 15 days training (training load was organized into four training zones depending on the intensity and quantified as in Mujika et al. (1996), adapted to Nordic skiing (the threshold for training adjustment was chosen as 30% of the mean of the previous day) + posttest1 + 1 week + posttest2. Similar training content for each group. H-HRV group: sleeping normobaric in hypoxia (simulated altitude of 2700 m) with HRV-guided training; daily hypoxic dose was similar between H-HRV and H; Night SpO2 was similar between H-HRV	Primary: VO _{2max} (maximal treadmill test: direct measurement). Secondary: basal HR, peripheral oxygen saturation (SpO ₂), RR interval with a heart rate monitor (HF and LF) 5 min in a supine position and 5 min in standing, VO ₂ at the second ventilatory threshold. Others: duration of hypoxic exposure, HR, blood parameters (erythrocyte concentration, hemoglobin, hematocrit, ferritin), questionnaire of overtraining.	VO _{2max} : significant intragroup changes in H-HRV (3.8 ± 3.1%). Basal HR: significant intergroup differences (H-HRV = 55.38 ± 10.02 vs. H = 55.59 ± 4bpm; H-HRV = 55.38 ± 10.02 vs. N = 47.11 ± 6.21bpm). SpO ₂ : significant intergroup differences (H-HRV = 90.4 ± 1.3 vs. N = 94.2 ± 0.8%). RR interval: no significant differences between intergroups (H-HRV = 9561.10 ± 9436.02 ms ² ; H = 12,199.41 ± 1293.14 ms ² ; N = 7441.2 ± 4954.16 ms ²). VO ₂ second VT: significant intragroup changes for H-HRV (6.7 ± 6.1%).	Selection	High	Allocation based on the results of a laboratory test or a series of tests.
						Performance	Unclear	Insufficient information to permit judgment of 'low risk' or 'high risk'.
						Detection	Unclear	The study did not address this outcome.
						Attrition	Low	No missing outcome data.
						Reporting	High	Not all of the study's prespecified primary outcomes have been reported.
						Other	Low	The study appears to be free of other sources of bias.

		subject's safety during training and/or hypoxic exposure.	and H, but lower than in N. H: traditional training sleeping in hypoxia (simulated altitude of 2700 m). N: traditional training sleeping in normoxia. Follow-up (post-test21 after 3 weeks of end postest1)					
Vesterinen_2016	Randomized controlled trial	Recreational endurance runners (men = 20; women = 20) N = 40: EXP = 20 + TRAD = 20 Location: not specified. Recruitment: advertisement and social media Inclusion criteria: 2 years' regular endurance running training. Exclusion criteria: disease or regular medication for chronic or long-term diseases.	12 weeks (4 weeks of preparation + 8 weeks of training). The same volume as before the study for PREP and the same volume as for PREP for INT. EXP: training MICT and HIIT according to HRV. TRAD: 50% sessions at low-intensity and 50% sessions at moderate/high-intensity. Week periodization, 3:1. No follow-up periods. 9 withdrawals: sicknesses (n = 2), injuries (n = 2), lack of adherence (n = 5)	Primary: VO _{2max} (maximal treadmill test: direct measurement). Secondary: Speed in Lactate 1, speed in Lactate 2, mean speed in the 3000 m test, time in the 3000 m test, RR intervals (rMSSD) 4 min in a supine position.	VO _{2max} : significant intragroup improvements (EXP = 3.7 ± 4.6%, TRAD = 5.0 ± 5.2%). Speed in L1 significant intragroups improvement in EXP (2.8 ± 3.7%). Speed in L2 significant intragroups improvement in EXP (2.6 ± 3.3%) and TRAD (1.9 ± 2.2%). Time in the 3000 m test: significant intragroup improvements in EXP (-14.3 ± 14.1 s)	Selection Performance Detection Attrition Reporting Other	High Unclear Unclear High High Low	Allocation based on the results of a laboratory test or a series of tests. Insufficient information to permit judgment of 'low risk' or 'high risk'. The study did not address this outcome. High rates of loss to follow-up. Not all of the study's prespecified primary outcomes have been reported. The study appears to be free of other sources of bias.

4.1.3. Excluded Studies

As indicated in Figure 1, five studies were excluded from the qualitative analysis. Three studies were excluded because the VO_{2max} was not considered as an outcome [24,25,34], and two studies were excluded because they were not RCTs [35,36].

4.2. Risk of Bias in the Included Studies

The risk of bias in the included studies is summarized in Table 2. This assessment was made following the Cochrane Collaboration guidelines [30]. In addition, publication bias was assessed using a funnel plot (Figure 2). The Egger test provided statistical evidence of funnel plot symmetry, suggesting the absence of a significant publication bias ($p = 0.101$).

Table 2. Risk of bias in the included studies.

Study	Risk-of-Bias Domains						Overall Risk of Bias
	Selection	Performance	Detection	Attrition	Reporting	Other	
Javaloyes_2019	Unclear	High	Unclear	Low	Unclear	Low	Unclear
Kiviniemi_2007	Unclear	Unclear	Unclear	Unclear	Unclear	Low	Unclear
Kiviniemi_2010	Unclear	Unclear	Unclear	High	Unclear	Low	Unclear
Nuuttila_2017	High	Unclear	Unclear	High	Unclear	Low	Unclear
Schmitt_2018	High	Unclear	Unclear	Low	High	Low	Unclear
Vesterinen_2016	High	Unclear	Unclear	High	High	Low	Unclear

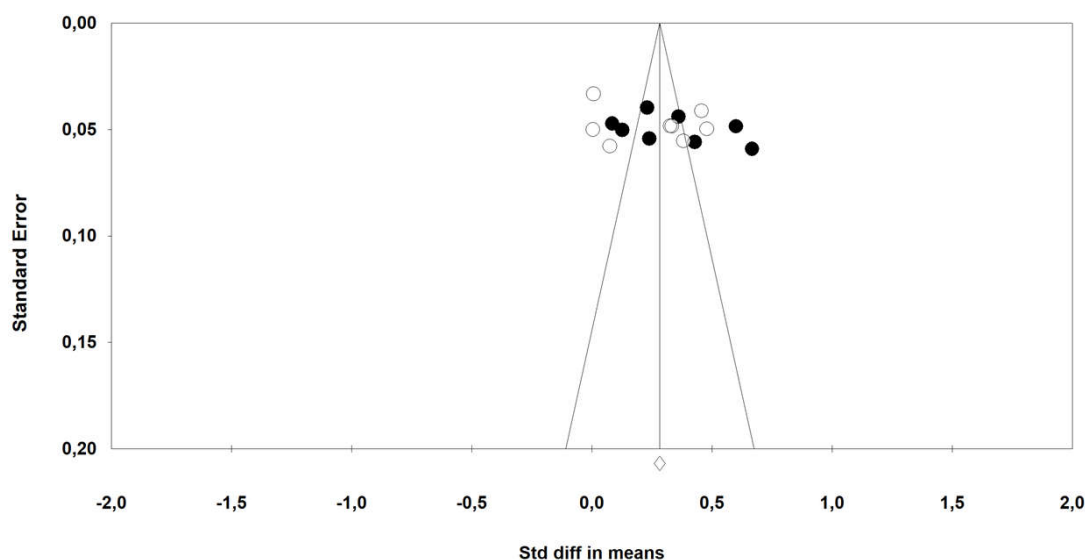


Figure 2. Funnel plot of standard error by standard differences in means (17 comparison; black circle, HRV-guided training; white circle, traditional training).

- Selection bias

In Javaloyes_2019, Kiviniemi_2007, and Kiviniemi_2010, neither the random component in the sequence generation nor the allocation concealment were described; therefore, the risk-of-bias selection was considered unclear. In Nuuttila_2017, Schmitt_2018, and Vesterinen_2016, the risk of bias was considered high because the randomization sequence was, in the first stage, based on the results of certain physical condition tests, sport discipline, age, or gender. Furthermore, in the second stage, the random component or the allocation concealment was not described.

- Performance and detection bias

The detection bias was considered unclear in all of the included studies because they did not address this outcome. The performance bias was also unclear in every study but Javaloyes_2019, which was considered high because only the participants were blinded, thus the blinding was incomplete.

- Attrition bias

In Javaloyes_2019 and Schmitt_2018, the attrition bias was considered low because there were no missing outcome data. On the other hand, Kiviniemi_2010, Nuuttila_2017, and Vesterinen_2016 presented high rates of follow-up loss for different reasons. These might be relevant in the ES observed. Moreover, no statistical procedure, such as intention-to-treat, was used to minimize this risk of bias. Therefore, they were considered as having a high risk of attrition bias. Finally, in Kiviniemi_2007, the attrition bias was unclear because this outcome was not addressed in the study.

- Reporting bias

The study protocols for the included studies were not available. Accordingly, Javaloyes_2019, Kiviniemi_2007, Kiviniemi_2010, and Nuuttila_2017 were considered as having an unclear reporting bias. For their part, Schmitt_2018 and Vesterinen_2016 did not report every outcome and were thus considered as having a high risk of reporting bias.

- Other biases

The included studies appear to be free from other sources of bias.

4.3. Synthesis of Results

The Kiviniemi_2010 and Schmitt_2017 studies were segmented for quantitative analysis according to their intervention groups. The comparisons were: Kiviniemi_2007 a, HRV (male subgroup, HRV-guided training) vs. standard training (ST); Kiviniemi_2010 a, HRV-I (male subgroup, HRV-guided training) vs. standard training (ST); Kiviniemi_2010 c, HRV-I (female subgroup, HRV-guided training) vs. standard training (ST); Kiviniemi_2010 g, HRV-II (female subgroup, HRV-guided training tailored for women) vs. HRV-I (female subgroup, HRV-guided training); Kiviniemi_2010 f, HRV-II (female subgroup, HRV-guided training tailored for women) vs. standard training (ST); Schmitt_2017 a HRV (HRV-guided training) vs. N (traditional training and normoxia sleeping); Schmitt_2017 b HRV (HRV-guided training) vs. H (traditional training and hypoxia sleeping). Therefore, the total number of individual studies analyzed were 17 ($k = 7$ for the experimental group; $k = 10$ for the control group).

- Primary outcome measures

There were five studies (Kiviniemi_2007, Kiviniemi_2010, Nuuttila_2017, Schmitt_2017 and Vesterinen_2016) with significant intragroup VO_{2max} improvements in the HRV-guided training group ($n = 95$), while no significant changes were found in Javaloyes_2019 ($n = 9$). On the other hand, in three studies (Kiviniemi_2010, Nuuttila_2017, and Vesterinen_2016), there were also significant intragroup VO_{2max} improvements in the control group ($n = 47$). The overall risk of bias was considered high in every study but for Javaloyes_2019, which was considered unclear. A random-effects meta-analysis of the six studies revealed a statistically significant ($p < 0.0001$) treatment effect for VO_{2max} in the HRV-guided training intervention (ES = 0.402; 95%CI = 0.273, 0.531). Moreover, the other training intervention was also statistically beneficial ($p < 0.0001$) for VO_{2max} improvements in the control group (ES = 0.215; 95% CI = 0.101, 0.329). However, the ES for the VO_{2max} was significantly higher ($p < 0.0001$) in the HRV-guided training group. The heterogeneity observed in the meta-analysis was significant and high in the overall analysis ($p < 0.0001$; $I^2 = 94.24\%$) and for the experimental ($p < 0.0001$; $I^2 = 9.36\%$) and the control group ($p < 0.0001$; $I^2 = 92.26\%$) (Figure 3).

