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Stress and Training Load Effects on Recovery, Well-Being and Sports Performance

Edited by
Filipe Manuel Clemente, Juan Pedro Fuentes García and
Rodrigo Ramirez-Campillo

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Contents

Preface to ”Stress and Training Load Effects on Recovery, Well-Being and Sports Performance”

While training, the magnitude of the exercise and type of stimulus used impact the psychophysiological responses of athletes, which can ultimately influence the way in which several well-being parameters vary. This Special Issue aims to establish a relationship between training exposure and variations of recovery/ readiness processes and the well-being of athletes, providing new insights that may enhance the body of knowledge about this topic of research. This Special Issue will present studies connecting the training process with variation in psychological aspects and neuromuscular and physiological outcomes. Among the studies published, such effects on both athletes in recreational sports and clinical populations are reviewed. This diverse range of research presented will enrich this edition and help us take steps in the direction towards the advancement of this field.

Filipe Manuel Clemente, Juan Pedro Fuentes García, Rodrigo Ramirez-Campillo
Editors

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Article

Physical Activity and Perceived Physical Fitness during the COVID-19 Epidemic: A Population of 40- to 69-Year-Olds in Japan

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Abstract: The COVID-19 pandemic has caused an abrupt change in lifestyle for many people with restrictions, often leading to a decrease in physical activity (PA), and thus contributing to a negative perception of health status. The purpose of this study was to examine the effects of the COVID-19 epidemic on physical activity and perceived physical fitness in Japanese adults aged 40 to 69 years. Data were collected from an online survey conducted between October 19 and 28, 2020. The analytic sample consisted of 1989 Japanese adults (mean age, 50.1 ± 6.9 years; women, 38.9%) who were aged between 40 and 69 years and completed the online survey. Overall, the PA time per week decreased by 32.4% between October 2019 and April 2020. A decrease in PA time was recorded in October 2020; however, a decline of 15.5% was observed. Compared to individuals who did not perceive a decline in physical fitness, individuals who perceived declining physical fitness during the COVID-19 state of emergency demonstrated a greater decrease in PA time in April 2020 (–50.5%), and this trend continued into October 2020 (–25.0%). These findings may indicate that Japanese adults aged 40 to 69 years who perceived declining physical fitness experienced a greater decrease in physical activity.

Keywords: physical activity; midlife; COVID-19

1. Introduction

The COVID-19 epidemic has dramatically changed the daily lives of people worldwide. Lifestyle and health behavior including physical activity (PA) and dietary habits during the COVID-19 lockdown have been investigated [1,2]. The impact of the COVID-19 pandemic on dietary intake, PA, and sedentary behavior among different groups such as university students, elite para-athletes, community-dwelling older people, and patients with type 2 diabetes have been examined [1,3–5]. For instance, the COVID-19 state of emergency or lockdown has led to PA restrictions worldwide [6]. In Japan, a state of emergency was declared in seven prefectures, including Tokyo, on April 7, 2020 and extended to the entire country on April 16 [7]. On May 25, the state of emergency was lifted nationwide for the first time in about a month and a half. Although the declaration of a state of emergency is not legally binding, the prefectural governors of the target areas can request that residents refrain from going out as part of a collective effort to reduce infection, except when doing so is necessary for the maintenance of daily life. Among Japanese older adults aged ≥ 65 years, the PA time per week decreased by 65 min (–26.5%) from January to April 2020 [8]. If PA rapidly decreases for an extended period, it may have a negative impact on health status.

Lower midlife PA is associated with a higher risk of cardiovascular diseases [9], metabolic syndrome [10], and neurodegenerative diseases [11]. PA in midlife is also related to perceived health [12]. Perceived health status affects negative health-related outcomes, such as mortality, functional impairment, and dementia [13–15]. Lifestyle changes during the state of emergency may affect perceived health status, which negatively affects future health conditions. A previous longitudinal study investigated changes in PA prior to lockdown restrictions being imposed, and across three time periods: pre-, during, and post-lockdown [16]. The results showed that vigorous and moderate intensity PA were significantly lower during and post-lockdown compared to pre-lockdown in those individuals who had been highly active pre-lockdown [16]. More than 40% of community-dwelling Japanese old-old adults perceived a decline their physical fitness during the COVID-19 state of emergency [17]. Although several studies have reported decreased PA due to the COVID-19 epidemic [18], few have determined its impact on perceived health and PA recovery. Clear associations between decreased PA due to the COVID-19 epidemic and poor perceived physical health status may exist.

The purpose of this study was to examine the effect of the first wave of the COVID-19 state of emergency on physical activity and perceived physical fitness in Japanese adults aged 40 to 69 years. We examined the PA time across three time periods: pre- (October 2019), during (April 2020) and post- (October 2020) COVID-19 state of emergency and compared those between Japanese adults aged 40 to 69 years who perceived declining physical fitness during the COVID-19 state of emergency and those who did not.

2. Materials and Methods

2.1. Study Sample

Data for this study were collected from an online survey panel administered through the sampling of Y cloud systems among Japanese adults. The Y cloud system is a crowd-sourcing service launched by Yahoo Japan Corporation, Inc. (Tokyo, Japan) in 2013. From 19 to 28 October 2020, 3048 Japanese adults completed the online survey. The inclusion criterion was adults aged 40 to 69 years. Responders who reported a history of stroke, Parkinson's disease, dementia, depression, and/or neurological disorders, or who gave the wrong answer to a question (choosing a specific option from multiple choices) to identify fraudulent responses were excluded. Responders who reported more than 960 min/day or 0 min/day of total PA time and more than a tenfold change (increasing or decreasing) of PA were also excluded. Finally, data from 1986 Japanese adults aged 40 to 69 years were analyzed. This study was conducted in accordance with the guidelines proposed by the Declaration of Helsinki, and the study protocol was reviewed and approved by the Ethics Committee of the Faculty of Medicine, Kagoshima University (#200101).

2.2. Assessment of PA

The abbreviated version of the International Physical Activity Questionnaire (IPAQ) [19] which consists of three-dimensional activity items—activity intensity level (light, moderate, and vigorous intensity), activity frequency per week, and activity time per day—was used to assess PA. Participants were asked to report their PA during three specific time periods: (1) October 2019, before the COVID-19 epidemic; (2) April 2020, during the first wave of the COVID-19 state of emergency; and (3) October 2020, after the COVID-19 state of emergency. Thus, they were asked to recall October 2019 and April 2020, as well as to report their current situation in October 2020. Following the guidelines for data processing and analysis of the IPAQ, only values of 10 or more minutes of PA were included in the calculation of summary scores. Responses of less than 10 min (and their associated days) were re-coded to 'zero'.

Additionally, activity time variables of each level exceeding '3 h' or '180 min' were truncated to be equal to '180 min' in a new variable. We determined the PA time (minutes/week) as added values for each activity level, which were then multiplied by activity frequency per week and activity time per day (minutes) at each activity level [8].

2.3. Assessment of Perceived Declining Physical Fitness

We investigated the participants' perceived decline in physical fitness during the COVID-19 state of emergency. Participants were asked to answer "yes" or "no" to the following question: "Do you perceive a decline in physical fitness after the state of emergency?" Participants who answered "yes" to this question were classified as perceiving a decline in their physical fitness. Incidentally, in this survey, respondents were asked about their physical fitness self-perception during the COVID-19 epidemic prior to completing the IPAQ.

2.4. Demographic Variables

Several variables such as age, gender, education, living alone, and living area were collected in the current study. The living area was classified as a special precaution area due to the COVID-19 pandemic. In Japan, 13 prefectures, including Tokyo, Kanagawa, Saitama, Chiba, Osaka, Hyogo, Fukuoka, Hokkaido, Ibaraki, Ishikawa, Gifu, Aichi, and Kyoto, were identified as areas of special precaution based on the situation in April 2020.

2.5. Statistical Analysis

PA time is presented as a median, with an interquartile range (IQR). The IPAQ guidelines state that physical activity is non-normally distributed in many populations and suggest reporting medians [20]. Therefore, we conducted the analysis using non-parametric tests, as appropriate. Changes in PA time were tested using Friedman's test for three time points, in October 2019, April 2020, and October 2020, overall and for each group, divided by perceived declining or non-declining physical fitness. Changes in PA time were also compared with October 2019 (before the COVID-19 epidemic) and tested using the Wilcoxon rank-sum test overall and for each group, divided by perceived declining or non-declining physical fitness. The changes in PA time also compared with October 2019 (before the COVID-19 epidemic) were tested using the Wilcoxon rank-sum test in overall and each group divided by perceived declining or non-declining physical fitness. Comparisons of demographic variables and PA time between perceived and non-perceived decline in physical fitness were tested using the paired t-test (for age), Mann–Whitney U test (for PA time), or chi-square test (for proportion). All analyses were conducted using IBM SPSS Statistics 26.0 (IBM Japan Tokyo, Japan). The level of statistical significance was set at $p < 0.05$.

3. Results

The demographic variables and PA time in October 2019, April 2020, and October 2020 for each age group (40–49, 50–59, and 60–69 years) are presented in Table 1. The mean (± standard deviation) age of the participants was 50.1 ± 6.9 years and 38.9% were women. Overall, the PA time decreased ($p < 0.001$) in April 2020 (median [IQR], 240 [80–540]) and October 2020 (300 [120–600]) compared to October 2019 (355 [150–660]).

Table 1. Demographic characteristics and physical activity (PA) time of the participants in each age groups.

	Overall (*n* = 1986)	**40–49 Years (*n* = 1046)**	**50–59 Years (*n* = 712)**	**60–69 Years (*n* = 228)**
Age, years	50.1 ± 6.9	44.8 ± 2.8	55.7 ± 2.8	63.4 ± 2.9
Women, *n* (%)	773 (38.9%)	453 (43.3%)	256 (36.0%)	64 (28.1%)
Education, *n* (%)				
Master/doctorate degree	110 (5.5%)	55 (5.3%)	38 (5.3%)	17 (7.5%)
Bachelor's degree	1011 (50.9%)	535 (51.1%)	345 (48.5%)	131 (57.5%)
Professional degree	311 (15.7%)	180 (17.2%)	105 (14.7%)	26 (11.4%)
High school graduate	465 (23.4%)	232 (22.2%)	185 (26.0%)	48 (21.1%)
Others	89 (4.5%)	44 (4.2%)	29 (4.1%)	6 (2.6%)
Living alone, *n* (%)	368 (18.5%)	201 (19.2%)	129 (18.1%)	38 (16.7%)
Living area, *n* (%)				
Special precaution areas due to COVID-19 pandemic	1415 (71.2%)	731 (69.9%)	532 (74.7%)	152 (66.7%)
Perceived declining physical fitness (yes), *n* (%)	671 (33.8%)	364 (34.8%)	227 (31.9%)	80 (35.1%)
PA time, minutes				
October-2019, median (IQR)	355 (150–660)	343 (150–700)	335 (150–630)	360 (161–685)
April-2020, median (IQR)	240 (80–540)	240 (60–560)	233 (80–490)	300 (120–600)
October-2020, median (IQR)	300 (120–600)	300 (100–600)	298 (120–600)	300 (120–630)

Abbreviations: PA, physical activity; IQR., interquartile range.

Table 2 illustrates the comparisons between participants who perceived a decline in physical fitness and those who did not in terms of demographic variables and PA time. Of the overall participants, 671 (33.8%) perceived a decline in physical fitness during the COVID-19 state of emergency. There were no significant differences between those who perceived a decline in physical fitness and those who did not in terms of education and who they lived with, but there was a higher rate of women and those who lived in special precaution areas among participants who perceived a decline in their physical fitness. Although there was no significant difference in PA time in October 2019 between those who had perceived a decline in physical fitness and those who had not, there was a significant difference in PA time between the former (180 [60–480]) and the latter (270 [100–600]) in April 2020 ($p < 0.01$). In October 2020, this difference was not evident, but there was a slight decrease in the group with a perceived decline in physical fitness (270 [90–580]) compared with the group with no perceived decline in physical fitness (300 [120–620]) ($p = 0.48$).