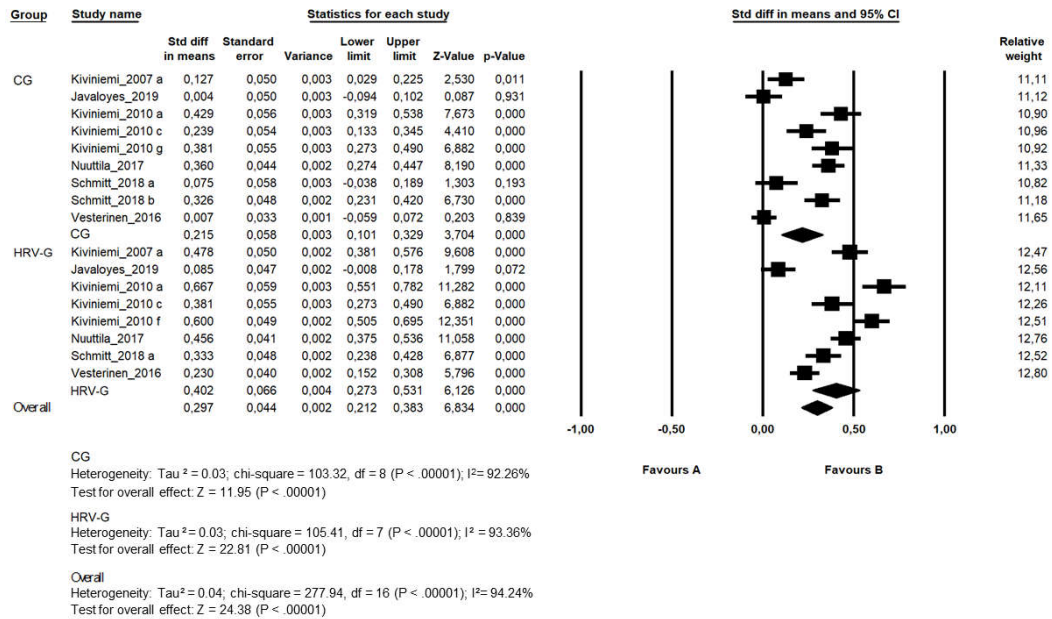


Figure 3. Standard differences in means (SDM) between post- and premeasures for VO_{2max} in included studies, segmented by the control group (CG) and heart-rate-variability-guided training group (HRV-G). Squares represent the SDM for each trial; the diamond represents the pooled SDM across trials; weight determines how much each individual study contributes to the pooled estimate; 95%CI, confidence interval.

- Moderator analyses

Owing to the high heterogeneity observed in the meta-analysis, the potential moderating effect of the following was considered to be of interest: (a) the athletes’ level (elite vs. amateur) and (b) the sex of the participants (‘men vs. women’ vs. ‘men and women’). We had originally planned to take into account the intervention duration; however, it was not finally included as a subgroup owing to there being only one study that considered an intervention period of 15 days (Schmitt_2017) while the others conducted an eight-week intervention. The sample size was used for the metaregression. Following the moderating variables (Table 3), the athletes’ level (elite vs. amateur) brought about statistically significant improvements ($p < 0.0001$) in both subgroups, while there were statistically significant differences between the subgroups ($p < 0.0001$) in favor of the nonprofessional subgroup (elite, ES = 0.17; amateur, ES = 0.36). According to the sex subgroups (‘men vs. women’ vs. ‘men and women’), there were statistically significant improvements ($p < 0.0001$) in the three subgroups and statistically significant differences ($p < 0.0001$) between the three subgroups in favor of the women (men, ES = 0.33; women, ES = 0.40; men and women, ES = 0.19). The metaregression findings (Figure 4) revealed that the sample size of the studies was directly related to the ES magnitude (regression coefficient = -0.016; standard error = 0.003; lower limit = -0.023; upper Limit = -0.011; Z-value = -5.42; $p \leq 0.0001$).

Table 3. Subgroup analyses for measuring their impact on VO_{2max} .

Research Studies			Variable: VO_{2max}			
Group	No Studies	References	SMD (95% CI)	I^2	p	p -Difference
Athlete level						
Elite	3	Javaloyes_2019; Schmitt_2018 a; Schmitt_2018 b	0.17 (0.03; 0.30)	89.63	<0.001	<0.001
Amateur	5	Kiviniemi_2010 a; Kiviniemi_2007 a; Kiviniemi_2010 c; Kiviniemi_2010 g; Nuuttila_2017; Vesterinen_2016	0.36 (0.24; 0.48)	94.66	<0.001	
Sex						
Women	3	Kiviniemi_2010 c; Kiviniemi_2010 f; Kiviniemi_2010 g	0.40 (0.25; 0.56)	88.36	<0.001	<0.001
Men	4	Javaloyes_2019; Kiviniemi_2007 a; Kiviniemi_2010 a; Nuuttila_2017	0.33 (0.17; 0.48)	94.98	<0.001	
Men and women	3	Schmitt_2017 a; Schmitt_2017 b; Vesterinen_2016	0.19 (0.06; 0.33)	92.10	0.006	

Note: SMD, standard mean difference; CI, confidence interval; VO_{2max} , maximal oxygen uptake; I^2 = I-squared.

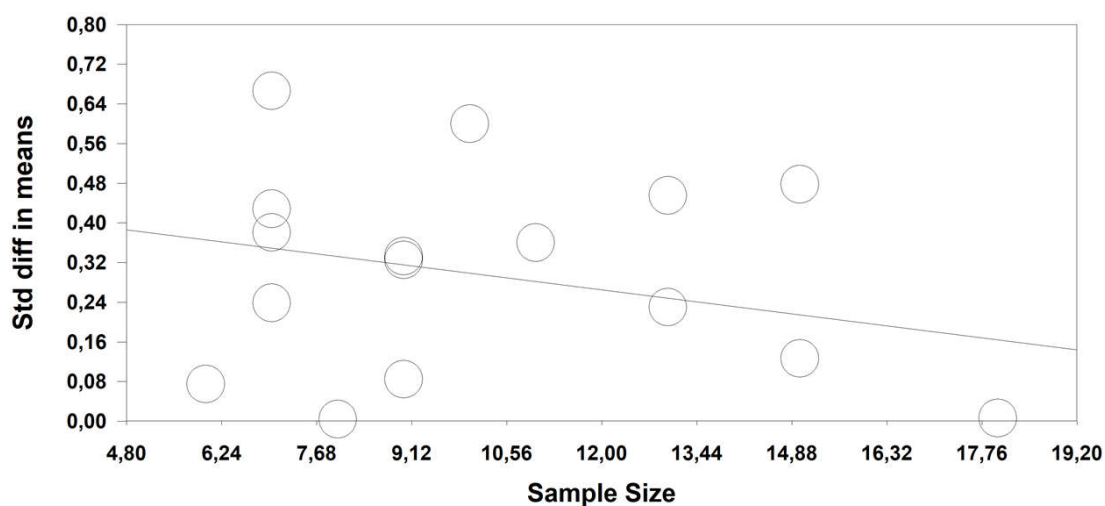


Figure 4. Metaregression of the number of participants (sample size) on standard differences in means (Std diff in means).

5. Discussion

5.1. Summary of Main Results

Six RCT studies evaluating the effects of an HRV-guided training intervention on endurance athletes were included in this review. The results of the meta-analyses provide some evidence that either HRV-guided training or traditional training may improve their performance in terms of VO_{2max} (HRV-G: ES = 0.402, $p < 0.0001$; CG: ES = 0.215, $p < 0.0001$). However, more favorable outcomes ($p < 0.0001$) for the experimental groups compared to the control groups were recorded across the studies. Moderators indicated larger effect sizes for interventions involving amateur endurance athletes (ES = 0.36, $p < 0.0001$) and women (ES = 0.40, $p < 0.0001$). On the other hand, the sample size of the studies was directly related to the ES magnitude ($p < 0.0001$).

5.2. Overall Completeness and Applicability of the Evidence

The total sample size of the studies meeting our original inclusion criteria was sufficiently large to warrant restricting the results to a meta-analysis of the RCTs. Data on the primary outcome (VO_{2max}) were measured directly using a gas exchange analysis system and a maximal test in each study. This is the most accurate way to obtain cardiorespiratory data. However, some studies

implemented this test using a treadmill (Kiviniemi_2007, Nuutila_2017, Schmitt_2017, and Vesterinen_2016) and others using a cycle ergometer (Javaloyes_2019 and Kiviniemi_2010). In the first case, training was based on running (Kiviniemi_2007, Nuutila_2017, and Vesterinen_2016) and skiing (Schmitt_2017), which implies similar technical execution in the test. In the second case, the Javaloyes_2019 study was carried out on cyclists, whereas the Kiviniemi_2010 study sample was composed of runners. Statistical improvements regarding VO_{2max} were found in the Kiviniemi_2007 and Kiviniemi_2010 studies. However, the specificity of the test may be a source of variability and potential imprecision in the second study results. Following the training specificity principle [37], the body's physiological and metabolic responses and training adaptations are specific to the type of exercise and the muscle groups involved. Thus, the evaluation method should be as similar as possible to the training in order to obtain the most reliable results. This needs to be taken into account when interpreting the results.

Despite the intervention durations being quite homogeneous in the included studies (eight weeks for each study apart from Kiviniemi_2007 and Schmitt_2017), the total duration of the training process, preparation weeks included, endurance sport modality, and training intensities used for the control group (standard training) were different. There was also a marked heterogeneity in the sample of the included studies: elite (Javaloyes_2019 and Schmitt_2017) and amateur (Kiviniemi_2007, Kiviniemi_2010, Nuutila_2017, and Vesterinen_2016) participants, or samples comprising only men (Javaloyes_2019, Kiviniemi_2007, Kiviniemi_2010, and Nuutila_2017), women (Kiviniemi_2010), or men and women (Schmitt_2017 and Vesterinen_2016). A standardized training protocol should be recommended to ensure the optimal benefits regarding VO_{2max} .

5.3. Quality of the Evidence

The quality of the evidence from the included studies can be considered unclear. Despite each study being a randomized controlled trial, the sequence generation or the allocation concealment was considered skewed in half of them. The performance bias was high only in Javaloyes_20019, while the detection bias was unclear in all the studies because incomplete blinding was considered. Attrition was high in Kiviniemi_2010, Nuutila_2017, and Vesterinen_2016 because of the high follow-up rates. In addition, the reporting bias was generally unclear due to the lack of a registered protocol.

5.4. Potential Biases in the Review Process

Although the systematic nature of the review process followed here decreases the potential for bias, the risk of bias in the review process remains. The greatest risk of bias present in this review was the study selection; specifically, the decision to limit the inclusion criteria to individual endurance sports, thus reducing the number of studies included and causing a potential limitation in the results.

- Agreements and disagreements with other studies or reviews

Based on the results from this systematic review with meta-analysis, and in response to Research Question 1, it is not surprising that the meta-analyzed results regarding improvements in athletes' VO_{2max} were associated with both training methodologies. According to Bartlett, O'Connor, Pitchford, Torres-Ronda, and Robertson [2] and Heyward [37], adequate prescribed training should maximize athletic performance when the specificity, overload, progression, initial level, individualization, diminishing return, and reversibility principles are followed. However, it was also found that the individual training adaptation according to the endurance athletes' daily HRV scores produced better VO_{2max} results than the standardized prescribed training, which answers Research Question 2. As pointed out by Vesterinen et al. [4,9], not every athlete improves their VO_{2max} after standardized group training. Similarly, Gallo, Cormack, Gabbett, Williams, and Lorenzen [38] reported that, in footballers, the internal load (perceived effort) of each athlete was different for a given external load; this definitely affects their individual performance during training and will be reflected in their individual performance improvements. Thus, daily individual HRV monitoring and training

guidance balancing the sympathetic and parasympathetic autonomic nervous system leads to greater athletic performance in endurance athletes compared to standardized prescribed training. This is relevant if training optimization is the objective, supporting the idea that training should be prescribed appropriately to avoid overtraining and/or injury [38]. In the same vein, it is also interesting to point out that, according to studies such as Hulin, Gabbett, Lawson, Caputi, and Sampson [39] and Williams et al. [16], training individualization is also related to minimizing overuse and reducing the injury risk, which may be a correlative benefit in the pursuit of endurance athlete training optimization.

On the other hand, the meta-analyzed results show that VO_{2max} improvements were greater when the sample comprised amateur endurance athletes. This answers Research Question 3. According to the initial training level principle [37], individuals with a low initial level of physical fitness should achieve more significant relative increases than those of average or high levels. This is in accordance with the results of Sanchez-Sanchez et al. [40], where greater performance improvements were obtained in lower-level football players compared to the higher-level players, concluding that the lower the athlete's initial fitness level, the higher the available window of adaptability. Conversely, in the systematic review with meta-analysis by Hammami, Gabbett, Slimani, and Bouhleb [41], the athlete's level was not a determinant variable in terms of VO_{2max} enhancement since it improved if they were elite or amateur players. It should be noted that this review was conducted on football players and that randomized and nonrandomized controlled trials were included.

According to our meta-analyzed results, and in response to Research Question 4, there were higher effect sizes regarding VO_{2max} improvements when the sample was not mixed, especially in the case of women. There is controversy concerning the influence of sex on sport performance. Recent studies conducted on endurance athletes concluded that either sex was not a predictor variable of performance [42] or that performance between men and women was different in swimming, cycling, and running [43]. In the case of the present systematic review with meta-analyses, we consider that the initial level of the sample influenced the result, given that, in the Kiviniemi_2010 study, when female samples were analyzed, the participants were amateur level athletes. Thus, a higher relative performance increment is predictable based on the athletes' level.