Table 2. Comparison between participants with and without a perceived decline in physical fitness.

	No Perceived Decline in Physical Fitness (*n* = 1315)	**Perceived Decline in Physical Fitness (*n* = 671)**	***p***
Age, years	50.2 ± 6.9	50.0 ± 6.9	0.57
Women, *n* (%)	469 (35.7%)	304 (45.3%)	<0.01
Education (Bachelor/master/doctorate degree), *n* (%)	587 (44.6%)	278 (41.4%)	0.17
Living alone, *n* (%)	237 (18.0%)	131 (19.5%)	0.42
Living area, *n* (%)			
Special precaution areas due to COVID-19 pandemic	913 (69.4%)	502 (74.8%)	0.01
PA time, minutes			
October 2019, median (IQR)	340 (140–660)	360 (150–700)	0.43
April 2020, median (IQR)	270 (100–600)	180 (60–480)	<0.01
October 2020, median (IQR)	300 (120–620)	270 (90–580)	0.48

Abbreviations: PA, physical activity; IQR., interquartile range.

Figure 1 shows the changes among the three time points, in October 2019, April 2020, and October 2020. Overall, there were significant differences in PA time among the three time points ($p < 0.001$). A significant 32.4% decline in PA time in April 2020 ($p < 0.01$) was observed, compared with October 2019. The decreased PA time improved in October 2020; however, the median PA time still decreased (–15.5%), compared with October 2019 ($p < 0.01$). PA time change results for the perceived non-declining and declining physical fitness groups are also illustrated in Figure 1. Participants who perceived declining physical fitness during the COVID-19 state of emergency showed more than a 50% decrease in PA time in April 2020 (–50.5%) ($p < 0.01$), and that decline remained (to a lesser degree) in October 2020 (–25.0%) ($p < 0.01$), compared with October 2019. Conversely, in participants who did not perceive declining physical fitness during the COVID-19 state of emergency, PA time during the COVID-19 state of emergency (April 2020) (–20.6%) and afterward (October 2020) (–11.8%) showed a smaller decline; however, those decreased PA times were still at a significant level ($p < 0.01$).

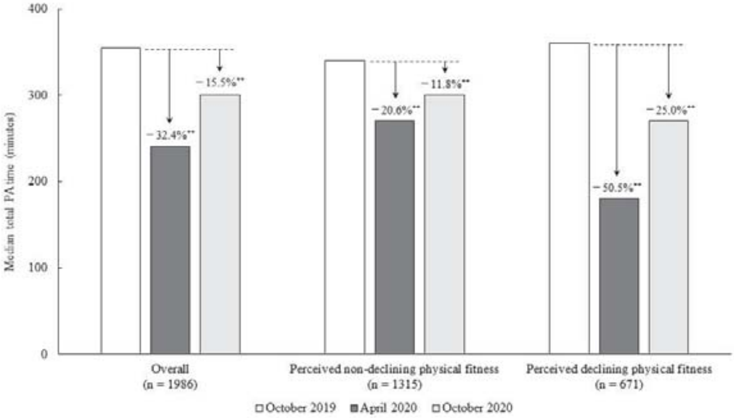

Figure 1. Changes in PA time in April and October 2020 compared with October 2019 in participants with and without a perceived decline in physical fitness. **: statistical significance ($p < 0.01$) compared with October 2019.

4. Discussion

This study examined PA time across three time periods: pre- (October 2019), during (April 2020) and post- (October 2020) COVID-19 state of emergency and compared PA times in those three time periods between Japanese adults aged 40 to 69 years who perceived a decline in physical fitness and those who did not. The current study confirmed that weekly PA time decreased from October 2019 to April 2020 by approximately 30%. This decrease in PA time improved in October 2020; however, a decline of approximately 15% was still observed. Specifically, the decrease in PA time in participants who perceived declining physical fitness during the COVID-19 state of emergency was remarkable, with declines of approximately 50% and 25% in April and October 2020, respectively. Those results would indicate secondary effects of the COVID-19 epidemic on our health, and continuously decreasing PA may contribute to negative health outcomes.

A previous study indicated that step counts decreased in the period after COVID-19, and differences were observed between regions, likely reflecting regional variation in COVID-19 timing, regional enforcement, and behavior change [6]. Among Japanese community-dwelling older adults, although the total amount of time devoted to PA in April 2020 (during the first wave of the COVID-19 pandemic) had significantly decreased from that in January 2020 (before the COVID-19 pandemic), PA time in June 2020 had recovered to the same level as before the COVID-19 pandemic (January 2020) [21]. The current study revealed that approximately 30% of the PA time decreased during the first wave of the COVID-19 pandemic among Japanese adults aged 40 to 69 years. The decrease in PA time in October 2020 was still approximately 15% lower than that before the COVID-19 pandemic (October 2019). Further long-term follow-up observation and examination of the effects of PA time changes on various health outcomes are needed.

Several previous studies examined that PA in midlife using the IPAQ to determine the prevalence of physical inactivity [22], and to examine the association of PA with neighborhood walkability [23] and the risk of health problems such as stroke [24], depression [25], and cognitive impairment [26]. Nearly all studies indicated that a lower PA in midlife had a negative impact on current and future health. During the COVID-19 lockdown, an online survey of a young cohort assessing its effect on PA levels showed an increase in sedentary behavior [27]. On the other hand, middle-aged adults with high and moderate PA levels had significantly higher life satisfaction and happiness than those with low PA levels [28]. Thus, to avoid negative impacts on health, unfavorable changes in PA time due to the COVID-19 state of emergency must reverse and recover to the level before the COVID-19 pandemic.

In this study, 33.8% (mean age 50.1 years) of participants perceived a decline in their physical fitness during the COVID-19 state of emergency. In general, self-rated health tends to deteriorate in older adults compared to middle-aged adults [29]. Poor self-rated health and physical fitness among middle-aged adults are associated with future negative health outcomes [30–33] and PA levels affect self-rated health status [34–36]. The current study identified an association between PA time and poor self-rated physical fitness during the COVID-19 state of emergency among Japanese adults aged 40 to 69 years, and those with perceived poor self-rated physical fitness showed less recovery of their PA after the first wave of the COVID-19 epidemic. In addition, a short-term reduction in steps resulted in a significant loss of leg fat-free mass [37]. Short-term physical inactivity also requires attention, and early improvement in PA is important [37]. Thus, the maintenance or minimization of changes in PA time due to the COVID-19 outbreak is very important, and if the PA decreases, it should be recovered as soon as possible.

One strength of this study is that it collected a representative sample of Japanese adults aged 40 to 69 years in the national area. Given the importance of PA and how affected it was by the restrictions imposed by the COVID-19 state of emergency, the results of this study will be important for planning and developing appropriate strategies and policies in the face of a pandemic. In addition to its strengths, some limitations should be considered when interpreting the results of the current study. Frist, participants were asked to recall their PA one year before the survey, in October 2019. Thus, it is possible that their subjective PA times could have been either underestimated or overestimated. Second, changes in PA time were assessed using the IPAQ and analyzed using non-parametric tests, following the IPAQ guidelines. We could not control for confounders, such as age, gender, and income. The decline in physical fitness is self-rated and not an objective measurement of physical fitness. Subjective PA levels may be over- or underestimated [38]. Furthermore, confounding factors between self-rated physical weakness and PA were not considered. In addition, the two groups were determined according to their perception of a decline in physical fitness at the end of the survey period, October 2020; as they were not divided according to objective measures, several April 2020 results might be different. For instance, some members of the "non-declining" group may have identified as members of the "declining" group if they had been asked in April 2020. For instance, some individuals of the "non-declining" group, if they were asked in April 2020, would answer as a "declining" group member. Therefore, some individuals may believe that there is a self-perception threshold regarding one's physical fitness status, despite any given individual's knowledge that they were, indeed, performing less PA. In future research, these points could contribute to understanding the associations between PA levels and self-perceived fitness. Third, participants were recruited from specific Internet service registrants.

5. Conclusions

The PA time per week for Japanese adults aged 40 to 69 years decreased by 32.4% between October 2019 and April 2020 with a continued 15.5% median decline in October 2020. The decrease in PA time in participants who perceived a decline in their physical fitness during the COVID-19 state of emergency was remarkable in April 2020 and October 2020. These findings indicate that Japanese adults who perceived a decline in their physical fitness experienced a greater decrease in PA and found it difficult to return to the same level of PA as the past year.

Author Contributions: H.M. conceived and designed the study, performed the analyses and drafted the manuscript. S.A., S.S., R.S. and D.S., prepared the data. S.A., S.S., R.S., Y.T., D.S. and Y.N. revised the manuscript. All authors participated in the interpretation of the results. 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.

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Article

Response of Knee Extensor Muscle-Tendon Unit Stiffness to Unaccustomed and Repeated High-Volume Eccentric Exercise

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Abstract: The purposes of this study were to investigate the muscle-tendon unit stiffness response and to compare the stiffness with those of other indirect markers induced by two bouts of unaccustomed eccentric exercise. Eleven untrained men performed two bouts of 200 maximal eccentric contractions of the right quadriceps 4 weeks apart. Changes in stiffness, pain evoked by stretching and pressure, plasma creatine kinase (CK) activity, and muscle thickness were followed for 7 days after each bout. Stiffness and pain peaked immediately and 1 day after the first exercise bout, whereas CK and thickness were highest 4 and 7 days after the first exercise bout, respectively ($p < 0.05$ for all). Muscular pain, thickness, and stiffness responses were lower by 53.3%, 99%, and 11.6%, respectively, after the repeated bout compared to after the first bout ($p < 0.05$ for all), while CK activity response did not differ significantly between bouts. High responders for an increase in muscle-tendon unit stiffness showed a repeated-bout effect for stiffness, pain, and CK activity (by 29%, 65%, and 98%, $p < 0.05$ for all), but the repeated-bout effect was not that clear in low responders. These findings suggest that a repeated eccentric exercise bout effect on stiffness in quadriceps is mostly not associated with muscle pain and CK activity, but there are large individual differences.

Keywords: muscle pain; stiffness; eccentric exercise; repeated-bout effect; knee extensor muscle

1. Introduction

Muscle stiffening and delayed-onset muscle soreness (DOMS) are typical sensations experienced soon after unaccustomed eccentric exercise [1]. Although the mechanisms and origin of the specific triggers of muscular stiffness are not clearly understood, tissue swelling is thought to be one direct cause [2]. The asynchronous development of swelling and stiffness suggests that they may not be causally linked [3]. Our recent study showing that postexercise stiffness develops much earlier than swelling [4] provided evidence that questions the early claim that these two processes are interrelated and instead suggested that they are likely to be two aspects of the same phenomenon. There are other possible causes, such as increased intracellular calcium concentration and contracture of damaged portions of muscle fibers [2,3]. In our earlier study of the interrelationships between muscle-tendon unit (MTU) stiffness, muscle pain and tenderness, swelling, and plasma CK release after acute eccentric exercise of the elbow flexors, we found strong correlations between muscle pain in response to stretching and increased resting elbow angle, and between peak plasma CK activity and the extent of swelling [4].

It is of practical importance that both muscle stiffness [4,5] and pain [4–6] responses are reduced when the exercise is repeated after the recovery from the previous bout. This phenomenon is called the repeated-bout effect (RBE) and is documented in both humans and animal models [7]. Several studies have demonstrated the protective effects of a previous exercise session on arm stiffness [5], leg stiffness [8], arm pain [5], leg pain [6], and muscle CK release [9] following the repeated bout. However, study results relating to whether there is an RBE on EIMD markers and MTU stiffness are inconsistent. For instance, stiffness of the plantar flexor MTU was not lower after a repeated exercise bout compared with the first bout [6]. By contrast, Janecki et al. [10] found less elbow flexor MTU stiffness following a repeated bout of 30 eccentric contractions. These differences may reflect differences in the EIMD induced by the exercise protocols, which used different combinations of exercise parameters such as volume and intensity. It may also be relevant that muscles of the upper limbs are much more susceptible to EIMD than are lower-limb muscles [11].

In the current study, we aimed to examine the dynamics of active stretch-induced muscle pain, pressure-evoked pain, swelling, CK release, and passive MTU stiffness after the first and repeated bout of eccentric exercise of the knee extensors. As a proxy of stiffness, the resistance of the passively stretched MTU was measured on an isokinetic dynamometer as has been used by others [12]. To compare indirectly the different aspects and time course of the responses to acute and repeated damage-evoking exercise between different muscle groups (lower vs. upper body), we used a protocol for high-volume eccentric lower-limb exercise to induce muscle damage of a magnitude comparable with that used to induce damage to the arm muscles [4]. We hypothesized that the reduced response of leg muscle stiffness after the second session because of the RBE would be related to the reduction in pain, which would confirm associations recently reported for arm exercise [4].

2. Materials and Methods

2.1. Participants

Eleven recreationally active men (mean ± standard deviation (SD); age, 21.3 ± 2.0 years; body weight, 81.5 ± 14.3 kg; height, 180.2 ± 3.5 cm) volunteered for the study. In order to assure that young, healthy, and non-familiar with eccentric types of activities, participants were recruited before being enrolled in the study who were asked about their age, health status, and physical activity prior to the study. Participants were included if they were 18 to 25 years, healthy, and not engaged in any type of resistance exercise during the previous 6 months. The exclusion criteria were the inability to abstain from strenuous physical activity during the entire study duration, any neuromuscular or skeletal problem, and the use of analgesic or anti-inflammatory drugs. The required minimum sample size was calculated before the study based on the expected passive knee extensor stiffness difference of 10% between exercise bouts, with an alpha level of 0.05 and the desired power of 80%. The calculation provided a sample size of at least ten participants.

Before participating in the study, the volunteers provided their written informed consent. The study conformed to the standards set by the latest revision of the Declaration of Helsinki and was approved by the Kaunas Regional Biomedical Research Ethics Committee (registration no. BE-2-17).

2.2. Study Design

The participants were familiarized with the equipment and measurements on the separate visit to the laboratory 3–4 days before the first exercise bout intervention. During the testing session, the following variables were measured before, immediately after, and 1, 2, 4, and 7 days after the exercise intervention: stretch- and pressure-induced muscle pain, MTU passive stiffness, swelling of vastus lateralis (VL) muscle as a change of muscle thickness, and plasma CK activity. The exercise intervention involved 200 maximal-effort eccentric isokinetic contractions of the knee extensors of the right leg performed twice, with a 4-week break between bouts. All exercise and testing was carried out by the same team of investigators. The temperature in the laboratory was kept stable between 20 and 22 °C.

2.3. The Intervention Exercise for Knee Extensors

The participant was seated on an isokinetic dynamometer (Biodex System 3 Biodex Medical Systems, Inc., Shirley, NY, USA) with the backrest fixed at 90° and with the trunk, pelvis, and right thigh stabilized to the chair of the dynamometer with Velcro straps. The axis of rotation was fixed at the knee joint, and the lever arm pad was attached 2–3 cm above the ankle. The participant was instructed to perform 20 sets (with 1 min breaks for recovery between sets) of 10 maximal voluntary eccentric contractions of the knee extensors in the range of 30–110° of knee flexion (0° = full knee extension) with the angular velocity set at $90^\circ \cdot s^{-1}$. The participant was instructed to exert a maximal-effort contraction of the knee extensors (resisted knee flexion) through the full range of motion (ROM). Between each contraction, the leg was fully relaxed for about 1 s to allow the machine to return the leg to the starting position at 30° knee flexion. Verbal encouragement to maintain maximal effort was given for each contraction. Torque was monitored throughout the 20 sets of exercises for the two exercise bouts.

2.4. Muscle Pain and Tenderness

Pain elicited by taking the knee to full flexion at 125° (stretched pain) during the endpoint of the passive stiffness test was evaluated using a visual analog scale of 0–10, where 0 represented "no pain" and 10 "intolerably intense pain" [13]. The pressure pain threshold (PPT) used to evaluate tenderness of knee extensors was measured with an algometer (1 cm diameter probe, Wagner Instruments, Greenwich, CT, USA) with the participant lying on the examination table with the knee extended. The probe was applied above the patella at a position 50% to a line between the anterior superior iliac spine and the superior border of the patella, laterally and centrally, for the VL and rectus femoris (RF), respectively. The probe was applied 10% to the reference line medially for the vastus medialis (VM). During the measurement, the investigator applied the tip of the device perpendicular to the skin and slowly increased pressure to 30 N. The participant reported the endpoint when the pressure first became a painful sensation. The applied force at the endpoint of the two trials was averaged and subtracted from 30 [4] and is referred to here as "PPT pain". The three measurement sites were firstly marked with a permanent marker with the guidance of real-time B-mode ultrasound, with the largest part of each muscle visible for repeated testing.

2.5. Passive Resistive Torque and MTU Stiffness of the Knee Extensors

To measure passive resistive torque, the participant was positioned lying on his back in a padded chair of the isokinetic dynamometer without shoes and with the right leg straight and parallel to the floor. The left leg was fully flexed at the hip and knee joints and held by the therapist to limit lumbopelvic motion (adapted from the work of Krause et al. [14]). The pelvis and the right thigh were fastened to the chair with Velcro straps. The axis of rotation of the right knee was fixed to the fulcrum of the dynamometer shaft with the knee out of the chair approximately by the distance of the 3 finger bases to ensure sufficient space for knee flexion to 125° without any lever arm strain, and the lever arm pad was fastened proximally to the malleolus. All dynamometer chair settings were noted for reproducibility. In this configuration and with the participant fully relaxed, the dynamometer passively flexed the knee to 125° at $5^\circ \cdot s^{-1}$. Passive stiffness was subsequently determined from the slope of the curve derived for the passive torque to angle relationship (with gravity correction for the weight of the leg). A third-degree polynomial using the least-squares method (R^2 value was used to evaluate the fit) was fitted to the linear portion of the curve for 85–100% of the maximal knee flexion. Measurements were accepted only if the knee muscle activity of the extensors (RF and VL) and flexors (biceps femoris muscle) was both <5% of the maximal electromyographic values [15], which had been predetermined by having the participant perform a maximum voluntary isometric contraction to obtain the peak of the root mean square. Three passive maximum peak torque and stiffness values were averaged for further analysis. The intraclass correlation coefficient for stiffness was 0.974 (95% confidence interval (CI): 0.904–0.993) in this study.

2.6. VL Muscle Thickness

Between the examinations of muscle pain and PPT pain, transverse images of the VL mid-belly were obtained using B-mode ultrasonography with a 10–15 MHz transducer (Echoblaster 128, UAB; Telemed, Vilnius, Lithuania) with minimal probe pressure application. The clearest images of the fascia captured were analyzed, and the probe placement was outlined with a permanent marker at the mid-belly in the same point for PPT of VL measurement. VL muscle thickness was then measured between the superficial and deep aponeurosis [16] using ImageJ image analysis software (Wayne Rasband, NIH, Bethesda, MD, USA). In this study, the intraclass correlation coefficient for muscle thickness was 0.895 (95% CI: 0.845–0.923). The average thickness from two VL images was used in further analyses.

2.7. Plasma CK Activity

About 0.25 mL of capillary blood was drawn from the finger and immediately centrifuged, and the plasma was used for measurement of CK activity using a bench-top biochemical analyzer [4] (Spotchem^{TM} EZ SP-4430, Menarini Diagnostics, Winnersh-Wokingham, UK) using soft reagent strips (Arkray Factory, Inc., Shiga, Japan).

2.8. Statistical Analysis

Descriptive data are presented as mean and SD; the standard error of the mean (SEM) is given where means are compared. The Shapiro–Wilk test was used to check whether the data were normally distributed. When not distributed normally, the data are reported as the median and interquartile range (IQR). Peak torque and work performed during the eccentric exercise were compared using paired *t*-tests. A repeated-measures analysis of variance was used to compare the change in MTU stiffness using the following parameters: condition (first or repeated bout) × time (before, immediately after, and 1, 2, 4, and 7 days after exercise). When significant main effects were found, post hoc testing was performed using paired *t*-tests with the Bonferroni correction for multiple comparisons. Partial eta-squared was used as a measure of the effect sizes (ES) in repeated measurements statistics. The data that failed the test for normal distribution were analyzed using the nonparametric Wilcoxon's signed-rank and Mann–Whitney *U* tests.

The relationships between the two measures of the change in stiffness, pain, plasma CK activity, and muscle thickness were identified using linear regression. The correlation coefficient (r) was used to identify the explained variance between tests. Significance was accepted as $p < 0.05$. As in our previous study on upper body exercise [17], the response to eccentric knee extension exercise was analyzed both on a whole group level as well as to more comprehensively represent the variability of the individual response, mainly for stiffness in this study, separately in low ($n = 4$) and high responders ($n = 4$) for MTU stiffness response to the first exercise bout. The test-retest reliability of the selected variables was found using the absolute agreement intraclass correlation coefficient (ICC 2,1) with a 95% confidence interval (CI) between the familiarization session and the baseline before first bout scores. Statistical analysis was performed using IBM SPSS Statistics for Windows (version 23.0, IBM Corp., Armonk, NY, USA).

3. Results

3.1. Eccentric Exercise

All participants successfully completed the two exercise sessions of 200 eccentric contractions. Peak torque declined progressively across the sets of 10 contractions to 55.1% ± 14.2% of the value in the first set of the first bout and to 69.0% ± 13.4% during the repeated bout (both $p < 0.001$) and did not differ significantly between bouts ($p > 0.05$). Similar dynamics were evident in the change in work from the first to the last set, which decreased to 62.4% ± 14.5% during the first bout and to 71.1% ± 14.2% during the repeated bout (both $p < 0.001$) with no difference between bouts ($p > 0.05$).

3.2. Repeated-Bout Effect

The differences in the responses of markers of EIMD after the first and repeated bouts are shown in Figure 1. Plasma CK activity peaked around day 4 being median 18.1 µkat (IQR: 13.7–216.5 µkat, $p = 0.017$) and was elevated for 7 days after the first bout, whereas it was elevated for only 1–2 days (11.3 µkat, IQR: 9.97–13.16 µkat, $p < 0.05$) after the repeated bout (Figure 1A). In 7 of 11 participants, CK activity was 5–200 times lower after the repeated bout. In the other four participants, a low CK activity response was evident after the first bout, which left little room for a decrease by several folds after the repeated bout. The difference between bouts did not reach significance because of this high variability in the CK response.

The stretched pain in the damaged muscle was maximal 1 day after the exercise. The median pain ratings were 6 points (IQR: 2.31–7.53 points) after the first bout and 4 points (IQR: 2.80–4.81 points) after the repeated bout ($p = 0.029$). Stretched pain in the damaged muscle remained evident during the 7 days after both exercise bouts. The RBE on pain development was seen on days 2–7 after the repeated exercise ($p < 0.01$ for all) (Figure 1B).

VL muscle thickness increased progressively in response to exercise and reached a peak by day 7 after the first exercise bout (F(1,9) = 12.1, $p = 0.007$, ES = 0.57). However, no increase was evident after the repeated exercise bout ($p > 0.05$) (Figure 1C).

Exercise-induced muscle tenderness, as measured by increased PPT pain in the RF, VM, and VL muscles (Figure 1D–F), increased over days 1 to 4, peaked on day 2 ($p < 0.05$ for all muscles) after the first bout, and had recovered fully by day 7. The repeated bout induced tenderness only in the VM muscle 1 day after exercise ($p = 0.028$) (Figure 1E). The maximum pressure pain observed after the first bout of exercise did not differ between the three muscles ($p > 0.05$).