6. Conclusions

6.1. Practical Implications

Training optimization to enhance performance in endurance athletes is a goal that is undergoing a constant process of improvement. Finding a procedure to objectively individualize the training would be ideal for achieving this goal. The meta-analyses results considered in this review suggest that HRV is a good indicator of physiological responses to training in endurance athletes. Consequently, using daily HRV scores for training individualization and prescription is an effective method for optimizing performance in endurance athletes. This is reflected in the improved VO_{2max} results when the training is guided by HRV, considering VO_{2max} as one of the main performance indicators. In addition, it should be taken into account that a lower initial athlete fitness level will be relevant in achieving greater VO_{2max} improvement. Although gender may be a variable that influences the performance gains, in our opinion, this result is primarily conditioned by the level of the athletes included in the analyzed studies. Therefore, we do not consider it to be a variable that clearly affects VO_{2max} improvements.

6.2. Research Implications

The results from this review suggest that, while there is evidence that HRV-guided training is effective at improving VO_{2max} in endurance athletes, there is still work to be done in terms of identifying the characteristics of the interventions that contribute to this effect and the characteristics of participants who are more likely to respond to such interventions. The most important point is that more research is required since only five studies were included in this review. Moreover, only

two of the studies used samples composed of elite endurance athletes, which gave different results regarding $\text{VO}_{2\text{max}}$ improvement. Consequently, the research should be extended to the professional field in order to clarify the effect of guiding training on $\text{VO}_{2\text{max}}$. This would also help to clarify whether the endurance sport modality is determinative of the $\text{VO}_{2\text{max}}$ enhancement when following this training methodology.

Using daily HRV scores to control the training load and intensity over eight weeks is enough to improve $\text{VO}_{2\text{max}}$ in endurance athletes. Nonetheless, the training protocol should be further standardized in terms of adjusting the number of preparation weeks or considering the measurement weeks within or around the training period, factors that determine the training duration. Moreover, the standardized training protocol used in the control groups varied between the studies, which considered low, moderate, or high training intensities, as well as different numbers of sessions per week and session durations. This might very well have influenced the $\text{VO}_{2\text{max}}$ results. Therefore, it is necessary to reach a consensus regarding a standardized training protocol to use in future studies. In this line, it has been recently published a protocol [44] that could clarify the studies design. Similarly, although each study in this review used the most accurate method available to obtain the cardiorespiratory data, in the future, we should consider using a measuring instrument that allows us to implement the most specific sport technique in order to minimize result variability and imprecision.

Regarding the quality of the studies, authors should consider: improving the sequence generation or allocation concealment, the blinding of the participants, personnel, and outcome assessors, the rates of follow-up loss, using statistical procedures such as intention-to-treat to minimize attrition bias, and registering their protocols before starting the randomized controlled trial.

Lastly, to reinforce knowledge regarding performance optimization in endurance athletes, a good way to supplement the effect of HRV-guided training might be to register the risk of injuries associated with overuse using tools such as the Oslo Sports Trauma Research Center Overuse Injury Questionnaire, since this considers additional aspects affecting the execution of athletes' training.

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Protocol

HRV-Guided Training for Professional Endurance Athletes: A Protocol for a Cluster-Randomized Controlled Trial

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Abstract: Physiological training responses depend on sympathetic (SNS) and parasympathetic nervous system (PNS) balance. This activity can be measured using heart rate variability (HRV). Such a measurement method can favor individualized training planning to improve athletes' performance. Recently, HRV-guided training has been implemented both on professional and amateur sportsmen and sportswomen with varied results. There is a dearth of studies involving professional endurance athletes following a defined HRV-guided training protocol. The objectives of the proposed protocol are: (i) to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period and (ii) to determine differences in the athletes' performance after following both training protocols. This will be a 12-week cluster-randomized controlled protocol in which professional athletes will be assigned to an HRV-based training group (HRV-G) or a traditional-based training group (TRAD-G). TRAD-G will train according to a predefined training program. HRV-G training will depend on the athletes' daily HRV. The maximal oxygen uptake (VO_{2max}) attained in an incremental treadmill test will be considered as the primary outcome. It is expected that this HRV-guided training protocol will improve functional performance in the high-level athletes, achieving better results than a traditional training method, and thus providing a good strategy for coaches of high-level athletes.

Keywords: HRV; endurance training; training performance; high level athletes; VO_{2max} ; running

1. Introduction

It is known that training is essential for improving physical performance [1] and that optimizing training for performance improvement in athletes is an important area of research within exercise physiology and sports medicine [2–4]. In this regard, different training methods for performance improvement have been tried and tested, such as intensified training [2,5,6] and submaximal tests [7]. However, it is also recognized that using the same standardized training program for a group of athletes can provoke a wide range of reactions in terms of performance and physiological adaptations [8,9].

As stated by Schmitt, Willis, Fardel, Coulmy, and Millet [10], an important component of the interindividual variability in physiological responses to standardized training is related to the balance between the parasympathetic (PNS) and sympathetic (SNS) activity of the autonomic nervous system (ANS) [11,12]. Heart rate variability (HRV) is one of the indicators that allows the noninvasive study

of autonomic nervous system activity in its sympathetic and parasympathetic branches [13,14]. HRV is understood as the variation in the time interval between two consecutive heartbeats. It is obtained by calculating the time interval that elapses between two consecutive R waves (i.e., RR interval fluctuation) on an electrocardiogram (ECG) [15]. The period between beats is not constant; consequently, high HRV values are associated with an efficient ANS, which promotes behavioral adaptation and cognitive flexibility during stress [16], whilst low HRV values are indicative of an inefficient ANS, resulting in maladaptive responses to stress and perceived threats [15]. Furthermore, HRV is considered to be an indicator of cardiovascular health level [17].

Given that the SNS is responsible for changes in heart rate (HR) due to stress, and that the HR is one of the first parameters used to control the body's functional capacity [18], HRV analysis has been established as a useful method for assessing the heart's ability to adapt to both endogenous and exogenous loads [19], and can be used for the individual assessment of responses to training loads. Indeed, in recent years, HRV has been used to analyze these imbalances between SNS and PNS in athletes [20] and to evaluate different aspects related to training [21] such as exercise intensity and duration [22], recovery and overtraining [23], training load [24] or psychophysiological profiles [25].

The control of training based on HRV, as an indicator of the precompetitive physical and psychological state in athletes, enables coaches and scientists to use these HRV records to adapt the recovery and training loads to each athlete in search of a better sports result. As indicated by Ortigosa-Márquez, Reigal, Carranque, and Hernández-Mendo [26], high HRV values indicate more parasympathetic than sympathetic activation in an athlete and, therefore, better recovery and preparation for dealing with high-intensity training sessions.

Traditionally, HRV has been measured with ECG [14]. One of the ways of quantifying HRV is through rMSSD (the root mean square of successive differences between adjacent RR intervals) [26] since it is a temporal statistical parameter that reports those variations occurring over the short term between RR intervals [27] and it is used to observe the influence of the SNP on the cardiovascular system [18]. Currently, there are other validated tools for determining HRV that facilitate measurement, such as the Kubios HRV, Elite HRV, Mobile Lab and HRV4Training applications (apps).

In recent years, experimental studies have been carried out evaluating HRV-guided training in endurance athletes. These studies have been conducted both on elite athletes in sports such as cycling [24] and skiing [10], and with amateur endurance athletes [28–31]. One should take into account the scarcity of studies that have been published to date, especially on elite-level endurance athletes, as well as the absence of a common protocol to follow in this type of research.

The protocol proposed in the present study contributes to the scientific literature in this field in several ways: (i) it proposes research focused on elite athletes, a sample population for which only two experimental studies have been carried out to date; (ii) the protocol is intended to research endurance runners, for which we are not aware of any research having been carried out on this type of sample and with these characteristics at the international level. Therefore, it is a novel study aiming to provide empirical support for HRV-guided training in long-distance runners, adapting daily training to the physiological responses of each individual athlete. The performance of these athletes could be compared to that of another group of long-distance runners who carry out traditional training over the same period of time.

Until just a few years ago, conducting research of this type, involving daily HRV measurements on each athlete and then adapting training based on these data, was only possible with the collaboration of high-cost laboratories. This study tests the use of noninvasive, commercial, low-cost and publicly accessible technology to evaluate the physiological responses obtained by adapting training to HRV.

Based on everything described above, we hypothesize that HRV-guided training will: (i) improve functional performance in high-level athletes and (ii) produce better performance results than a traditional training method. The objectives of the proposed protocol are: (i) to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period, and (ii) to determine differences in the athletes' performance after following both training protocols.

This will be a 12-week cluster-randomized controlled trial protocol in which professional athletes are assigned to either an HRV-based training group (HRV-G) or a traditional-based training group (TRAD-G). A block randomization method will be chosen to randomly assign participants to interventions in equally sized sample groups. This protocol has been designed following the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) Statement [32]. To describe the intervention, the TIDieR (Template for Intervention Description and Replication) checklist by Hoffmann et al. [33] has been used.

2. Materials and Methods

2.1. Study Setting

To detect an intervention-related effect in professional athletes, other studies with similar protocols [24,31] compared athletes from two clubs or associations. Similarly, our sample will comprise athletes from two sport institutions in Almería (Spain): the C.D. Atletas de Almería, based in the city of Almería (Spain) and the Asociación Espeleológica Velezana, based in Vélez Rubio (Spain).

2.2. Eligibility Criteria

The inclusion criteria for participating in the program will be: (i) to belong to the Spanish Athletics Federation; (ii) to have been training and competing in Spanish Athletics Federation competitions for at least two years; and (iii) to be in the first third of the classification for the last five races of the previous season. Regarding the exclusion criteria, the following will be taken into account: (i) having cardiovascular pathologies, abnormal blood pressure parameters or diagnosed respiratory problems; (ii) being treated for psychological problems, or regularly taking a drug(s) that has a direct or indirect effect on the nervous system (e.g., anxiolytics, antidepressants or neuroleptics); (iii) substance use that is not permitted by the International Association of Athletics Federations (IAAF); (iv) occasional consumption of medication for a disease related to the cardiorespiratory system (e.g., influenza) that might alter performance and (v) not performing at least 90% of the workouts during the intervention.

The trial steering members will be responsible for checking that the subjects interviewed meet the inclusion criteria. The Spanish Athletics Federation's medical team will certify that the subjects do not meet any of the exclusion criteria. After being informed of the study design and potential risks, all athletes will sign a written informed consent document. The model consent form is shown in Appendix A.

2.3. Interventions

Based on the methodology used by Javaloyes et al. [24] and Vesterinen et al. [31], the intervention will be divided into two training periods for both study groups (HRV-G and TRAD-G): a four-week preparation period (PR) and an eight-week training period (TR). Both will maintain the weekly training volume. The training carried out will mainly be running. The PR period will be common to both groups and will be a familiarization phase for the training sessions and their intensities. During this period, the training intensity will gradually increase for the first three weeks and then decrease in the fourth week. This will mean three weeks of overloading and one week of recovery (3:1). The training to be carried out by the athletes is presented in more detail in Table 1. In the TR period, each group will carry out the corresponding intervention. The TRAD-G group will train according to a predefined training program, which will include sessions carried out at low intensity (approximately 50% of the total), and other sessions of moderate and high intensity, with a structure similar to that carried out during the PR period (Table 2). The training prescribed to the HRV-G group will depend on the subjects' HRV, in accordance with authors such as Javaloyes et al. [24], Kiviniemi, Hautala, Kinnunen, and Tulppo [34], and Lamberts, Swart, Noakes, and Lambert [35].

Table 1. Periodization and training distribution for the heart rate variability group (HRV-G) and traditional-based training group (TRAD-G) during the preparation period (PR).

Weeks	High Intensity	Moderate Intensity	Low Intensity
1		90 min between VT1 and VT2	3–4 sessions between 30 and 35 min below VT1
2	4 × 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
3	50 min at VT2 4 × 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
4			3–4 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions will be performed with a 15- to 20-min warm up and 20 min of cooling down.

Table 2. Periodization and training distribution for TRAD-G during training period (TR).

Weeks	High Intensity	Moderate Intensity	Low Intensity
5	50 min at VT2		3–4 sessions between 30 and 35 min below VT1
6	4 × 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
7	50 min at VT2 4 × 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
8			3–4 sessions between 30 and 35 min below VT1
9	50 min at VT2		3–4 sessions between 30 and 35 min below VT1
10		90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
11	50 min at VT2 4 × 12 min > VT2/3-min rest	90 min between VT1 and VT2	2–3 sessions between 30 and 35 min below VT1
12			3–4 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions will be performed with a 15- to 20-min warm up and 20 min of cooling down. Approximately 50% of the total sessions will be at low intensity.

To quantify the HRV, a Smartphone application known as “HRV4Training” (see <http://www.hrv4training.com/>) will be used. This tool has been validated by Plews et al. [36], showing a low typical estimate error (CV% (90% CI) = 3.8 (3.1; 5.0)) and a clear electrocardiographical correlation ($r = 1.00$ (1.00; 1.00)). It provides the root mean sum of the successive differences between R – R intervals (rMSSD) data using photoplethysmography. rMSSD is more suitable and reliable than other indexes [13,37]; nonetheless, the HRV data will be transformed by taking the natural logarithm, thus allowing parametric statistical comparisons that assume a normal distribution. In this way, a 7-day rolling average will be calculated (LnrMSSD_{7-d}). The PR period will be used as a standardized phase to obtain the baseline LnrMSSD_{7-d} and its range of normality (upper and lower limits). Following the indications of Plews, Laursen, Kilding, and Buchheit [38], this will be calculated as the mean \pm 0.5 \times SD. During the TR period, the LnrMSSD_{7-d} will be calculated daily in order to adapt the training prescribed to the HRV-G athletes. Moreover, the range of normality will be updated weekly. If the LnrMSSD_{7-d} is within the range of normality, the athletes will perform a moderate or high-intensity session. If the weekly LnrMSSD_{7-d} average falls below the normal range, a low intensity workout or rest will be undertaken. Athletes will perform a maximum of two consecutive sessions of moderate or high intensity; likewise, they will not accumulate more than two consecutive rest sessions. The modified scheme of Kiviniemi et al. [34] presented in Figure 1 will be followed.

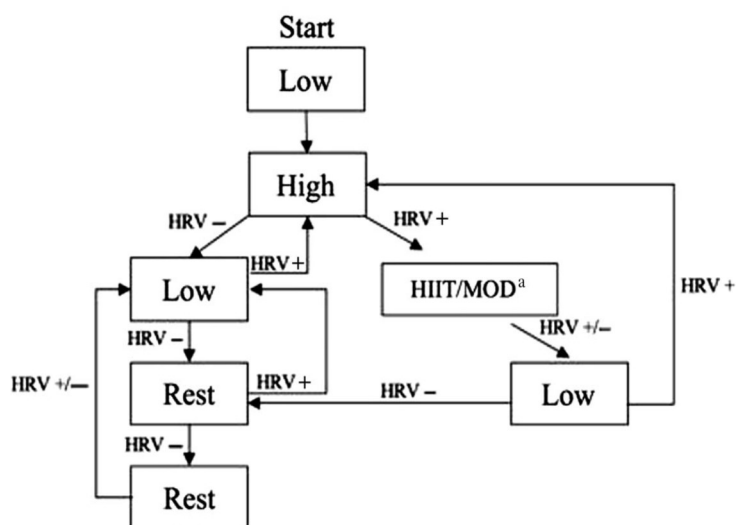


Figure 1. HRV-guided training schema. Modified from Kiviniemi et al. [34]. Note: When LnrMSSD_{7-d} remained inside their normal range, high-intensity or moderate-intensity training sessions were prescribed. If LnrMSSD_{7-d} fell outside their normal range (below), low intensity or rest were prescribed. HIIT/MOD = high/moderate-intensity interval training; HRV = heart rate variability; LnrMSSD_{7-d} = 7-day rolling average of the natural logarithm of the root-mean-squared differences of successive RR intervals.

In accordance with Javaloyes et al. [24], all participants will be instructed to measure their HRV data at home each morning after waking up and emptying their bladders. They will be instructed to lie in a supine position and not perform any further activity during the recordings. Data will be recorded over a 60-s period e.g., [36]. The daily control and recording of the rMSSD, as well as the LnrMSSD_{7-d} calculation used to prescribe the training of the HRV-G athletes, will always be carried out by the trial steering members. These members will receive the information from each athlete via phone or email and, in turn, will inform the HRV-G coach of the training intensity corresponding to each athlete. This procedure will also serve as a strategy for maintaining and monitoring the athletes' adherence to the training programs. Concomitant care, or any other intervention, will not be allowed during the trial for either the HRV-G or the TRAD-G. Athletes from both groups will carry out the training in their usual location.

2.4. Outcomes

The primary outcome of this study will be the maximal oxygen uptake (VO_{2max}) obtained in an incremental treadmill test. The secondary outcomes will be: the maximal speed in m/s, maximal heart rate, respiratory exchange ratio, ventilatory thresholds (VT1 and VT2) and their derived speed, heart rate, respiratory exchange ratio and VO_2 obtained in the incremental treadmill test. Other measurements considered as secondary outcomes will be: the time, speed, heart rate, rating of perceived exertion (RPE) and lactate in the 3000 m running test. Body composition and rMSSD will be considered as other variables.

Measurements will be taken before and after the training period, which will correspond to weeks 5 (pretest) and 12 (post-test). Over the assessment weeks, care will be taken that participants do not carry out any high-intensity training sessions. Each assessment week will consist of two testing sessions with a 48-h recovery period. The first testing session will include maximal graded exercise test and body composition measurements. In the second testing session, athletes will perform a 3000 m running test. The rMSSD will be measured daily, as explained in the intervention section.

The incremental treadmill test will be performed by the Physical Exercise and Human Performance Research Group at the University of Murcia (Spain). This is a more objective way of determining

physical fitness and represents the maximal performance capacity of an individual [39]. First, with the athlete in the supine position, a cardiovascular examination will be carried out at rest by means of cardiac auscultation, blood pressure and an electrocardiogram (ECG). The electrodes for recording the ECG and heart rate will be kept in place throughout the test. The Cardioline Cube[®] electrocardiograph will be used. To perform the incremental treadmill test, the Runner srl (Cavezzo Italy) treadmill will be used, as it was in other studies such as Ballesta-García, Martínez-González-Moro, Ramos-Campo, and Carrasco-Poyatos [40]. Similar to other studies, such as Nuutila et al. [30] or Vesterinen et al. [31], a prior 2-min aerobic warm up will be performed at 6 km/h. The test itself will start at a velocity of 7 km/h. The speed will be increased by 0.1 km/h every 6 s. The incline will remain at 1% throughout the test. The athletes will be encouraged to perform at maximum effort. The test will end when the subject can no longer run; the subject will indicate this with a hand gesture. The recovery phase will then begin at 4km/h for 3 min followed by rest for a further 2 min. The tests will be considered maximal and valid when the theoretical heart rate (220-age) exceeds 85% and the respiratory exchange ratio (RER) is greater than 1.15 [41]. During the stress test, the subjects will breathe through a mask connected to a gas analyzer (Metalyzer 3b[®], Cortex, Leipzig, Germany). All gas exchange parameters will be measured breath-by-breath and averaged every 30 s. The VO_{2max} will be defined as the oxygen consumption plateau [42]. The aerobic (VT1) and anaerobic (VT2) thresholds will be determined. Before each test, the gas analyzer will be manually calibrated. The test's maximal speed (V_{max}), maximal heart rate (HR_{max}), and respiratory exchange ratio (RER) will be recorded. The V_{max} or HR_{max} will be defined as the highest speed, or heart rate, reached for a finished stage. The speed, heart rate, respiratory exchange ratio and VO_2 at each ventilatory threshold will also be recorded as V_{VT1} , V_{VT2} , HR_{VT1} , HR_{VT2} , RER_{VT1} , RER_{VT2} , VO_{2VT1} , and VO_{2VT2} , respectively. All tests will be carried out under similar environmental conditions (an ambient temperature of 20–22 degrees).

As in other studies [30,43], the 3000 m running test will be conducted individually on a 400 m outdoor running track. Participants will be instructed to run at their maximum speed. Before the test, a 15 min standardized aerobic warm-up will be performed, consisting of running at a low to moderate intensity. Capillary blood samples (5 μ L) for blood lactate concentration analysis will be taken from the fingertip using a Scout+ analyzer (SensLab GmbH, Leipzig, Germany). Lactate is considered a useful indicator for measuring the metabolic cost and intensity of effort in aerobic-anaerobic sports [44]. Following Ribas [45], it will be considered in this test to relate it to running intensity. Lactate samples will be taken at four different points in time, in accordance with Rodríguez and Valero [46]: (i) just before the test (Lactate_{pre}), (ii) just after the test (Lactate_{post}), (iii) 3 min after the test (Lactate_{post3}) and (iv) 5 min after the test (Lactate_{post5}). Other variables, such as heart rate, time, speed, and rated perceived exertion (RPE), will also be measured. Heart rate will be recorded at five different points in time: (i) just before the test (HR_{pre}), (ii) just after the test (HR_{post}), (iii) 1 min after the test (HR_{post1}), (iv) 3 min after the test (HR_{post3}) and (v) 5 min after the test (HR_{post5}). The time will be recorded every 1000 m at three different points in time: (i) after running 1000 m, (ii) after running 2000 m and (iii) after running 3000 m (right at the end of the test). The speed will be calculated from these three points using the formula: speed = distance in m/time in seconds. The RPE will be measured at the end of the test using the modified Börg CR-10 scale of perceived exertion [47]. According to the authors, a 0 rating corresponds to rest; a 3 rating to moderate intensity; a 5 rating to hard intensity; a 7 rating to very hard intensity; and a 10 rating to maximal intensity. This tool has recently been determined as a stand-alone method for training load monitoring purposes in several sports and physical activities with men and women in different age categories (children, adolescents and adults) at various expertise levels [48].

Body composition will be analyzed just before the treadmill test using the InBody120 analyzer (Biospace Co. Ltd., Seoul, South Korea). Height will be measured using a measuring rod (Seca 213), and the body mass index (BMI) will be calculated according to the formula: $BMI = kg/m^2$.

The time schedule for enrolment, interventions and assessments is shown in Figure 2.

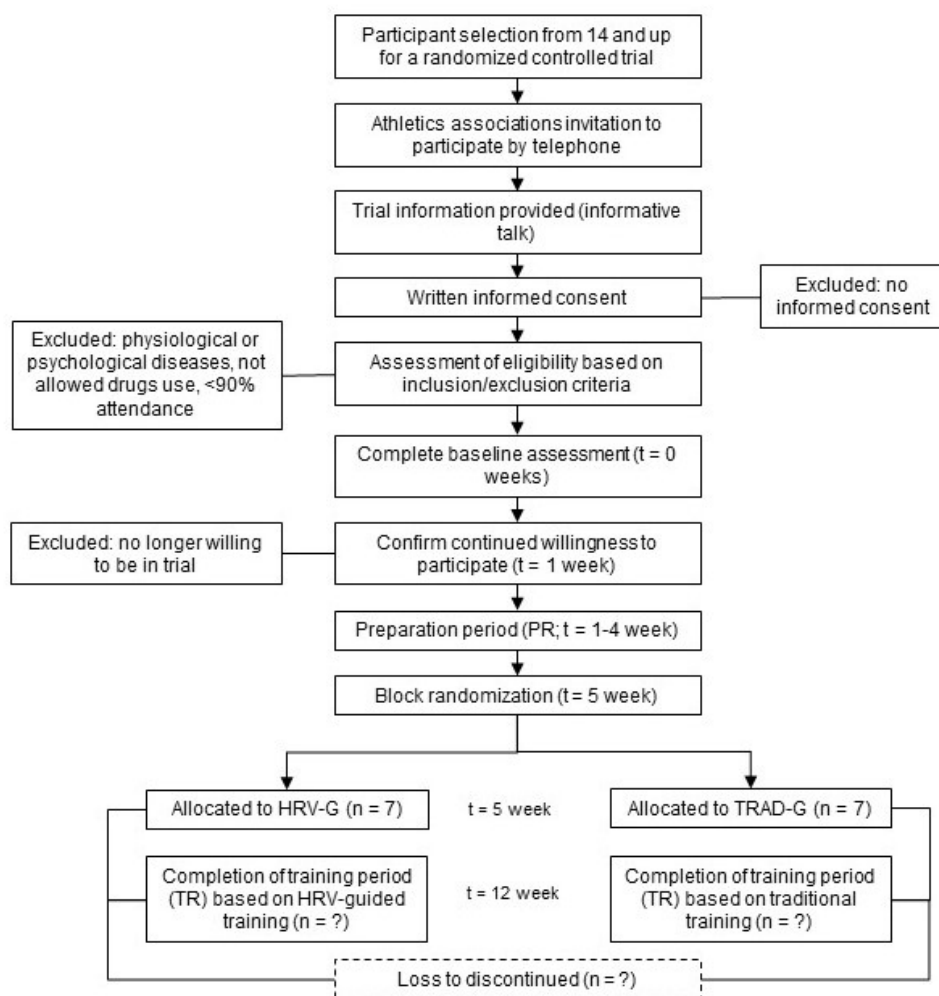


Figure 2. Schedule of enrolment, interventions and assessment. Note: HRV-G = heart rate variability-based training group; TRAD-G = traditional based training group; PR = preparation period; TR = training period.