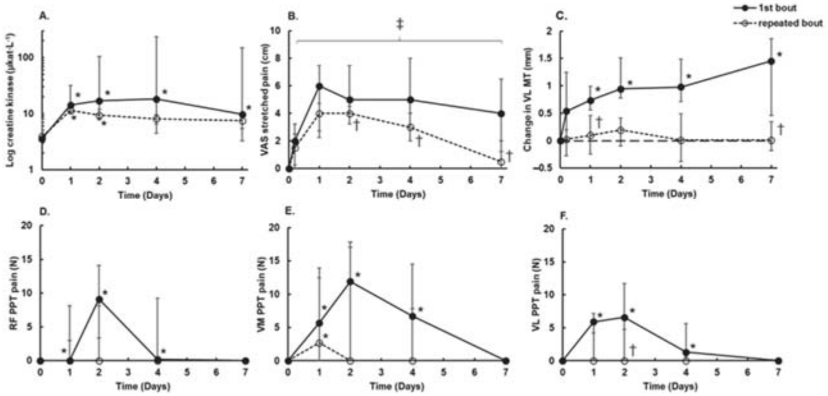

Figure 1. Responses to a repeated bout of 200 knee extension eccentric contractions. (**A**) Plasma creatine kinase (CK) activity expressed on a log scale. (**B**) Visual analog scale (VAS) to rate stretched muscle pain. (**C**) Thickness of vastus lateralis (VL). (**D**) Pressure pain threshold (PPT) in rectus femoris (RF). (**E**) PPT pain in vastus medialis (VM). (**F**) PPT pain in VL. Data are presented as the median and interquartile range (IQR) ($n = 11$). Filled circles and solid lines are data for the first bout; open circles and dashed lines are data for the repeated bout. * Significant difference from before exercise ($p < 0.05$). ‡ Significant difference from before exercise for both exercise bouts ($p < 0.05$). † Significant difference from the first exercise bout ($p < 0.05$).

Data for MTU stiffness of the knee extensors are shown in Figure 2. The slope of the passive torque at the last 15° of knee flexion responded differently between the two exercise bouts (Figure 2A,B). MTU stiffness reached a peak at 17.8% (CI: 6.9–28.6%) above the baseline value immediately after the first bout ($F(5,40) = 5.8$, $p < 0.001$, ES = 0.42) and at 6.1% (CI: 1.3–13.6%) above the baseline immediately after the repeated exercise bout ($p > 0.05$, Figure 2C). MTU stiffness remained higher on day 1 after the first exercise bout but not on day 1 after the repeated bout (bout effect for pre-, post- and 1 day postexercise values, $F(1,8) = 6.3$, $p = 0.036$, ES = 0.44). On days 2–7, MTU stiffness did not differ from the baseline value or between exercise bouts (Figure 2C). Peak resistive torque at the endpoint of passive knee flexion was lower after the repeated compared to first bout ($F(1,8) = 6.2$, $p = 0.038$, ES = 0.44) (Figure 2B).

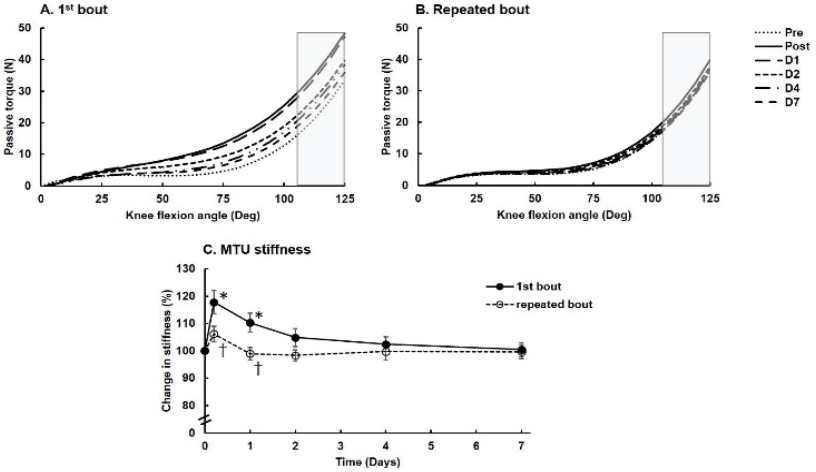

Figure 2. Changes in passive MTU stiffness of the knee extensors. Angle–torque relationships for (**A**) the first bout and (**B**) the repeated bout. The gray shaded boxes indicate the stiffness calculation for the last 15° (110–125°) of passive knee flexion. (**C**) Passive MTU stiffness after 7 days is shown as mean and SEM ($n = 11$). Filled circles and solid lines are data for the first bout; open circles and dashed lines are data for the repeated bout. * Significantly different from the pre-exercise value ($p < 0.05$). † Significantly different from the first exercise bout ($p < 0.05$).

3.3. Correlations between MTU Stiffness and Markers of EIMD

Baseline MTU stiffness did not correlate with changes in any of the markers of EIMD ($p > 0.05$) (Table 1). There was a significant, but modest correlation between peak stiffness and peak muscle swelling ($r = 0.50$, $p = 0.04$) but not between peak stiffness and peak pain ($r = 0.18$, $p > 0.05$) or peak plasma CK activity ($r = 0.33$, $p > 0.05$) (Table 1).

Table 1. Correlations between stiffness and peak pain in response to stretching with peak pain to pressure, swelling, and plasma CK activity ($n = 11$).

	Initial Stiffness		Peak Stiffness		Peak Pain on Stretch	
	r	*p*	r	*p*	r	*p*
Peak pain on stretch	−0.18 (−0.06,0.03)	0.47	0.18 (−1.91,3.83)	0.49	-	-
Peak RF PPT	−0.29 (−0.02,0.01)	0.25	0.42 (0.34,1.68)	0.09	0.30 (−0.06,0.24)	0.22
Peak VM PPT	−0.20 (−0.01,0.01)	0.43	0.35 (−0.22,1.23)	0.16	0.59 (0.05,0.27)	0.01
Peak VL PPT	−0.06 (−0.02,0.02)	0.82	0.35 (−0.38,2.11)	0.16	0.43 (−0.02,0.41)	0.08
Peak stiffness	−0.21 (−0.10,0.01)	0.41	-	-	0.18 (−0.06,0.13)	0.49
Peak Swelling	0.01 (−0.07,0.07)	0.98	0.50 (0.39,9.89)	0.04	0.18 (−0.64,1.32)	0.48
Peak CK	−0.05 (−0.01,0.01)	0.86	0.33 (0.08,0.44)	0.39	0.28 (−0.03,0.08)	0.28

RF, rectus femoris; PPT, pressure pain threshold; VM, vastus medialis; VL, vastus lateralis; CK, creatine kinase.

3.4. High Responders vs. Low Responders

The stiffness, VAS score, and plasma CK activity of the four participants with the highest MTU stiffness (30.3%, CI: 13.1–47.6%, $p = 0.011$) and four participants with the lowest MTU stiffness (5.9%, CI: 0.5–11.3%, $p = 0.04$) immediately after the first exercise bout are presented in Figure 3. MTU stiffness did not change after the repeated bout in any group ($p > 0.05$) (Figure 3A). Stretched pain in the exercised muscle increased over days 1–7 after the first bout ($p < 0.05$) but was much lower ($p < 0.05$) on days 4–7 after the repeated bout for the high responders (Figure 3B). The pain did not differ between the bouts in low responders ($p > 0.05$). Plasma CK activity in the high responders tended to be higher and last longer over the 7 days compared with that in the low responders, for whom it increased nonsignificantly only 2 days after the first exercise bout ($p > 0.05$) (Figure 3C). Plasma CK activity after the repeated bout was reduced on day 1 after exercise ($p = 0.043$, compared to the first bout) in the high responders, but no training effect was noted for the low responders ($p > 0.05$). VL muscle thickness did not differ between high and low responders independently of the exercise bout ($p > 0.05$ for all comparisons).

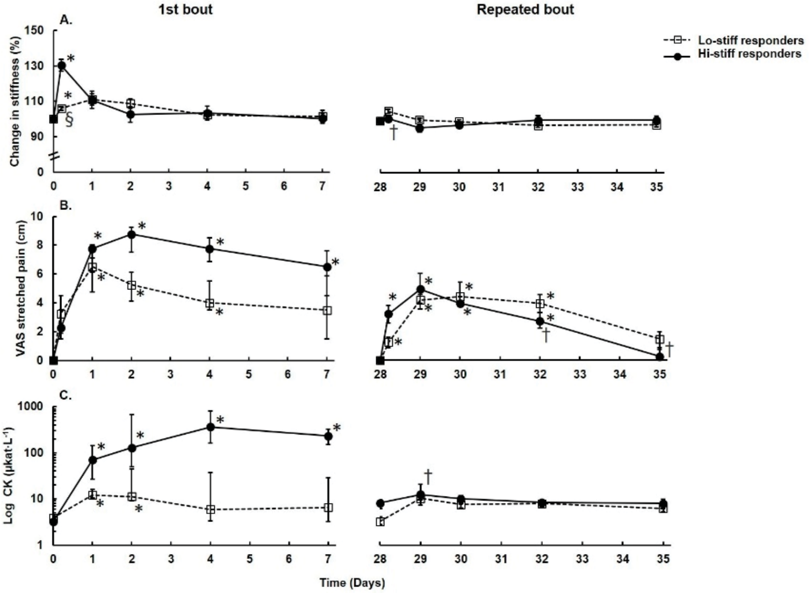

Figure 3. Variable responses to the repeated bout identified as the change in passive MTU stiffness. Filled circles and solid lines are the data for the highest stiffness response ("high responders," $n = 4$); open circles and dashed lines are the data for the lowest stiffness response ("low responders," $n = 4$). (**A**) Change in passive MTU stiffness. (**B**) Visual analog scale score for the pain of stretched muscle. (**C**) Plasma creatine kinase (CK) activity on a log scale. Data are presented as the median and interquartile range (IQR) except for stiffness data, which are presented as the mean and standard error of the mean. * Significant difference from the pre-exercise value ($p < 0.05$). † Significant difference from the first exercise bout ($p < 0.05$). § Significant difference between high and low responders ($p < 0.05$).

4. Discussion

The primary purpose of this study was to examine the dynamics of exercise-induced muscle pain, swelling, CK release, and passive stiffness after the first and repeated bout of eccentric exercise of the knee extensors. The secondary aim was to indirectly compare the different aspects of the response to acute and repeated damage-evoking exercise between lower vs. upper bodymuscle groups as the current study applied a protocol for high-volume eccentric leg exercise to induce muscle damage of a magnitude comparable with that for the arm muscles in the previous study [4]. The results of the present study, in which 200 maximal-effort eccentric contractions of the knee extensors were used, have confirmed the major RBE on the stretched pain, pressure pain threshold (tenderness), and swelling in the exercised muscle. However, the protective effect against exercise-induced MTU stiffness was small and was not evident for the change in plasma CK activity, mainly because both of these markers had large interindividual variability. The high responders for MTU stiffness, however, exhibited a more pronounced response for muscle pain and plasma CK activity after the first exercise bout. In addition, the induction of changes in all three markers was blunted after the repeated exercise bout for the high responders. Even though to a small degree, the results of the current study additionally support the link between stiffness, pain, and CK activity as observed previously in a study on elbow flexors [4].

The peak knee extensor MTU stiffness was detected already immediately after the first eccentric exercise bout and lasted for 1 day, after which it gradually resolved within

7 days. This pattern corresponds with that of peak gastrocnemius stiffness 24 h after exercise [18]. The change in peak passive torque measured immediately after exercise may be related to an acute increase in the water content within the muscle after highly metabolically demanding exercise [19]. We have noted a large variation of the changes in MTU stiffness in response to exercise. Variability in the extent of EIMD, as reflected by differences in muscle force loss, increased plasma CK activity, muscle soreness, swelling, and reduced ROM (increased MTU stiffness) may well have a genetic component, which may be reflected in the different individual responses in exercise-induced upregulation of heat shock proteins [20]. Regarding methodology for MTU stiffness, in our study, it depended on the properties of many structures, including agonists, antagonists, and noncontractile connective tissue, rather than providing a direct measure of the stiffness of the specific muscle [21].

There are several possible explanations of the origin of stiffness associated with EIMD. These encompass the release of Ca^{2+} because of membrane damage during eccentric exercise, increase in Ca^{2+} entry [3] and activity of calpains via stretch-activated channels [22], and changes in titin secondary to effects on the calcium-dependent pathway [23]. In the current study, we did not examine the precise mechanisms underlying the development of MTU stiffness after exercise. However, we found a relationship between peak stiffness and peak muscle swelling, although this does not seem to be causal because the markers had very different dynamics, as has been observed in elbow flexors [4].

In the present study, MTU stiffness in leg muscles lasted a shorter time compared with the exercise-induced stiffness observed in elbow flexors, which did not return to baseline until ~2 days after the bout of eccentric exercise [4]. The different responses of the upper vs. lower limbs might be explained by the extent of muscle damage evoked because the arm muscles are more susceptible to EIMD [11]. The plasma CK activity response was four times higher in a study of elbow flexors [4] than observed in the knee extensors in the present study. This difference implies that greater damage can be induced with only 60 maximal eccentric contractions of the smaller arm flexors compared with the 200 eccentric contractions performed by the large musculature of the leg extensors in our study. Other studies have also reported greater susceptibility to damage in the arm than leg muscles in young adults performing exercise at the same relative intensity [24].

With elbow flexors eccentric exercise, we have previously reported significant RBE for ROM, upper arm circumference, and stretch-induced pain, while only a small RBE for plasma CK activity and pressure-induced pain had been detected [4]. A magnitude of the protection of the repeated exercise bout, a "protective index", could be calculated for each marker of muscle damage as a difference between the response to the first and repeated bout divided by the value of the response to the first bout and multiplied by 100; as such, a higher number of the protective index means a larger protective effect. The protective effect of the first bout for pain was ~50% for the arm muscles in our previous study [4], and it was ~33% for the leg muscles in the current study. These findings are consistent with those of Chen et al. [25], who found no significant difference between various muscle groups in the magnitude of the protection against soreness after the repeated exercise bout of 50 eccentric contractions. Notwithstanding, the protective effect of the first exercise bout against muscle soreness was reported as larger for elbow flexors than for knee extensors [11], and the protective effect of the first bout was 99–100% for plasma CK activity, which contrasts with the findings of our studies. These differences may reflect the high response variability between individuals.

The increase in MTU stiffness in this study was less substantial after the repeated than the first bout, as a similar finding to Margaritelis et al. [8]. Pincheira et al. [6] did not observe an RBE for exercise-induced stiffness and postulated that RBE is likely to be associated with noncontractile elements of the muscle rather than changes in mechanical factors or neural activation. The MTU stiffness response to eccentric exercise differed substantially between individuals in our study (Figure 3). It is notable that the high responders for MTU stiffness exhibited an RBE for MTU stiffness, stretch-induced pain, and plasma CK activity. By contrast, the low responders did not exhibit an RBE for any of the measured variables. These results here are largely consistent with previous findings that slow muscle recovery following eccentric exercise is characteristic in high responders and reflects mainly secondary damage, as indicated by swelling and increased CK activity, and that the faster recovery following a repeated bout occurs because of suppressed secondary damage response [17].

The major limitation of the current study is small sample sizes, whereas this corresponds with those in other similar studies where the numbers of participants ranged between 7 and 18 [1,26]. There was, however, a lack of statistical power in subgroup analyses that were used to present the typically high variability of response levels as presented in Figure 4 of our previous study [17]. The other limitation of the present study was that the knee extensors were investigated as the whole complex of contractile and adjacent connective tissues rather than by measuring the passive tension of only the muscle fibers. The advantages of using shear wave elastography might be considered when measuring the response of stiffness of a muscle or tendon unit to eccentric exercise. Using shear wave elastography, muscle stiffness of the elbow flexors when tested at long length (160°) peaked at 1 h after exercise that induced EIMD and persisted up to 21 days after exercise [27].

5. Conclusions

In summary, we have reported the RBE for MTU stiffness of the knee extensors in response to high-volume intense eccentric exercise. Changes in MTU stiffness were minor associated with muscle pain and plasma CK activity. However, there were large individual differences, which may also explain why RBE for MTU stiffness is difficult to detect at a group level.

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Institutional Review Board Statement: The study was conducted according to the guidelines of the Declaration of Helsinki and approved by the Ethics Committee of the Kaunas Regional Biomedical Research Ethics Committee (registration no. BE-2-17).

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: The datasets used and analyzed during this study are available from the corresponding author on reasonable request.

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

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Article

Impact of Real and Simulated Flights on Psychophysiological Response of Military Pilots

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Abstract: Objective: The present research aimed to analyse the autonomic, anxiety, perceived exertion, and self-confidence response during real and simulated flights. Methods: This cross-sectional study participated 12 experienced male pilots (age = 33.08 (5.21)) from the Spanish Air Force. Participants had to complete a real and a simulated flight mission randomly. The heart rate variability (HRV), anxiety, self-confidence, and rating of perceived exertion were collected before and after both manoeuvres, and HRV was also collected during both simulated and real flights. Results: When studying the acute effects of real and simulated flights, the mean heart rate, the R-to-R interval, the cognitive anxiety and the perceived exertion were significantly impacted only by real flights. Furthermore, significant differences in the mean heart rate and RR interval were found when compared to the acute effects of real and simulated flights (with higher acute effects observed in real flights). Additionally, when compared the HRV values during simulated and real flights, significant differences were observed in the RR and heart rate mean (with lower RR interval and higher heart rate mean observed during real flights). Conclusion: Real flights significantly reduced the RR interval and cognitive anxiety while increased the heart rate mean and the rating of perceived exertion, whereas simulated flights did not induce any significant change in the autonomic modulation.

Keywords: HRV; army; simulator; flight; anxiety; perceived exertion

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1. Introduction

Flight simulators have been used in aviation as an economical and safety tool to train pilots, providing a simulated environment which could mimic real conditions [1]. In order to study the transfer between training conditions and aircraft [2], previous studies have used different psychophysiological tools to investigate the cognitive demands of both simulated and real flights [3,4]. This is relevant since this operation requires higher cognitive demands [2,5–7] and, therefore, the evaluation of the mental workload has emerged as a cornerstone.

Heart rate variability (HRV) is a non-invasive tool which studies the successive heartbeats variation [8]. This evaluates the balance between sympathetic and parasympathetic nervous systems, as well as it has been considered a measure of heart-brain interaction since it could be modified by cognitive, attentional, anxiogenic, or physical stimulus [9–11]. Thus, when the activity of the sympathetic nervous system predominates, the HRV is reduced. Instead, when parasympathetic activity increases, the HRV is higher. For these reasons, the HRV is considered as a cognitive load biomarker [11–13].

HRV measures have been performed in military pilots to study the sympathetic activity during flights [14,15]. In this regard, Sauvet, et al. [16] showed that flights induced a progressive decrease of RR intervals, increasing the sympathetic activity. The sympathetic activation could induce increased anaerobic metabolism or increased anxiety and stress perceptions among other psychophysiological effects [17]. Autonomic modulation analysis was used in a military population as a stress marker, showing how different military manoeuvres, independently of land or flight units, produced an increased sympathetic modulation [18]. Moreover, the fatigue induced by flights can also be detected by spirometry, handgrip strength, and stress and exertion rates as previous authors showed in this special population [19–22].

The development of flight simulators has allowed its use as a way of training, avoiding high-risk conditions with catastrophic consequences. Due to this fact, as well as to economise training (since real flights have enormous costs), the number of real flights has decreased whereas the number of hours in the flight simulator increased. However, the psychophysiological response of these conditions (real and simulated flights) and the comparison between real and simulated flight condition have been poorly studied. Therefore, the aims of the present study were: (1) To analyse the acute effects of a simulated and a real flight in the autonomic modulation, anxiety, perceived exertion, and self-confidence; and (2) to compare the autonomic modulation during a real and a simulated flight. Since flight simulator cannot mimic the physical demands of a real flight, our hypotheses are: (1) The impact of acute effects (on HRV and anxiety) will be higher in the real than in the simulator; and (2) the HRV will be lower during a real flight than during a simulated flight.

2. Materials and Methods

2.1. Participants

Twelve experienced military pilots participate in this cross-sectional study. Pilots had a mean age of 33.08 (5.21) years and an experience of 13.25 (5.15) years of military service. Procedures were approved by the university ethics committee (approval number: 206/2019) as well as pilots agreed to participate in this study, giving written consent. Table 1 shows pilots' characteristics.

Table 1. Characteristics of military pilots.

Variable	Mean (SD)
Age (years)	33.08 (5.21)
Military service (years)	13.25 (5.15)
Fear to an accident(0–100)	28.33 (25.17)

2.2. Procedure

Participants were evaluated before, during and after two flights: (1) A real flight with a F5 aircraft; and (2) a simulated flight with an operational F-5 M (Indra Company, Madrid, Spain) flight simulator. Each protocol, real or simulated flights, were performed on consecutive separate days. The order between simulated and real flights was randomised. The mission, both simulated and real, were the same with: (1) An individual takeoff; (2) G-warm up/G- awareness below FL 180; (3) air–air mission with two set-ups; (4) air–ground attack on the selected target, and (5) landing without reduction of visibility. Both real and flight protocols lasted 45 min. For analysis purposes, the whole protocol duration (with all the manoeuvres) was used to study the HRV during real and simulated flights.

HRV, anxiety, perceived exertion, and self-confidence were assessed immediately after and before real and simulated missions. The HRV baseline was recorded once starting the protocol. Moreover, the HRV was assessed during both real and simulated flight mission. In order to avoid potential confounding factors, participants were asked to not take alcohol, coffee, or caffeinated drinks 24-hours before undergoing the protocol, since their consumption could affect the nervous system, and, therefore, HRV variables. None of the participants had smoking habits or took cardioactive medication such as antidepressant, antipsychotic, or antihypertensive medication [23].

2.3. HRV Acquisition and Preprocessing Steps

The HRV was recorded using a reliable heart rate monitor (Polar RS800CX, Oy, Kempele, Finland) [24] and analysed with the Kubios HRV software (v. 3.3) [25]. The Task Force's recommendations of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology [26] were followed in this study. Thus, the baseline measure lasted 5 min (which is considered a short-term record). RR data recorded by the heart rate monitor was exported to the Kubios HRV software where different preprocessing steps were applied. In this regard, a middle filter was applied to correct possible artefacts. Those RR intervals which are shorter/longer than 0.25 s, compared to previous beats average were automatically identified, as well as they were replaced by cubic spline interpolation [27,28].

2.4. Outcomes

2.4.1. HRV

Time, frequency and non-linear variables were calculated using the Kubios HRV software. In the time domain, the mean heart rate (mean HR), RR intervals, RR50 count divided by the total number of all RR ranges (Pnn50), and the square root of differences between adjacent RR intervals (RMSSD) were extracted. In the frequency domain, the low frequency (LF, 0.04–0.15 Hz) and high frequency (HF, 0.15–0.4 Hz) ratio (LF/HF) and Total Power were included. The non-linear measures, such as RR variability from heartbeat to short term Poincaré graph (width) (SD1) and RR variability from heartbeat to long term Poincaré graph (length) (SD2). Further information regarding HRV variables can be found in the following articles [9,29].