2.5. Sample Size and Power

Calculations to establish the sample size will be performed using RStudio 3.15.0 software (PBC, Boston, USA). The significance level will be set at $p \leq 0.05$. According to the mean standard deviation established for VO_{2max} in a previous study [31] ($SD = 1.5 \text{ mL/kg/min}$) and an estimated error (d) of 1.1, a valid sample size providing a 95% confidence interval (CI) in each group will be 7 ($n = CI^2 \times d^2/SD^2$). Thus, a final sample size of 7 for each group will provide a power of 93% if between and within a variance of 2.

2.6. Recruitment

Each club or association involved in athletics in Almería (Spain) will be screened to identify the percentage of high-level or professionally federated athletes. When there are at least 7 high-level or professionally federated athletes, the club/association officers will be contacted by telephone to inform them of the study objective. Once they agree, an informative talk will be carried out with the athletes and the coach to inform them of the study objective, the time period in which it will take place, and the required commitment by the athletes to measure their daily HRV according to the established protocol, to attend the pre and post-test sessions and to attend at least 90% of the training sessions. The coaches will be informed of the required commitment to adapt each athlete's training session to the daily HRV score if their club is randomized into the HRV-G group. If they agree to participate, then they will have

to sign the written consent and meet the eligibility criteria necessary to be recruited into the study. The recruitment process will be conducted by the trial steering members.

2.7. Allocation and Blinding

A block randomization method will be used to allocate participants to the groups, which will contain equal sample sizes. The block size will be determined by the data monitoring committee according to the statistical power provided. Blocks will be chosen randomly by tossing a coin to determine the participants' assignment into the groups. This procedure will be carried out by the data monitoring committee. The athletes and the data monitoring committee will be blinded to the exercise group assignment.

2.8. Data Analysis

Data will be analyzed using Jamovi (Jamovi Project 2018, version 0.9.1.7, Sydney, Australia) and RStudio 3.15.0 software (PBC, Boston, MA, USA). Prior to data analysis, the Shapiro Wilk test and the Levene test will be performed to determine the normal distribution of the variables and the homogeneity of variance. Descriptive data will be reported as mean \pm SD and range. All the data will be analyzed based on the intention-to-treat principle (last observation carried forward). If the sample is normally distributed, Student's t-test will be calculated to compare variables before and after the intervention. For a variable to be considered as having a normal distribution, 95% of values will have to be within two standard deviations of the mean. If the sample is nonparametric, the U-Mann Whitney test will be used to compare variables before and after the intervention. The standardized mean differences (Cohen's effect size) will be calculated together with the 95% confidence intervals [49]. The effect sizes (ES) will be calculated using Cohen's d [49]. The relationship between variables will be assessed using the Pearson r correlation coefficient. If r is higher than 0.7, the determination coefficient (r^2) will be used to determine the percentage of Y variation with regard to the X variation. Significance will be accepted at $p \leq 0.05$.

2.9. Monitoring

A data monitoring committee will be set up during the study recruitment period. Interim analyses will be supplied to the committee in strict confidence, together with any other analyses that the committee may request. Based on the data monitoring committee's advice, the trial steering members will decide whether or not to modify the trial intake.

In our study, an adverse event will be defined as any untoward medical occurrence in a subject without regard to the possibility of a causal relationship. Adverse events will be collected after the subject has provided consent and enrolled in the study. If a subject experiences an adverse event after the informed consent document is signed (entry) but the subject has not started to receive study intervention, the event will be reported as not being related to the study's exercise program. For this study, the following will be considered serious adverse events: severe or permanent disability, use of prohibited substances and any other significant hazard as determined by the study members. Serious adverse events occurring after a subject has stopped participating in the study will not be reported unless the researchers feel that the event may have been caused by the study protocol procedure.

2.10. Ethics and Dissemination

This protocol, the informed consent template contained in Appendix A and other requested documents (if any) will be reviewed and approved by the Bioethical Committee at the University of Almería with respect to the scientific content and compliance with applicable research and human subject regulations. Following initial review and approval, this protocol will be reviewed by the researcher at least once a year at Clinicaltrials.org, where it is registered with the ID: NCT04150952.

Any protocol modifications which might have an impact on conducting the study, potentially benefit a subject or affect a subject's safety, or change the study objectives, study design, subject

population, sample sizes, study procedures, along with significant administrative issues, will require a formal amendment to the protocol. Such an amendment will be agreed upon by the Bioethical Committee at the University of Almería prior to implementation, and the clubs/associations enrolled will be notified. Administrative changes to the protocol that are minor corrections, and/or clarifications having no effect on the way the study is to be conducted, will be agreed upon by the researchers and documented in a memorandum. The Bioethical Committee at the University of Almería may be notified of the administrative changes.

All study-related information will be stored securely at the study site. All records that contain names or other personal identifiers, such as locator forms and informed consent forms, will be stored separately from the study records and identified by a code number. Forms, lists, logbooks, appointment books and any other listings that link participant ID numbers to other identifying information will be stored in a separate, locked file.

3. Discussion

This protocol describes the rationale, design, and methods of an HRV-guided training design for professional endurance athletes. It will allow accomplishment of a randomized controlled intervention to determine changes in the performance of high-level athletes after following an HRV-guided or a traditional training period. Moreover, the differences in the athletes' performance after following both training protocols will be determined. To design this protocol with professional endurance athletes, the guidelines described in Kiviniemi et al. [34] have been followed. This procedure has also been adapted in other professional sports such as cycling [24,50] and skiing [10], as well as to amateur endurance athletes [28–31].

This is the first time that this kind of protocol will be applied in endurance elite athletes. After its implementation, we expect that both high-level athletes groups (HRV-G and TRAD-G) improve: (i) VO_{2max} and other secondary outcomes measured in the treadmill test (the maximal speed in m/s, maximal heart rate, respiratory exchange ratio, or ventilatory thresholds), (ii) the time, speed, heart rate, rating of perceived exertion (RPE) and lactate in the 3000 m running test. Additionally, HRV-G will be better regarding performance results than the TRAD-G. These findings will suggest that training guidance balancing the sympathetic and parasympathetic autonomic nervous system leads to greater athletic performance in endurance athletes compared to standardized prescribed training. This is relevant for training optimization and for minimizing overuse and reducing injury risk.

4. Conclusions

Experimental research conducted in recent years shows that improvements in variables related with athletes' performance (e.g., VO_{2max}) can be obtained through HRV-guided training. However, accordingly to these studies, results do not allow a consensus to be established regarding the performance benefits of HRV-guided training for endurance athletes.

From studies carried out until now, this article describes a novel protocol to conduct a randomized controlled trial with endurance athletes. So far, no other HRV-guided training research has been conducted with these types of professional athletes. Besides, this protocol proposes to use emergent technologies in the training and the research fields, such as smartphone applications; in this case, HRV4training, an app scientifically validated that allows calculation of the daily HRV measurement for each athlete. Although more research is needed, the implementation of the protocol described here will contribute to this scientific field of study.

Author Contributions: Conceptualization, M.C.-P., A.G.-Q. and A.G.-G.; methodology, M.C.-P., A.G.-Q., A.G.-G. and I.M.-G.-M.; investigation, M.C.-P., A.G.-G. and A.G.-Q.; writing—original draft preparation, M.C.-P. and A.G.-Q.; writing—review and editing, A.G.-G. and I.M.-G.-M. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

Mr. Antonio Granero-Gallegos and Ms. María Carrasco-Poyatos, lead researchers on the project: Physiological and psychological effects from heart rate variability-based training in professional athletes have informed:

Mr/Ms ID. , about the present study's general proceeding, its objectives, duration, purpose, inclusion and exclusion criteria, associated risks and benefits, as well as the possibility of leaving it without having to give reasons. In knowledge of all the above, and the measures that will be adopted to protect the participant's personal data, according to the current regulations,

I CONSENT to participate in the present research.

Signed: Mr/Ms ID.

Signed: Mr. Antonio Granero-Gallegos, ID 23245990M and Ms. María-Carrasco-Poyatos, ID 75096834V

Project Leading Researcher.

Antonio Granero-Gallegos

María Carrasco-Poyatos

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Heart rate variability-guided training in professional runners: Effects on performance and vagal modulation

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ABSTRACT

Purpose: To analyze the training structure following a heart rate variability (HRV) -guided training or traditional training protocol, determining their effects on the cardiovascular performance of professional endurance runners, and describing the vagal modulation interaction.

Methods: This was an 8-week cluster-randomized controlled trial. Twelve professional endurance runners were randomly assigned to an HRV-guided training group (HRV-G; $n = 6$) or a traditional training group (TRAD-G; $n = 6$). The training methodology followed by the HRV-G was determined by their daily HRV scores. Training intensities were recorded daily. HRV4Training was used to register the rMSSD every morning and during a 60-second period. Cardiovascular outcomes were obtained through an incremental treadmill test. The primary outcome was the maximal oxygen uptake (VO₂max).

Results: Total training volume was significantly higher in TRAD-G, but moderate intensity training was significantly higher in HRV-G ($X \pm SD_{Dif} = 1.98 \pm 0.1\%$; $P = 0.006$; $d = 1.22$) and low intensity training in TRAD-G ($X \pm SD_{Dif} = 2.03 \pm 0.74\%$; $P = 0.004$; $d = 1.36$). The maximal velocity increased significantly in HRV-G ($P = 0.027$, $d = 0.66$), while the respiratory exchange ratio increased in TRAD-G ($P = 0.017$, $d = 1$). There was a small effect on the LnRMSSD increment ($P = 0.365$, $d = 0.4$) in HRV-G. There were statistical inter-group differences in the Δ maximal heart rate when Δ LnRMSSD was considered as a covariable ($F = 7.58$; $P = 0.025$; $d = 0.487$). There were significant and indirect correlations of LnRMSSD_{TEST} with VO₂max ($r = -0.656$, $P = 0.02$), Δ LnRMSSD with Δ VO₂max ($r = -0.606$, $P = 0.037$), and Δ LnRMSSD_{CV} with Δ VENT ($r = -0.790$, $P = 0.002$).

Conclusions: higher HRV scores suggest better cardiovascular adaptations due to higher training intensities, favoring HRV as a measure to optimize individualized training in professional runners.

1. Introduction

The key to achieving optimal adaptations in endurance training is to find the most efficient stimulus [1]. The importance of macrocycle (weeks or months) training periodization is well-recognized in endurance training interventions. However, some experimental studies [2,3] highlight the relevance of microcycle (day-to-day) periodization in order to achieve better cardiovascular adaptations, especially when operating under the constraints of limited resources and time.

Monitoring the cardiac autonomic nervous system (ANS) is currently

being used as a promising method to optimize the training prescription, as variability in physiological responses to standardized training is related to the balance between parasympathetic and sympathetic activity [4]. The parasympathetic branch of the ANS is of particular importance as it represents the body's integrative system for adaptively self-regulating and maintaining homeostatic balance [5]. In this regard, heart rate variability (HRV) is commonly used as an indicator of day-to-day autonomic nervous system activity. Higher parasympathetic activation is reflected in higher HRV values, which are indicative of better recovery [6]. Recently, validated smartphone tools have become

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available, such as HRV4Training, that have enabled day-to-day HRV recording, making it easier to adapt the training loads to each athlete for performance optimization.

According to Seiler et al. [7], the recovery of well-trained runners is better than that of novices since endurance training adaptations are associated with reduced sympathetic branch activation [1]. Therefore, endurance training adaptations should induce higher HRV values. If the HRV is recorded daily, higher intensity sessions might be included in the training periodization, with the athletes benefiting from greater peripheral and central physiological adaptations associated to more polarized training. According to this assumption, a recent meta-analysis [8] showed that the individual training adaptation, based on the endurance athletes' daily HRV scores, produced better VO_{2max} results than the standardized training prescribed, with the training level being a determinant factor. However, when the sample is composed of runners, there is still a lack of consensus regarding the training design, when it is HRV-guided or traditionally prescribed [9,10], in terms of the total volume achieved and the proportion of training performed at high, moderate or low intensities. It is also unclear as to whether HRV-guided training induces better physiological performance, leading to a higher running velocity [9,2,10] or enhancing the oxygen consumption [2,3,9]. Moreover, HRV does not always improve statistically after an HRV-guided intervention [2,3,11,9], and its correlation to performance outcomes has only been analyzed in one study [10].