2.4.2. Anxiety Measurements

The Competitive State Anxiety Inventory-2R (CSAI-2R) (Spanish version) was used to assess the pre-competitive anxiety of the participants [30,31]. This questionnaire has 17 items, where cognitive anxiety, somatic anxiety and self-confidence can be extracted. Moreover, anxiety was also measured by the State-Trait Anxiety Inventory (STAI-E) (Spielberger et al., 1971), which consisted of 20 items and where the participants reported their state of anxiety at that time. The score range for the test is 20–80, indicating a higher level of anxiety a higher score [32]. Additionally, the perceived exertion was assessed by the rating of perceived exertion (RPE) 6–20 scale [33].

2.5. Statistical Analysis

The SPSS statistical package (version 20.0; SPSS, Inc., Chicago, IL, USA) was used to analyse the data. Following the results of a Shapiro–Wilk test, non-parametric tests were employed.

The Wilcoxon signed-rank test was used to examine the difference between the pre- and post-measures for each variable. Moreover, in order to compare the impact of real vs. simulated flight, the post-values were normalised (by subtracting the baseline). Effect sizes [r] were calculated for the non-parametric tests, classified as follows: 0.5 is a large effect, 0.3 is a medium effect and 0.1 is a small effect [34,35].

3. Results

3.1. HRV and Perceived Anxiety and Self-Confidence before and after a Real and a Simulated Flight

Table 2 shows the acute effects of a real vs. a simulated flight mission on the HRV of experienced pilots. Significant results were obtained in the mean HR and RR variables (p-value < 0.05) when compared the baseline and the post-measure after a real flight. Regarding simulated flight, significant differences were not found between baseline and post-measure. Moreover, significant differences were obtained when compared to real vs. simulated flight. In this regard, significant differences were found in mean HR and RR, indicating higher mean HR and lower RR intervals after a real flight.

Table 2. Acute effects of a real vs. a simulated flight mission on heart rate variability (HRV).

Flight Conditions		**Baseline**	**Post**	**Baseline vs. Post Measure**		**Acute Effects of a Real vs. a Simulated Mission**	
	Variables	**Mean (SD)**	**Mean (SD)**	***p*-Value**	**Effect Size**	***p*-Value**	**Effect Size**
Real F. Simulated F.	Mean HR	69.82 (10.70)	98.06 (11.89) 74.56 (14.39)	0.003 * 0.155	0.847 0.411	0.005 *	0.809
Real F. Simulated F.	RR	885.54 (136.14)	631.91 (77.11) 837.70 (144.35)	0.003 * 0.155	0.847 0.411	0.005 *	0.809
Real F. Simulated F.	Pnn50	19.34 (17.18)	8.65 (8.28) 16.04 (12.08)	0.062 0.929	0.539 0.026	0.139	0.426
Real F. Simulated F.	RMSSD	44.28 (25.96)	27.77 (13.50) 37.01 (14.39)	0.062 0.424	0.539 0.231	0.139	0.426
Real F. Simulated F.	SDNN	51.05 (27.29)	41.48 (13.88) 45.30 (11.91)	0.477 0.594	0.205 0.154	0.241	0.338
Real F. Simulated F.	HF	35.49 (18.01)	24.43 (12.12) 28.76 (16.45)	0.155 0.075	0.411 0.513	0.721	0.103
Real F. Simulated F.	LF	64.44 (18.00)	75.51 (12.14) 71.17 (16.47)	0.155 0.075	0.411 0.513	0.721	0.103
Real F. Simulated F.	LF/HF	2.95 (2.94)	4.20 (3.14) 3.69 (2.65)	0.213 0.328	0.359 0.282	0.508	0.191
Real F. Simulated F.	Total Power	2931.89 (3830.04)	1953.40 (1200.12) 2090.28 (1078.28)	0.859 0.286	0.051 0.308	0.508	0.191
Real F. Simulated F.	SD1	31.37 (18.42)	19.64 (9.54) 26.18 (10.18)	0.062 0.424	0.539 0.231	0.139	0.427
Real F. Simulated F.	SD2	64.54 (34.65)	55.04 (17.79) 58.09 (14.66)	0.594 0.534	0.154 0.179	0.285	0.309

* *p*-value < 0.05. F: Flight; HR: Heart rate; RR: Time between intervals R-R; pNN50: Percentage of intervals >50 ms different from the previous interval; RMSSD: The square root of the mean of the squares of the successive differences of the interval RR; LF/HF: Low frequency (LF) ratio (ms2)/High frequency (HF) (ms2); Total power: The sum of all the spectra; SDNN: Standard deviation of normal-to-normal intervals; SD1: Dispersion, standard deviation, of points perpendicular to the axis of line-of-identity in the Poincaré plot; SD2: Dispersion, standard deviation, of points along the axis of line-of-identity in the Poincaré plot.

Table 3 shows the acute effects of a real and a simulated flight mission on the rating of perceived exertion, anxiety, and self-confidence of experienced military pilots. In this regard, differences between baseline and post-measure were found in the rating of perceived exertion (*p*-value = 0.010) and the cognitive anxiety (*p*-value 0.024), showing an increased rating of perceived exertion and lower cognitive anxiety after a real flight. Regarding differences between real and simulated flight, statistically, and significant differences were not achieved by any of the studied variables.

Table 3. Acute effects of a real vs. a simulated flight mission on the rating of perceived exertion, anxiety and self-confidence.

Flight Conditions		Baseline	Post	Baseline vs. Post-Measure		Acute Effects of a Real vs. a Simulated Mission	
	Variables	Mean (SD)	Mean (SD)	*p*-Value	Effect Size	*p*-Value	Effect Size
Real F. Simulated F.	RPE	8.42 (1.88) 7.92 (2.23)	11.17 (2.33) 8.82 (2.64)	0.010 * 0.089	0.742 0.491	0.218	0.355
Real F. Simulated F.	STAI-E	27.42 (8.26) 24.82 (3.87)	28.17 (9.37) 24.75 (4.55)	0.574 0.459	0.162 0.213	0.633	0.138
Real F. Simulated F.	Cognitive anxiety	6.92 (3.15) 7.00 (2.63)	6.17 (2.37) 6.33 (2.19)	0.024 * 0.074	0.653 0.515	0.931	0.024
Real F. Simulated F.	Somatic anxiety	9.33 (1.72) 8.75 (1.71)	9.67 (2.23) 9.08 (1.97)	0.776 0.285	0.082 0.308	0.832	0.061
Real F. Simulated F.	Self-confidence	18.67 (2.39) 19.33 (1.23)	19.08 (1.93) 19.25 (1.76)	0.180 0.705	0.387 0.109	0.221	0.354

* *p*-value < 0.05. F: Flight; CSAI-2R: Competitive State Anxiety Inventory-2R; RPE: Rating of perceived exertion; STAI: State-Trait Anxiety Inventory.

3.2. HRV during a Real and a Simulated Flight

Table 4 shows the HRV during simulated and real flights. Results showed significant differences between real and simulated flights, exhibiting higher mean HR and lower RR interval during a real flight.

Table 4. HRV during simulated and real flights.

Variables	Real Flight Mean (SD)	Simulated Flight Mean (SD)	*p*-Value	Effect Size
Mean HR	93.81 (15.41)	70.83 (12.48)	0.003 *	0.847
RR	660.30 (106.86)	875.58 (139.10)	0.003 *	0.847
Pnn50	11.75 (8.83)	17.69 (14.26)	0.131	0.436
RMSSD	32.52 (14.34)	38.93 (18.04)	0.110	0.461
SDNN	53.20 (23.73)	47.98 (20.55)	1.000	<0.001
HF	20.95 (12.95)	29.06 (17.28)	0.155	0.411
LF	79.02 (12.96)	70.89 (17.27)	0.155	0.411
LF/HF	5.87 (4.11)	3.84 (2.94)	0.131	0.436
Total Power	2750.74 (1733.18)	2721.90 (2647.31)	0.929	0.026
SD1	23.02 (10.15)	27.56 (12.78)	0.110	0.462
SD2	71.14 (31.89)	61.77 (26.66)	0.657	0.128

* *p*-value < 0.05. HR: heart rate; RR: Time between intervals R-R; pNN50: Percentage of intervals >50 ms different from the previous interval; RMSSD: The square root of the mean of the squares of the successive differences of the interval RR; LF/HF: Low frequency (LF) ratio (ms2)/High frequency (HF) (ms2); Total power: The sum of all the spectra; SDNN: Standard deviation of normal-to-normal intervals; SD1: Dispersion, standard deviation, of points perpendicular to the axis of line-of-identity in the Poincaré plot; SD2: Dispersion, standard deviation, of points along the axis of line-of-identity in the Poincaré plot.

4. Discussion

This study aimed to analyse the autonomic, anxiety, perceived exertion, and self-confidence response during real and simulated flight. Regarding the first hypothesis, "the impact of acute effects (on HRV and anxiety) will be higher in the real than in the simulator", cannot be accepted since not all the variables reached the significance level (*p*-value < 0.05). Only significant differences (between baseline and post measures) were found in the mean HR, RR interval, cognitive anxiety and perceived exertion in the real flight. Moreover, significant differences in the acute effects were observed in the mean HR and RR intervals between real and simulated flights (with significantly higher values of mean HR and lower RR interval values after the real flight). Regarding the second hypothesis, "the HRV will be lower during a real flight than during a simulated flight" cannot be totally accepted since only mean HR and RR interval showed significant differences between real and simulated flights. Higher values of mean HR and lower values of RR interval were observed during real flights.

These results highlighted the importance of flight simulator for training purposes in pilots. Not all the expected differences were reached, which could mean that planning and task design of the simulated task are close to the real condition. However, since the simulator cannot mimic the G forces or vibration, the significant differences which can be observed in the RR interval, mean HR, and perceived exertion can be derived from the physical demands of real flights. Furthermore, the differences in the cognitive anxiety observed before a real and simulated flight can be due to the responsibility and risk derived from real flights. Therefore, in order to totally mimic real conditions, future flight simulators should incorporate immersive virtual reality technology simulating G forces and vibration. Additionally, the relevant information that provides the HRV (in both cognitive and physical spheres) could be used to design more individualised training controlling the training load and therefore increasing the efficiency and the performance of pilots during flights.

The study of the HRV informed about the balance between parasympathetic and sympathetic nervous systems. Previous studies in aviation field showed how simulated and real flights could reduce the HRV [3,5,15,21]. This is relevant due to the negative impact of increased sympathetic activity that produces a reduction in memory and decision making processes [36,37]. However, our results showed that HR mean and RR intervals were negatively impacted by real flights while other time domain, frequency domain or non-linear measures were not. Since the mean heart rate was below 100 beats/min, it would suggest that these differences were due to a parasympathetic activity reduction rather than an increase in sympathetic activity. A previous study that analyses the effect of defence and attacks air combat manoeuvres on air combat fighter pilots' psychophysiological response did not detect significant differences between pre versus post or pre versus during flight [15]. Authors stated that this behaviour could be due behaviours' anticipatory anxiety response which started before the flight manoeuvres [15]. Nevertheless, in our study, statistically significant differences were not observed in the HRV when compared real vs. simulator or even pre vs. post (for both real and simulator conditions).

In this regard, previous studies have highlighted the usefulness, due to the high sensitivity, of this variable (RMSSD) to measure the autonomic modulation [21,38]. However, our results did not show any significant effect on this variable. Hypothetically, these results could also be explained since all the analysed pilots were expert. A previous study reinforces this explanation since lower HRV values were related to a lower level of experience [20]. Authors explained that this could be an adaptive response to this stressful environment [20]. Another study showed that a pilot's first flight was the most stressful [7,39]. Thus, novice pilots might exhibit higher sympathetic, reducing the HRV. Therefore, future studies should focus on comparing the different level of expertise in pilots.