Therefore, the aims of the present study were: 1) to analyze the training structure (volume and intensity) performed by an HRV-guided training group and a traditional training group (HRV-G and TRAD-G), comprising professional endurance runners; 2) to determine the effect of the two training methods on the athletes' cardiovascular performance and vagal modulation; and 3) to describe the influence of HRV on the performance outcomes in both groups. It was hypothesized that HRV-G will train at higher intensity and lower volume. This will have a positive impact on their cardiovascular performance and in their recovery, leading to higher HRV scores. Moreover, HRV scores will be associated to runners' performance.

2. Materials and methods

2.1. Design

This study was an 8-week cluster-randomized controlled trial in which professional endurance runners were assigned to an HRV-based training group (HRV-G; $n = 6$) or a traditional training group (TRAD-G; $n = 6$). The trial design followed the CONSORT guidelines [12]. The study was approved by the University of Almería's Bioethics Committee (UALBIO2019/026). The study was registered prospectively with ClinicalTrials.gov (NCT04150952).

2.2. Participants

The professional endurance runners were recruited from two sport institutions in Almería province (Spain): the C.D. Atletas de Almería, based in the city of Almería (Spain) and the Asociación Espeleológica Velezana, based in Vélez Rubio (Spain). The HRV data were collected in Almería whereas the incremental treadmill test data were collected at the University of Murcia (Spain). The inclusion and exclusion criteria are detailed elsewhere [13]. All participants signed a consent form before beginning the study.

2.3. Interventions

The training design followed in the preparation period (PR; the first four weeks) for both groups, or in the intervention period (TR; the following eight weeks) for the HRV-G and TRAD-G groups, are detailed in Tables 1, 2, 3. The training prescription methodology followed by TRAD-G group was based on a traditional training prescription,

Table 1

Periodization and training distribution for HRV-G and TRAD-G during PR period.

Weeks	High intensity	Moderate intensity	Low intensity	
1		8 × 2 min > VT2/1-min rec	110 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
2	30 min at VT2	10 × 2 min > VT2/1-min rec	120 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
3	40 min at VT2	12 × 2 min > VT2/1-min rec	140 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
4		4 × 3 min > VT2/1-min rec	70 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions were performed with a 15- to 20-minute warm up and 20 min of cooling down.

Table 2

Periodization and training distribution for TRAD-G during TR period.

Weeks	High intensity	Moderate intensity	Low intensity	
5	20 min at VT2	70 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1	
6	30 min at VT2	8 × 3 min > VT2/2-min rec	90 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
7	40 min at VT2	10 × 3 min > VT2/2-min rec	110 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
8	50 min at VT2	12 × 3 min > VT2/130-min rec	140 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
9	50 min al VT2	10 × 3 min > VT2/1-min rec	140 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
10	40 min al VT2	8 × 3 min > VT2/1-min rec	120 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
11	30 min al VT2	6 × 3 min > VT2/1-min rec	110 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
12				4-5 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions were performed with a 15- to 20-minute warm up and 20 min of cooling down. Approximately 50% of the total sessions will be at low intensity.

meanwhile the HRV-G group training was determined by their daily HRV scores and based on the Kiviniemi et al. [2] scheme modified by Javaloyes et al. [14]. More information is detailed in the protocol of Carrasco et al. [13]. Figs. 1 and 2 are an example of the HRV fluctuations during the TR period in HRV-G and TRAD-G, respectively.

2.4. Outcomes

The maximal oxygen uptake relative to weight (VO_{2max}) was considered as the study's primary outcome. The secondary outcomes recorded were: i) the maximal velocity scores (V_{max}), the heart rate (HR_{max}), the percentage of the heart rate relative to the maximum (HR_{rel}), the maximal oxygen uptake in liters per minute (VO_{2max}), the respiratory exchange ratio (RER), and the ventilatory rate (VENT). Other variables considered were the body mass index, the LnrMSSD, and the training volume at high (\geq Ventilatory Threshold 2, VT2, or Zone 3),

Table 3
Periodization and training distribution for HRV-G during TR period.

Weeks	High intensity	Moderate intensity	Low intensity
5	20 min at VT2	50 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
6	40 min at VT2	70 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
7	50 min at VT2	90 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
8	60 min at VT2	110 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
9	60 min al VT2	110 min between VT1 and VT2	4-5 sessions between 30 and 35 min below VT1
10	50 min al VT2	100 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
11	40 min al VT2	80 min between VT1 and VT2	3-4 sessions between 30 and 35 min below VT1
12			4-5 sessions between 30 and 35 min below VT1

Note: VT1 = first ventilatory threshold; VT2 = second ventilatory threshold. High-intensity and moderate-intensity sessions were performed with a 15- to 20-minute warm up and 20 min of cooling down. Approximately 50% of the total sessions will be at low intensity.

moderate (between VT2 and VT1, or Zone 2) and low (\leq VT1 or Zone 1) intensities in the TR or PR periods.

The primary and secondary outcomes were obtained through an incremental treadmill test. This test procedure and the instruments involved are explained in the Carrasco et al. protocol [13] together with the HRV and body mass index measurements. According to Piatrikova et al. [15], the weekly average for the LnrMSSD calculation was implemented with its coefficient of variation ($LnrMSSD_{CV} = [LnrMSSD_{SD}/LnrMSSD_{MEAN}] \times 100$) since it was also found to be a marker of day-to-day HRV variability and representative of training adaptation (a higher LnrMSSD_{CV} was associated with a less optimal response). Moreover, the final test-day LnrMSSD was considered (LnrMSSD_{TEST}). The total training volume and the training volume at each intensity were recorded daily by the respective coaches.

The pre-test and post-test were conducted in weeks 5 and 12, coinciding with the first and the last weeks of the training period, in which low to moderate training sessions were carried out by the runners.

2.5. Sample size and power

According to the mean standard deviation established for the VO_{2max} in our study (SD = 6.19 ml/kg/min) and an estimated error (*d*) of 4.8, a total of six subjects was determined to be a valid sample size for each group, providing a 95% confidence interval (CI) ($n = CI^2 \times d^2/SD^2$). Thus, a final sample size of 6 for each group provides a power of 88% if it is between and within a variance of 2. Calculations to establish the sample size were performed using RStudio 3.15.0 software. The significance level was set at $P \leq 0.05$.

2.6. Randomization and blinding

Participants were randomly allocated to each group using a block randomization method. The treatment was randomly assigned to the groups by coin tossing. The block size was determined according to the statistical power provided, making sure that the sample size was the same for each group (HRV-G, TRAD-G, $n = 6$). This process was implemented by the principal investigator. Participants and research staff were blinded. Only the coaches knew about the groups so that they could adapt, where appropriate, the training sessions to the daily HRV recording.

2.7. Statistical methods

Prior to data analysis, the Shapiro-Wilk test was used to determine the normal distribution of the variables. Levene's test was also performed to determine the homogeneity of variance. Descriptive data are presented as mean \pm SD and range. The variables were normally distributed, so to test for differences between the groups at baseline, an unpaired two-tailed *t*-test was used. To compare the groups after the intervention, an analysis of covariance (ANCOVA) was used with the baseline values, while the final test-day LnrMSSD (LnrMSSD_{TEST}), the change in LnrMSSD ($\Delta LnrMSSD$), and the change in its coefficient of variation ($\Delta LnrMSSD_{CV}$), were included as co-variables in order to adjust for any potential influence in the dependent variables. To test for differences within groups, a paired two-tailed *t*-test was used. Owing to our small sample size, the standardized mean differences (Cohen's effect size) were calculated together with the 95% confidence intervals so as to

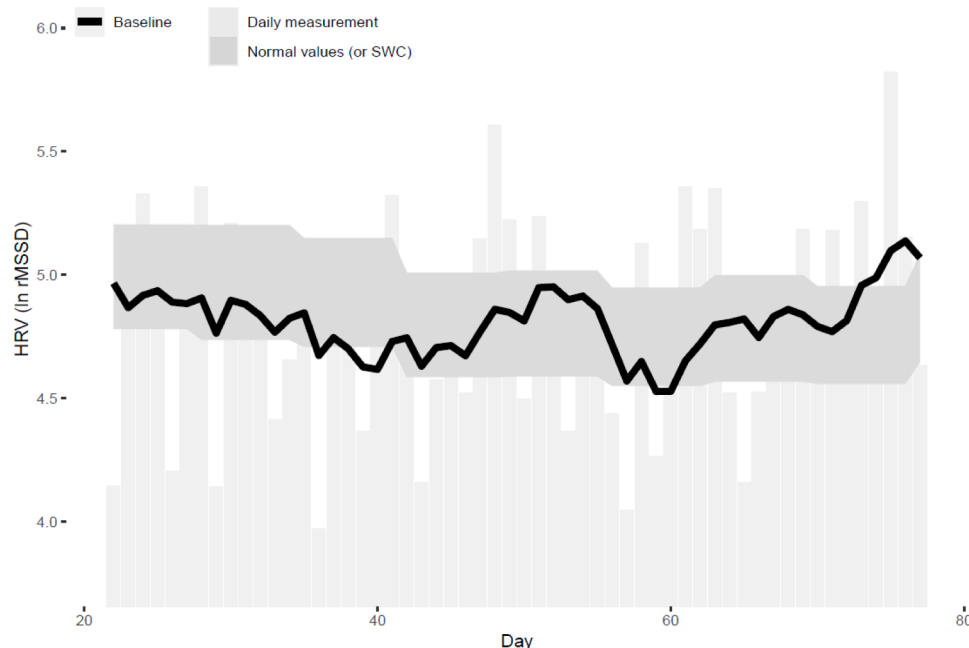


Fig. 1. Example of individual response of HRV in an HRV-G runner.

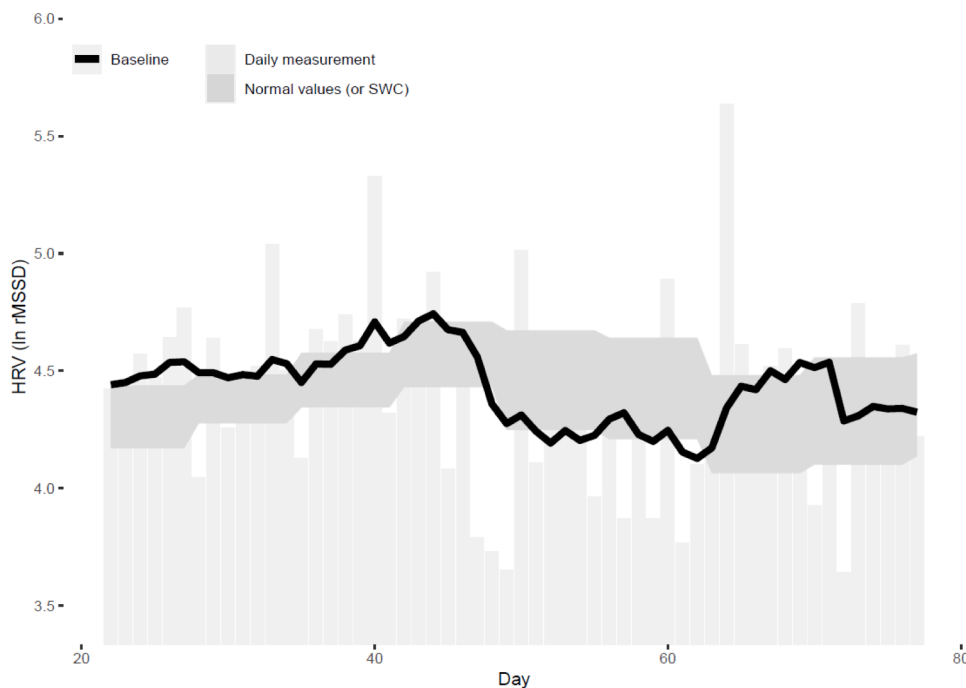


Fig. 2. Example of individual response of HRV in a TRAD-G runner.

describe if the treatment effect had a relevant magnitude. An effect size (ES) value of 0.20 indicates a small effect, 0.50 a moderate effect, and 0.8 a large effect [16]. A bivariate Pearson correlation was utilized to assess the relationships between the LnRMSSD and the physiological outcomes. The correlation thresholds were 0.1, small; 0.3, moderate; 0.5, large; 0.7, very large; and 0.9, nearly perfect [17]. The level of significance was set at $P \leq 0.05$. The statistical analyses were conducted using the IBM SPSS Statistics V.24 program for Windows (SPSS Inc., IL, USA), Microsoft Excel 2010 (Microsoft Corporation, WA, USA) and Rstudio 3.15.0 software.

3. Results

The participant flow as this randomized trial progressed is detailed in Fig. 3. There were no losses or exclusions after the randomization process, thus 12 participants completed the study (HRV-G, $n = 6$; TRAD-G, $n = 6$). Recruitment was carried out in September 2019 and the intervention was conducted in the months of October and November 2019. There were no special harm events that forced the intervention to stop. The baseline characteristics of the two groups are shown in Table 4.

Results for Objective 1: Regarding the training volume and intensity

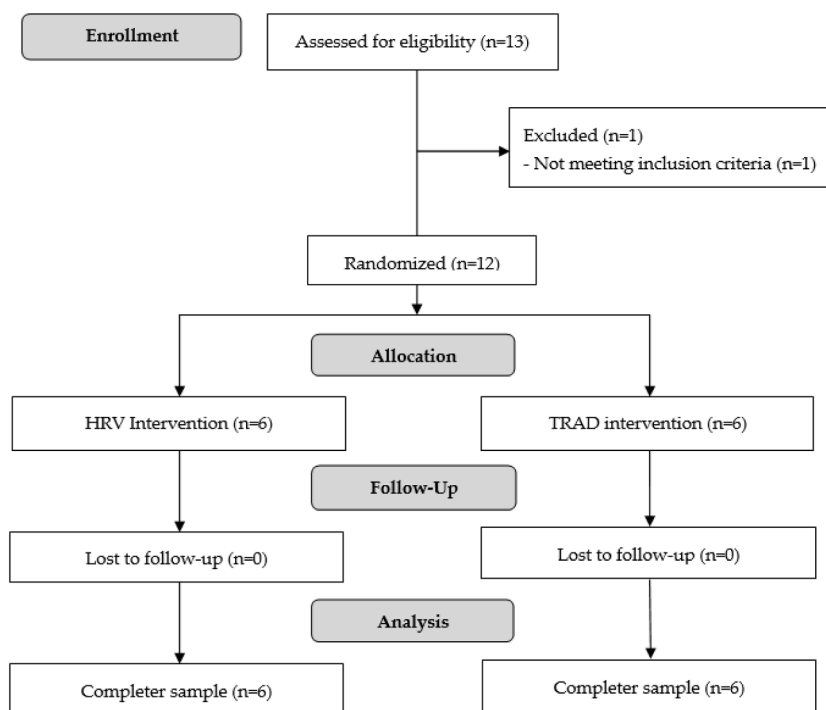


Fig. 3. Flow diagram of the progress of the randomized trial.

in the TR period, the HRV-G group accomplished a total of 46 h, 17 min, and 24 s (46 h 17' 24") distributed as 7 h 43' 12" at \geq VT2, 17 h 29' 24" between VT2 and VT1, and 21 h 4' 48" at \leq VT1, which results in 16.68%, 37.77%, and 45.54% of the training time at the respective intensities. For its part, the TRAD-G group trained for 56 h 46' 12" distributed as 9 h 26' 24", 20 h 19' 48", and 27 h for the \geq VT2, between VT2 and VT1, and \leq VT1 intensities, respectively, which results in 16.63%, 35.81%, and 47.56% of the respective intensities. There were significant differences favoring TRAD-G regarding the total training volume ($X \pm SD_{Dif} = 629 \pm 12.96$ min; $P = 4.78 \times 10^{-7}$; $d = 6.57$ [3.71, 9.44]) and the training minutes at \geq VT2 ($X \pm SD_{Dif} = 103.17 \pm 7.05$ min; $P = 2 \times 10^{-6}$; $d = 5.63$ [3.1, 8.15]), between VT2 and VT1 ($X \pm SD_{Dif} = 170.83 \pm 19.36$ min; $P = 6 \times 10^{-6}$; $d = 4.98$ [2.69, 7.28]), and at \leq VT1 ($X \pm SD_{Dif} = 355 \pm 23.21$ min; $P = 3 \times 10^{-6}$; $d = 5.38$ [2.95, 7.82]) (Fig. 4). On the other hand, the training time in Zone 2 was significantly higher in HRV-G ($X \pm SD_{Dif} = 1.98 \pm 0.1\%$; $P = 0.006$; $d = 1.22$ [-0.009, 2.46]), while TRAD-G spent significantly more time training in Zone 1 ($X \pm SD_{Dif} = 2.03 \pm 0.74\%$; $P = 0.004$; $d = 1.36$ [0.11, 2.62]) (Fig. 5). There were also statistical differences between groups regarding the training time at moderate-to-high intensity (Zones 2 + 3) favoring HRV-G ($X \pm SD_{Dif} = 2.03 \pm 0.74\%$; $P = 0.004$; $d = 1.36$ [0.11, 2.62]).

Results for Objective 2: At the end of the intervention, there were no significant inter-group differences in the primary or secondary outcomes. In contrast, the Vmax increased significantly in HRV-G ($P =$

Table 4
Characteristics at baseline.

Variables	n	Mean	SD	Min	Max
Age (years)					
HRV-G	6	31.5	8.36	25	46
TRAD-G	6	31.2	10.3	19	42
Height (cm)					
HRV-G	6	177	6.66	165	185
TRAD-G	6	173.2	9.12	160.5	186.5
Weight (kg)					
HRV-G	6	70.9	7.87	57.3	77.1
TRAD-G	6	65.9	12.03	54.6	88
BMI (kg/m2)					
HRV-G	6	22.5	1.46	20.8	24
TRAD-G	6	21.8	2.43	18.8	25.3
LnrMSSD (ms)					
HRV-G	6	4.41	0.32	4.07	4.99
TRAD-G	6	4.44	0.31	4.15	4.96
Vmax (m/s)					
HRV-G	6	18.6	1.46	17	21.8
TRAD-G	6	19.2	1.79	16,00	21.2
HRmax (bpm)					
HRV-G	6	187.8	15.65	161	205
TRAD-G	6	180	11.61	159,00	192
HRrel (%)					
HRV-G	6	99.5	4.71	92.5	105.1
TRAD-G	6	95.3	3.97	89.3	100.6
VO₂max (ml/kg/min)					
HRV-G	6	58.8	4.99	53,0	64,0
TRAD-G	6	61	7.37	52,0	70,0
VO₂max (l/min)					
HRV-G	6	4.16	0.4	3.61	4.84
TRAD-G	6	3.97	0.44	3.43	4.58
RER (l)					
HRV-G	6	1.12	0.03	1.1	1.17
TRAD-G	6	1.12	0.03	1.07	1.17
VENT (bpm)					
HRV-G	6	146.83	14.63	128	166
TRAD-G	6	142.78	11.35	128	160,00

Note: BMI = body mass index (kg/m2), LnrMSSD = the Neperian logarithm of the square root of the mean value of the sum of the squares of the differences between the adjacent RR intervals (ms). Treadmill test outcomes: Vmax = maximal velocity (km/h), HRmax = maximal heart rate (bpm), HRrel = the percentage of the heart rate relative to the maximum (%), VO₂max = maximal oxygen uptake (l/min) and relative to weight (ml/kg/min), RER = respiratory exchange ratio (l), VENT = ventilatory rate (bpm).

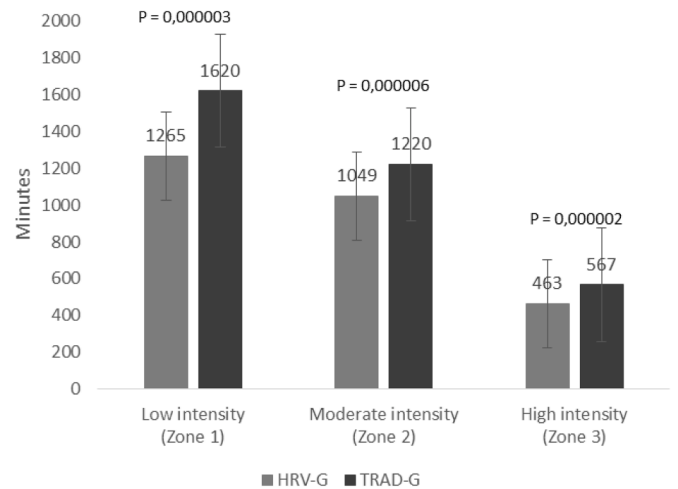


Fig. 4. Total training time at low-, moderate-, and high- intensities for HRV-G and TRAD-G.

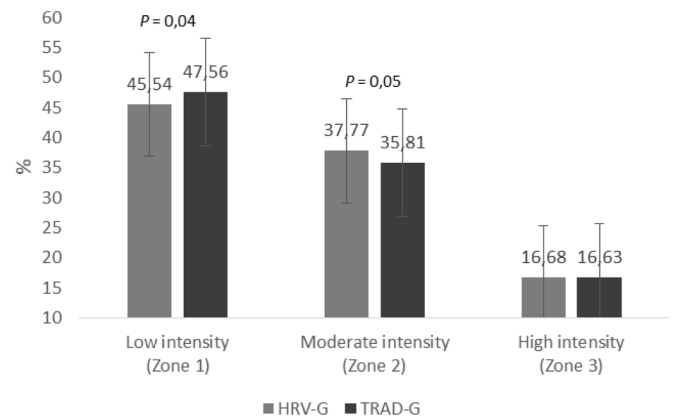


Fig. 5. The proportion of training at low-, moderate-, and high- intensities for HRV-G and TRAD-G.

0.027, $d = 0.66$), and the RER increased significantly in TRAD-G ($P = 0.017$, $d = 1$). Moreover, in HRV-G, there was a large effect in the VENT reduction ($P = 0.11$, $d = 1.01$) and a small effect in the VO₂max reduction ($P = 0.267$, $d = 0.32$) as well as an increment in the LnrMSSD ($P = 0.365$, $d = 0.4$). These results are shown in Table 5.

Results for Objective 3: The ANCOVA results showed that there were significant differences between groups in ΔHR_{max} ($F = 7.58$; $P = 0.025$; $d = 0.487$) and ΔHR_{rel} ($F = 8.47$, $P = 0.02$, $d = 0.514$) when the covariable was $\Delta LnrMSSD$, but also when the covariable was the baseline result (ΔHR_{max} : $F = 5.646$, $P = 0.045$, $d = 0.414$; ΔHR_{rel} : $F = 7.18$, $P = 0.028$, $d = 0.473$) (Table 6). On the other hand, a significant interaction of the LnrMSSD_{TEST} with the post-test HR_{max} ($F = 5.305$, $P = 0.05$, $d = 0.399$) and the VO₂max ($F = 10.653$, $P = 0.011$, $d = 0.571$) was found, along with a significant interaction of $\Delta LnrMSSD_{CV}$ with $\Delta VENT$ ($F = 11.663$, $P = 0.009$, $d = 0.593$), but these were not statistically different. Moreover, there were significant and indirect correlations of the LnrMSSD_{TEST} with the post-test VO₂max ($r = -0.656$, $P = 0.02$), the $\Delta LnrMSSD$ with the ΔVO_{2max} ($r = -0.606$, $P = 0.037$), and the $\Delta LnrMSSD_{CV}$ with the $\Delta VENT$ ($r = -0.790$, $P = 0.002$).

4. Discussion

The objectives of the present study were to analyze the training structure following an HRV-guided training or a traditional training methodology, determining their effect on performance in a sample of

Table 5
Differences in pre- and post-test for the BMI, the heart rate variability and the ergometer test outcomes on each group (HRV-G and TRAD-G).

Variables	Pre-training			Post-Training		p	95% CI for Mean Difference		Cohen's d
	n	Mean	SD	Mean	SD		Lower	Upper	
BMI (kg/m²)									
HRV-G	6	22.57	1.46	22.85	1.37	0.301	-0.91	0.34	0.63
TRAD-G	6	21.85	2.43	21.85	2.9	1000	-0.57	0.57	0,00
LnrMSSD (ms)									
HRV-G	6	4.41	0.32	4.57	0.47	0.365	-0.56	0.25	0.398
TRAD-G	6	4.44	0.31	4.41	0.28	0.857	-0.38	0.44	0.102
Vmax (m/s)									
HRV-G	6	18.6	1.46	19.8	2.11	0.027	0.21	2.25	0.66
TRAD-G	6	19.2	1.79	19.2	1.19	0.986	-2.35	2.38	0,00
HRmax (bpm)									
HRV-G	6	187.8	15.65	188.3	16.45	0.749	-4.29	3.29	0.03
TRAD-G	6	180	11.61	181,00	11.95	0.733	-8.11	6.12	0.08
HRrel (%)									
HRV-G	6	99.5	4.71	99.8	4.95	0.749	-2.27	1.75	0.06
TRAD-G	6	95.3	3.97	95.8	4.21	0.732	-4.31	3.25	0.15
VO₂max (ml/kg/min)									
HRV-G	6	58.8	4.99	57.2	5.11	0.267	-1.76	5.09	0.32
TRAD-G	6	61	7.37	61	7.29	1000	-1.33	1.33	0,00
VO₂max (l/min)									
HRV-G	6	4.16	0.4	4.08	0.42	0.228	-0.06	0.21	0.24
TRAD-G	6	3.97	0.44	3.96	0.42	0.87	-0.11	0.12	0.02
RER (l)									
HRV-G	6	1.12	0.03	1.13	0.07	0.574	-0.07	0.04	0.19
TRAD-G	6	1.12	0.03	1.15	0.03	0.017	-0.06	-0.01	1
VENT (bpm)									
HRV-G	6	146.83	14.63	139.56	16.63	0.11	-2.35	16.88	1.012
TRAD-G	6	142.78	11.35	142.48	10.13	0.883	-4.67	5.27	0.087

Note: BMI = body mass index (kg/m²), LnrMSSD = the Neperian logarithm of the square root of the mean value of the sum of the squares of the differences between the adjacent RR intervals (ms). Treadmill test outcomes: Vmax = maximal velocity (km/h). HRmax = maximal heart rate (bpm). HRrel = the percentage of the heart rate relative to the maximum (%). VO₂max = maximal oxygen uptake (l/min) and relative to weight (ml/kg/min). RER = respiratory exchange ratio (l). VENT = ventilatory rate (bpm).