Previous studies have investigated the impact of flights on the anxiety and rating of perceived exertion. Regarding anxiety, it was shown [21] that anxiety was higher before flight missions, highlighting the anticipatory anxiety response of pilots. This was in line with our results when higher cognitive anxiety was found before a real flight which could be due to the responsibility of flight a real aircraft. This is congruent with the HRV results where differences were not obtained (significant differences were not achieved between pre and post or during flight assessments), and an anticipatory response emerged as a possible explanation. Moreover, regarding the rating of perceived exertion, real flight exhibited higher values than simulated one. This could be due to the stress and anxiogenic response induced by mechanical load such as vibration or G forces which pilots have to suffer during a real flight. This is also supported by the acute effects observed on the autonomic modulation where significant differences were reported on RR and heart rate mean.

This study has some limitations which should be highlighted. First, the sample size was small, so probably only large differences have reached the statistical significance level. Second, the sample was composed by experienced pilots which mean that results cannot be extrapolated to novice pilots. Third, the SPSS package uses z-ratio, which is applicable for at least ten observations. Although this study has twelve observations, results from Wilcoxon signed-rank test could be affected by this issue. Fourth, the breathing rate was not controlled, so the respiratory sinus arrhythmia can impact RR intervals. Lastly, the simulator did not mimic all the real conditions such as vibration, G forces or even oxygen deprivation. Thus, flight simulator could be improved, incorporating this to the simulation in order to make more realistic training for pilots.

5. Conclusions

Real flights significantly reduced the RR interval and cognitive anxiety while increased the heart rate mean and the rating of perceived exertion whereas simulated flights did not induce any significant change in the autonomic modulation

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Body Composition in International Sprint Swimmers: Are There Any Relations with Performance?

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Abstract: The paper addresses relations between the characteristics of body composition in international sprint swimmers and sprint performance. The research included 82 swimmers of international level (N = 46 male and N = 36 female athletes) from 8 countries. We measured body composition using multifrequency bioelectrical impedance methods with "InBody 720" device. In the case of male swimmers, it was established that the most important statistically significant correlation with sprint performance is seen in variables, which define the quantitative relationship between their fat and muscle with the contractile potential of the body (Protein-Fat Index, r = 0.392, $p = 0.007$; Index of Body Composition, r = 0.392, $p = 0.007$; Percent of Skeletal Muscle Mass, r = 0.392, $p = 0.016$). In the case of female athletes, statistically significant relations with sprint performance were established for variables that define the absolute and relative amount of a contractile component in the body, but also with the variables that define the structure of body fat characteristics (Percent of Skeletal Muscle Mass, r = 0.732, $p = 0.000$; Free Fat Mass, r = 0.702, $p = 0.000$; Fat Mass Index, r = −0.642, $p = 0.000$; Percent of Body Fat, r = −0.621, $p = 0.000$). Using Multiple Regression Analysis, we managed to predict swimming performance of sprint swimmers with the help of body composition variables, where the models defined explained 35.1 and 75.1% of the mutual variability of performance, for male and female swimmers, respectively. This data clearly demonstrate the importance of body composition control in sprint swimmers as a valuable method for monitoring the efficiency of body adaptation to training process in order to optimize competitive performance.

Keywords: sprint swimmers; body composition; results prediction; body fat; skeletal muscle mass

1. Introduction

A sports training system represents a long-lasting and multicomplex process in which programmed training loads are applied. It is well documented in scientific literature that has examined long-term athlete development that selected athletes need eight to twelve years of systematic sports training with approximately two or three hours of daily practice to achieve an elite level with full performance potential [1,2].

The improvement of the sports training system has led to better sport achievements with the help of increased athlete performance. The same phenomena were established in swimming in the last 50 years, regardless of swimmers' age and gender [3].

Biologically, development of athletes as a specifically selected and highly trained population, is constantly subjected to adaptation. It is well known that there are many extraneous factors (genetics, anatomical, neurological, hormonal, psychological, cognitive etc.) that must be incorporated within the planning of any specific form of physical training. All these systems are influenced by the hetero-chronic phenomenon, that is, they are time independent in their biological development, but all of them affect the physiological systems of the body [4].

One of the mechanisms of sports training is the morphological adaptation of the body. Adaptation in sports is always triggered by training and training components, such as the type of intensity or training load, which is aimed at changing the morphological characteristics of the body to reach a typical body structure [4–9]. This phenomenon is only occurring to those body tissues that are subjected to biological adaptation, as are fat and muscle tissue or even a bone tissue component [9–11].

The published evidence on morphological factors which contributes to the development of elite performance in swimming, has not been represented sufficiently in top scientific literature in the previous 35 years [11–15]. Competitive swimmers, especially Olympic elite swimmers, were found to be taller than sub-Olympic. The longitudinal data from Russian national swimming teams demonstrated that current swimmers tend to be taller than in the past. For example, 17-year-old male swimmers have an average body height of more than 185 cm [16].

In swimming, as a form of human locomotion in water, as a physical medium, metabolic power (the energy expended in the unit of time) exponentially increase as a function of speed (as a “measure” of exercise intensity). The maximum swimming speed will be achieved by the swimmer who can achieve higher maximum metabolic power for lower energy consumption during swimming [17]. The overall efficiency of swimming depends on the propulsive efficiency and the hydrodynamic resistance (different aspects of passive and active drag form) that the body creates as it moves through the water. Different forms of water resistance, as a sum of overall drag, during swimming is affected by the swimmer’s body shape and morphology, body density and body position in a water—as passive drag components; or depends on lots of comprehensive factors such as friction drag, frontal area and body shape resistance drag, and wave drag—as active drag components [15,18]. To enable an athlete to swim faster, the swimmer must either increase the propulsive force or reduce the drag force, or, ideally, do both [17,19,20].

Body composition is a term that describes the relative proportions of all main components of the body, including fat, bone, muscle, and water mass [21]. In the last decade, bioelectricl impedance analysis (BIA), especially direct segmental multifrequency methods, have been widely used in science and sport practice—along with the other traditional body composition methods as skinfold measurements, dual-energy X-ray absorptiometry, body density measurement, and total body water estimates—and had become a standard method for determination of complete body structure according to the body segments [7,22–24].

Evaluating the relationship of body composition to swim performance on the sample of 280 competitive female swimmers it was found that Body Height, Body Mass, Lean Body Mass (LBM), and Residual Lung Volume (RV) were in correlation, which is statistically significant but inverse to the time recorded for the 100-yard swim [25]. During competitive swim season, significant increase of LBM and decrease of fat mass has occurred primarily during the part of the season when training was intense [12]. Increasing muscle size and improvement of muscle contractile quality, and decreasing fat mass, at a level of optimal balance, may have positive effect on swimming performance, and is likely to be important for maximizing competitive swimming performance. Moreover, seasonal swim performance changes tend to follow changes in muscle mass, so regular control of body composition during the season may be helpful and beneficial in order to improvement training and performance [11].

This research is aimed at defining the relations between swimming performance and the characteristics of body composition in international sprint swimmers, measured with the multifrequency bio-impedance method. The secondary goal of the research is to define a multidimensional model of performance prediction, based on the most sensitive variables of body composition in swimmers.

This model can serve as a tool for controlling, achieving, and maintaining optimal body composition of sprinter swimmers.

All the results obtained in this research may be used to improve sports training technologies in sprint swimming disciplines in the athletes of an international level.

2. Materials and Methods

This research was conducted using Cross-Sectional Designs, applied according to the research methods accepted for physical activity and sport [26]. Moreover, in this study, we used a multicentric study design and laboratory testing method. This study possesses the characteristics of fundamental and applied research, aimed at expanding the existing knowledge about the body composition of international swimmers in relation to sprint performance potential. Sprint performance has been expressed as FINA point score (Federatioin Internationale de Natation, http://www.fina.org/content/fina-points), where FINA points score allows standardized comparisons of swimming performances in different events.

2.1. Research Sample

Eighty-two swimmers of an international level (N = 46 male athletes, age—22.9 ± 4.2 years, training experience—14.60 ± 5.6 years, FINA Score = 785 ± 71, Min = 638–Max = 883 and N = 36 female swimmers, FINA Score = 727 ± 98, Min = 642–Max = 910, age—21.0 ± 4.7 years, training experience—12.7 ± 4.6 years) participated in this research. The participants were from the following 8 countries: Serbia (N = 33, 20 males and 13 females), Slovenia (N = 23, 9 males and 14 females), Lithuania (N = 10 males), Russia (N = 5 males), Estonia (N = 3 females), Belarus (N = 3 females), and Bosnia and Herzegovina (N = 3 females). Participants used the following stroke styles: 32 freestyle (15 males and 17 females), 14 backstroke (5 males and 9 females), 20 breaststroke (16 males and 4 females), and 14 fly swimmers (10 males and 6 females). All swimmers were competitors at the distance of 50 m or 100 m.

2.2. Measurement Procedure

Before testing, the participants, all of them volunteers, were informed about measurement conditions and procedures. The overall process of sample tracking was carried out during the period 2012–2017. The study was conducted at the premises of three scientific laboratories: the laboratory of the Faculty of Sport and Physical Education at the University of Belgrade (Serbia); the laboratory of the Faculty of Sport at the University of Ljubljana (Slovenia); and the laboratory of the Faculty of Sport Bio-medicine at Lithuanian Sports University, Kaunas (Lithuania). The research was performed in accordance with the conditions of Declaration of Helsinki: Recommendations Guiding Physicians in Biomedical Research Involving Human Subjects (http://www.cirp.org/library/ethics/helsinki/), and with the approval and consent of the Ethics Committee of all three faculties, and under the supervision of The Institutional Ethical Board of Faculty of Sport and Physical Education, University of Belgrade Serbia, approved the study (No. 484-2).

2.2.1. Body Composition Variables

We measured body composition using bioelectrical impedance analysis (BIA) with InBody 720 device that used Tetapolar 8 points by tactical electrodes system with DSM-BIA (Direct Segmental Multifrequency Bioelectrical Impedance Analysis) (Biospace Co, Ltd., Seoul, Korea). Inbody 720 device [27] demonstrated high test-retest reliability and accuracy (ICC 0.9995). It is regarded to be highly statistically reliable and valid for measuring both overall and segmental body composition in female and male athletes [22,24,28].

All participants were measured in accordance with manufacturer's suggestions and previously published procedures [7,8,27,29].

For this study, we used 14 variables, 5 of which were basic and 9 were derived (index) variables, defining the morphology and composition of the body according to the following criteria: basic component variables, voluminosity independent variables, longitudinal independent variables, and index variables.

Basic component variables (5): BH—body height, cm; BM—body mass, kg; BF—body fat mass, kg; SMM—skeletal muscle mass, kg; FFM—fat free mass, kg.

The voluminosity independent variables (3): PBF—percent of body fat, calculated as: BF (body fat, kg)/BM (body mass, kg), %; PSMM—percent of skeletal muscle mass, calculated as: SMM (skeletal muscle mass, kg)/BM (body mass, kg), %; PFFM—percent of fat free mass, calculated as: FFM (fat free mass, kg)/BM (body mass, kg), %;

Longitudinal independent variables (3): BMI—body mass index, calculated as: BM (body mass, kg)/BH^2 (body height, m), kg Body mass·m^{-2}; FMI—fat mass index, calculated as: FM (fat mass, kg)/BH^2 (body height, m), kg Body fat·m^{-2}; SMMI—skeletal muscle mass index, calculated as: SMM (skeletal muscle mass, kg)/BH^2 (body height, m), kg Skeletal muscle·m^{-2}; FFMI—fat free mass index, calculated as: FFM (fat free mass, kg)/BH^2 (body height, m), kg FFM·m^{-2};

Derivated (index) variables (2): PFI—protein fat index, calculated as the relation between proteins, as a pure contractile tissue in the body (kg) and body fat mass, as a ballast or non-contractile tissue in the body (kg), kg; IBC—index of body composition, calculated as the relation between BMI (kg) and the percent of body fat mass (%), arbitrary units.