Table 6
Differences between groups in the change (pre-post) in performance variables adjusted by the baseline scores, ΔLnrMSSD and ΔLnrMSSDcv.

Variable	n	Change (pre-post)		Group*Baseline			Group*ΔLnrMSSD			Group*ΔLnrMSSDcv		
		Mean	SD	F	p	ES η ²	F	p	ES η ²	F	p	ES η ²
ΔLnrMSSD (ms)												
HRV-G	6	-0.16	0.38	0.957	0.357	0.107				0.227	0.646	0.028
TRAD-G	6	0.03	0.39									
ΔVmax (m/s)												
HRV-G	6	-0.68	3.08	5.55	0.046	0.41	0.038	0.85	0.005	1.525	0.252	0.16
TRAD-G	6	-0.57	1.57									
ΔHRmax (bpm)												
HRV-G	6	0.17	26.69	5.646	0.045	0.414	7.58	0.025	0.487	0.024	0.88	0.003
TRAD-G	6	-1.67	7.9									
ΔHRrel (%)												
HRV-G	6	-0.3	14.81	7.18	0.028	0.473	8.47	0.02	0.514	0.031	0.865	0.004
TRAD-G	6	-0.87	4.17									
ΔVO₂max (ml/kg/min)												
HRV-G	6	0.67	7.58	0.338	0.577	0.04	0.586	0.466	0.068	0.734	0.417	0.084
TRAD-G	6	1	10.1									
ΔVO₂max (l/min)												
HRV-G	6	0.12	0.67	0.352	0.569	0.042	0.108	0.751	0.013	0.682	0.433	0.079
TRAD-G	6	-0.04	0.48									
ΔRER (l)												
HRV-G	6	-0.012	0.09	0.403	0.543	0.048	0.003	0.96	0.0003	0.195	0.67	0.024
TRAD-G	6	-0.04	0.07									
ΔVENT (bpm)												
HRV-G	6	9.52	12.68	0.655	0.442	0.076	2.397	0.16	0.231	0.54	0.483	0.063
TRAD-G	6	-1.95	15.64									

Note: LnrMSSD = the Neperian logarithm of the square root of the mean value of the sum of the squares of the differences between the adjacent RR intervals (ms). Treadmill test outcomes: Vmax = maximal velocity (km/h). HRmax = maximal heart rate (bpm). HRrel = the percentage of the heart rate relative to the maximum (%). VO₂max = maximal oxygen uptake (l/min) and relative to weight (ml/kg/min). RER = respiratory exchange ratio (l). VENT = ventilatory rate (bpm).

professional endurance runners, and describing the vagal modulation interaction. First of all, the total training volume and the time training in Zones 1, 2 and 3 were significantly higher in TRAD-G. Nevertheless, the proportion of training time at high intensity (Zone 3) was similar for both groups, but HRV-G spent significantly more time training at

moderate intensity (Zone 2), and TRAD-G trained more in Zone 1. This training structure did not generate differences between groups but some performance aspects significantly changed in both groups, such as the maximal velocity or the respiratory exchange ratio, favoring the HRV-guided training. Moreover, there was a non-significant but small-to-

high effect in the LnrMSSD increment and in the VO_{2max} , and a decrease in the ventilation rate in HRV-G. These changes in LnrMSSD had a significant impact on the maximal and relative heart rate, and were significantly correlated to the changes in VO_{2max} , meaning that an increase in cardiac vagal activation positively influences the endurance runners' cardiovascular performance.

Regarding the training structure, the HRV-G training was more efficient, resulting in less total training volume than in a traditional training methodology, as was reported by Vesterinen et al. [18]. In addition, higher proportions of moderate-to-high intensities (Zone 2 and Zone 2 + 3) were achieved by the HRV-G group while a higher proportion of low-intensity training (Zone 1) was achieved by the TRAD-G group. The distribution of volume and intensities in TRAD-G fitted with the traditional characteristics of elite endurance athletes' training [19]. On the other hand, the higher training intensities achieved by HRV-G show that their recovery was better than theoretically expected, as pointed out by Seiler et al. [7] and Hackney [20]. Thus, their LnrMSSD remained within the normal range more often, demonstrating the physiological adaptations associated to training, as can be seen in Figs. 1 and 2. In this way, designing the training based on the daily HRV scores allows professional runners to train at higher intensities and lower volume than when following a traditional and theoretical training method. If intensity is the key to optimizing aerobic training [1], HRV-guided training may provide a practical tool for better adapting the training prescription to professional endurance runners, increasing the importance of microcycle (day-to-day) training periodization.

Regarding the training time at the different intensities, similar training proportions in Zones 1, 2 and 3 were found in the study by Javaloyes et al. [21] on professional cyclists. On the other hand, our results do not agree with those of Javaloyes et al. [14] since they found a higher proportion of low and high-intensity training in professional cyclists, and Vesterinen et al. [11] where similar proportion of times in the different training zones between groups were found in the recreational runners. This indicates that the focus of the training design determines the intensity of the training sessions when moderate or high-intensity sessions can be chosen. In this regard, a simpler schema to design the HRV-guided training could help to unify the interventions in professional athletes. In accordance with Seiler et al. [7], it seems that the recovery is essentially identical following a moderate-intensity training session or a session over the VT 2 in highly trained endurance runners, while the athlete's level does affect the recovery. Therefore, including only high or low-intensity training options could be a more optimal procedure when the sample is composed of professional athletes.

Regardless of this, the training structure followed by HRV-G reported a significant increase in maximal velocity ($\Delta V_{max}=0.68 \pm 3.08$ km/h, $P = 0.027$, $d = 0.66$) and a small-to-high effect in the VO_{2max} ($\Delta VO_{2max}=0.67 \pm 7.58$ ml/kg/min, $P = 0.267$, $d = 0.32$), with a decrease in the ventilation rate ($\Delta VENT=9.52 \pm 12.68$ bpm, $P = 0.11$, $d = 1.01$). These outcomes fit with certain physiological effects expected when training in Zone 2 [22]. Similarly, the training structure followed by TRAD-G is reflected in the significant increment in the RER ($\Delta RER = 0.03 \pm 0.001$ l, $P = 0.017$, $d = 1$) and in the low effect associated to the change in the other physiological outcomes measured. This is in accordance with the physiological effects generated by training in Zone 1, as it is sufficient to build a solid aerobic base; however, when the objective is to improve endurance performance, higher training velocities are necessary to stimulate the neuromuscular system to maintain higher competition velocities [1]. Therefore, the higher proportion of moderate-intensity training in HRV-G resulted in better physiological performance after the intervention than the higher proportion of low-intensity training followed by TRAD-G. These results are in the line with other similar studies carried out on professional or amateur athletes [2,3,10,14,8,21,23,18,24,25]. However, when a block periodization was followed in these studies, significant changes were found in variables such as the VO_{2max} or peak power in their HRV-guided training groups, or the V_{max}

in their traditional training groups. Therefore, including a higher proportion of high-intensity training might improve the physiological results.

Continuing with the intervention effect, and in accordance with other studies [15,2,3], there were no significant inter or intra-group changes regarding the LnrMSSD. However, the moderate tendency for increased cardiac vagal activation found in the HRV-G at the end of this study was considered relevant as this could be related to this group's higher proportion of training in Zone 2. According to López-Chicharro et al. [1] or Hackney [20], endurance training between the ventilatory thresholds 1 and 2 is related to a reduction in sympathetic activation. This assumption was proven in several experimental studies, such as Iellamo et al. [26] and Nuutila et al. [10]. Additionally, it seems that training below VT 1 (Zone 1) had no effect on the autonomic nervous system balance in highly trained runners [7]. On the other hand, the autonomic cardiac vagal modulation is unclear when the training intensity is increased below the VT 2 [26,7,27,3,28,10]. Thus, a large proportion of training at moderate intensity (Zone 2) had a moderate effect on the professional endurance runners' cardiac vagal activation increment, which could be enhanced if the training volume (for example, the duration of the intervention) were increased. Nevertheless, the effects that the major total training volumes or higher proportions of training in Zone 3 have on the LnrMSSD should continue to be studied.

In this regard, the change in cardiac vagal activation found after the intervention ($\Delta LnrMSSD$) had a significant impact on our sample's maximal and relative heart rate change, which was significantly lower in HRV-G than in TRAD-G ($\Delta HR_{max HRVG}=0.17 \pm 26.69$ bpm, $\Delta HR_{max TRADG}=-1.67 \pm 7.9$ bpm, $F = 7.58$, $P = 0.025$, $d = 0.487$; $\Delta HR_{relHRVG}=-0.3 \pm 14.81$ bpm, $\Delta HR_{relTRADG}=-0.87 \pm 4.17$ bpm, $F = 8.47$, $P = 0.02$, $d = 0.514$). Moreover, the change in the $\Delta LnrMSSD_{cv}$ influenced the change in the ventilation rate, which decreased in HRV-G with a large effect size despite the absence of significant differences between groups. Furthermore, significant indirect correlations were found between $\Delta LnrMSSD$ and ΔVO_{2max} ($r = -0.606$, $P = 0.037$), between $\Delta LnrMSSD_{cv}$ and $\Delta VENT$ ($r = -0.790$, $P = 0.002$), and even between $LnrMSSD_{TEST}$ and posttest VO_{2max} ($r = -0.656$, $P = 0.02$). This association between LnrMSSD and the performance variables was also found in other studies carried out with professional swimmers [15] and recreational runners [11,29,10], but the performance-related measure was the maximal speed, which represents a submaximal intensity outcome. However, the relevance of our findings can be found in the performance measures that represent the runners' maximal capacity (maximal heart rate or VO_{2max}). As is shown by these results, the cardiac vagal activation was conditioning our runners' performance - the higher the change in LnrMSSD, the lower the change in maximal capacity outcomes. These results are in accordance with the assumption that increased cardiac vagal activation allows the training demands to be met with more stable physiological responses [1,22], which entails more optimal physiological responses to maximal effort. On the other hand, the baseline scores also had a significant impact on our sample's maximal and relative heart rate changes. According to the initial level training principle [30], the initial level of the HRV-G runners was somewhat better than that of the TRAD-G runners, even though there were no significant differences between groups. Therefore, the cardiac vagal activation impacts positively on the professional runners' performance, although the baseline scores should also be taken into account.

5. Conclusions

Using HRV as a daily guide to professional runners' training demonstrates that their recovery is better than expected, allowing them to achieve higher intensities at a lower training volume. However, polarized training, which includes only low or high training intensities, is recommended to strengthen cardiovascular performance and cardiac vagal modulation. Nevertheless, the increment in HRV scores suggest better cardiovascular adaptations, favoring HRV as a measure for

optimizing training individualization and cardiovascular performance.

6. Limitations and practical applications

A polarized training method, combining high and low intensities, and/or longer interventions (>8 weeks), should be considered in further research. The athletes' level (professional or amateur) should be taken into account to homogenize the performance results. However, using the HRV4Training device allows coaches and athletes to directly collect day-to-day recovery data without compromising the data's validity. HRV measurements can optimize the adjustments made to the training process planning, reducing the burden of repetitive performance testing and adapting the training individualization to maximize the professional athletes' potential. According to the previously reviewed research, HRV monitoring will also help coaches in deciding whether or not to increase the sessions' training intensity, thus reducing the risk of overtraining. Recording the training load and HRV can also assist coaches and athletes in controlling other factors that influence stress recovery rather than reducing the training itself.

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