All the variables used are taken from the previously published research [7,8,29,30].

2.2.2. Swimming Performance Variable

The criterion variable was the value of the absolute best result at 50 m or 100 m distance in 50 m pool, achieved in a given competitive microcycle and expressed as FINA score [31]. Body composition was measured on one occasion only within a competitive microcycle, i.e., at least seven days before or after a race during a particular swimming season. Relevant body composition was compared with the actual peak of the results achieved.

2.3. Statistical Procedures

We processed raw results using basic descriptive statistics, namely a central tendency (MEAN), statistical dispersion (SD, cV%, Min and Max measured values), and the parameters of measurement errors. The methods used are the result of the multicentric nature of the study (SEM—Standard Error of Measurements, absolute and relative). The distribution regularity of variables was tested with the help of Kolmogorov–Smirnov nonparametric test (KSZ). We established the partial relations between criteria and body composition variables using Pearson's correlation analysis. With the help of Fisher t-to-z transformation for an independent sample, we calculated if there were statistical differences in the correlations between male and female swimmers, having the same body composition variables and criteria, i.e., performance level. We also used a mathematical modeling by means of Multivariate Regression Analysis (MRA) as a multidimensional prediction system, which helped us to define complex relations between the criteria (swimming performance and FINA Scores) and body composition variables used as predictors [32,33]. All statistical analysis were performed using corresponding statistical software SPSS 19.0. The level of statistical significance is defined at 95% and the probability values of $p < 0.05$ [34].

3. Results

Table 1 demonstrates all statistical data about Male and Female swimmers, respectively. The basic anthropometrical and body composition data were shown that mean value for BH is 186.3 ± 5.4 cm, for BM is 82.4 ± 6.5 kg, for BMI is 23.73 ± 1.35 kg·m^{-2}, percent of body fat (PBF) is 9.82 ± 3.35, and percent of skeletal muscle mass (PSMM) is 52.36 ± 1.83 considering male sprint swimmers sample. Female swimmers had the following basic anthropometrical and body composition mean values: BH = 173.4 ± 5.8 cm, BM = 62.8 ± 4.9 kg, BMI = 20.88 ± 1.13 kg·m^{-2}, PBF = 15.79 ± 4.84, and PSMM = 47.01 ± 2.93. All variables had a normal distribution, except for one—IBC at female sample (KS *p* = 0.031), which indicates that the results can be used as acceptably representative for interpretation of the population of international sprint swimmers.

Table 1. Descriptive statistics for male and female swimmers.

Variables	Male													
	BH	BM	BMI	BF	SMM	FFM	PBF	PSMM	PFFM	FMI	SMMI	FFMI	IBC	PFI
Mean	186.3	82.4	23.73	8.12	43.13	74.29	9.82	52.36	90.18	2.35	12.41	21.38	2.80	2.18
Std. Dev.	5.4	6.5	1.35	3.08	3.55	6.25	3.35	1.83	3.35	0.88	0.63	1.17	1.25	1.08
cV%	2.90	7.83	5.69	37.07	8.23	8.41	33.81	3.50	3.71	36.17	5.08	5.47	43.21	47.25
SEM	0.78	0.95	0.20	0.37	0.52	0.92	0.41	0.27	0.49	0.09	0.09	0.17	4.29	4.59
SEM (%)	0.42	1.15	0.84	4.56	1.21	1.24	4.18	0.52	0.54	3.83	0.73	0.80	4.29	4.59
Min	178.4	71.8	21.42	2.4	36.2	63.1	2.99	48.44	81.14	0.68	10.95	18.85	1.34	0.89
Max	201.5	96.9	27.13	17.8	54.0	93.1	18.86	55.79	97.01	4.79	13.92	23.86	7.66	6.46
KSZ	0.942	0.770	0.542	1.066	0.756	0.678	0.777	0.632	0.765	1.128	0.554	0.428	1.384	1.380
KS *p*	0.338	0.594	0.931	0.206	0.616	0.747	0.581	0.820	0.603	0.157	0.919	0.993	0.051	0.052
	Female													
Mean	173.4	62.8	20.88	9.87	29.52	52.93	15.79	47.01	84.27	3.31	9.80	17.56	1.48	1.21
Std. Dev.	5.8	4.9	1.13	3.01	2.30	5.18	4.84	2.93	4.83	1.11	0.58	0.99	0.48	0.51
cV%	3.36	7.79	5.41	30.50	7.79	9.79	30.65	6.23	5.73	33.53	5.92	5.64	32.43	38.84
SEM	0.97	0.82	0.19	0.40	0.50	0.86	0.78	0.49	0.81	0.16	0.10	0.17	0.07	0.07
SEM (%)	0.56	1.31	0.91	4.05	1.69	1.62	4.94	1.04	0.96	4.83	1.02	0.97	4.73	5.79
Min.	163.0	53.8	19.25	4.4	22.3	41.0	7.50	39.05	70.09	1.47	8.39	15.24	0.73	0.46
Max.	184.4	73.3	23.94	17.5	35.0	62.4	29.91	52.14	92.48	6.51	11.22	20.24	2.61	2.45
KSZ	0.642	0.861	1.052	0.781	0.600	0.630	0.706	0.737	0.710	0.861	0.597	0.589	1.456	1.289
KS *p*	0.804	0.448	0.218	0.575	0.864	0.823	0.702	.649	0.695	0.449	0.868	0.879	0.037	0.072

Table 2 demonstrates a correlation matrix for body composition variables and athletes' performance with Fisher r-to-z transformation results. All statistically different r values between gender considering observed body composition variables are shown in bold format. The largest statistically important difference between individual variables of body composition in relation to the sprint swimming performance as per gender was found in PFFM (t = −2.74, *p* = 0.006) i PBF (t = 2.72, *p* = 0.007), while the smallest difference was in BF (t = 2.13, *p* = 0.033) i FFMI (t = −2.14, *p* = 0.032). In as many as five variables, it was established that the gender was not a source of influence on the performance in sprint swimming (BH, BM, BMI, IBG, and PFI, Table 2).

Table 2. Correlation matrix for body composition variables and athletes' performance with Fisher r-to-z transformation results.

Body Composition Variables		FINA Score Pearsons Correlation Coefficient		Fisher r-to-z Transformation	*p*
		Male	Female		
BH (cm)	r value	0.187	0.535	−1.76	0.078
	p significance	0.212	0.001		
BM (kg)	r value	0.215	0.396	−0.87	0.384
	p significance	0.151	0.017		
BMI (kg·m^{-2})	r value	0.087	−0.085	0.75	0.453
	p significance	0.566	0.621		
BF (kg)	r value	−0.148	−0.566	2.13	0.033
	p significance	0.326	0.000		

Table 2. *Cont.*

Body Composition Variables		FINA Score Pearsons Correlation Coefficient		Fisher r-to-z Transformation	*p*
		Male	Female		
SMM (kg)	r value	0.350	0.730	−2.43	0.015
	p significance	0.017	0.000		
FFM (kg)	r value	0.294	0.702	−2.61	0.009
	p significance	0.047	0.000		
PBF (%)	r value	−0.224	−0.695	2.72	0.007
	p significance	0.135	0.000		
PSMM (%)	r value	0.353	0.732	−2.44	0.015
	p significance	0.016	0.000		
PFFM (%)	r value	0.223	0.697	−2.74	0.006
	p significance	0.136	0.000		
FMI (kg·m^{-2})	r value	−0.170	−0.642	2.55	0.011
	p significance	0.260	0.000		
SMMI (kg·m^{-2})	r value	0.323	0.684	−2.17	0.030
	p significance	0.029	0.000		
FFMI (kg·m^{-2})	r value	0.228	0.621	−2.14	0.032
	p significance	0.127	0.000		
IBC (Arbitraly Unit)	r value	0.391	0.687	−1.85	0.064
	p significance	0.007	0.000		
PFI (kg)	r value	0.392	0.655	−1.60	0.110
	p significance	0.007	0.000		

Table 3 demonstrates MRA statistical results with the multiple equation models of athletes' sprint swimming performance prediction by body composition characteristics. Of those variables, which are part of the predictor system for the swimming sprint performance, the three most influential ones in males are BM, BH, and PSMM (body mass, height, and the muscle percentage) while for females those are PBF, PSMM, and PFI (the percentage of fat and muscles and the structural ratio of fat and protein). For males, 35.1% of the common variance with the criterion is explained, while 75.1% for female swimmers. Thus, there is significantly more other factors then body composition influencing sprint swimming performance in males (64.9%) than in females (24.9%).

Figures 1 and 2 show linear regression for a calculated FINA score (FINA_Score_Predicted) and FINA score performance (FINA_Score_Swim) for male and female sprint swimmers. On the basis of the defined multiple equation models (Table 3, performance prediction), the relations between the calculated y value (FINA_Score_Predicted), the criteria of the sprinter swimming performance (FINA_Score_Swim), and the calculated value of intercept, it is evident that for the male swimmers (Figure 1, intercept—435.143) the line of relation is slanted toward the x-axis, meaning that the model underestimates sprint performance (FINA_Score_Swim) the better the sprint performance. Namely, the better the international male swimmer's sprint performance is, the influence of body composition on the prediction of results is lesser. Compared to international female sprint swimmers, the value of intercept is inferior; that is at the level of 118.114 (Figure 2), indicating that the better the sprint performance, the body composition characteristics will be more proportionally accurate for the sprint swimming prediction.

Table 3. Multivariate Regression Analysis (MRA) statistics results with the multiple equation models of athletes' performance prediction by body composition characteristics—Model Summary.

Model	Dependent Variable	Predictors (Variable, t and *p* Values)	R	R^2	Adj. R^2	SEE	ANOVA	
							F Relation	*p* Value
Male	FINA_Score	PFI (1.58, 0.123), BM (2.28, 0.028), PSMM (2.03, 0.049), BH (−2.20, 0.034), IBC (−1.56, 0.126), SMMI (−1.72, 0.093)	0.662	0.438	0.351	57.48	5.06	0.001
Female	FINA_Score	PFI (2.66, 0.013), BM (2.19, 0.037), BMI (−2.28, 0.030), FFMI (2.50, 0.019), PSMM (2.88, 0.008), IBC (−2.60, 0.015), PBF (3.00, 0.006)	0.895	0.801	0.751	55.99	16.10	0.000
Multiple equation models of athletes' performance prediction by body composition characteristics for male and female swimmers								
Male	$\text{FINA score\_M} = 5884.616 - (\text{BH} \times 65.548) + (\text{BM} \times 74.835) + (\text{PSMM} \times 116.793) - (\text{SMMI} \times 411.608) - (\text{IBC} \times 268.620) + (\text{PFI} \bullet 316.588)$							
Female	$\text{FINA score\_F} = -12241.319 + (\text{BM} \times 6.225) - (\text{BMI} \times 716.712) + (\text{PBF} \times 272.470) + (\text{PSMM} \times 111.570) + (\text{FFMI} \times 1051.171) - (\text{IBC} \times 2253.208) + (\text{PFI} \times 2359.262)$							

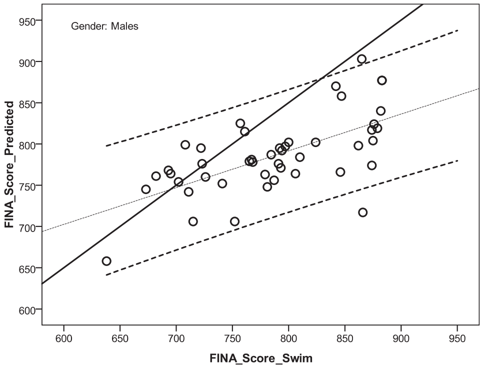

Figure 1. Linear regression for a calculated FINA score and FINA score performance for male sprint swimmers ($y = 435.143 \times \text{FINA\_Score\_Swim}^{0.446}$).

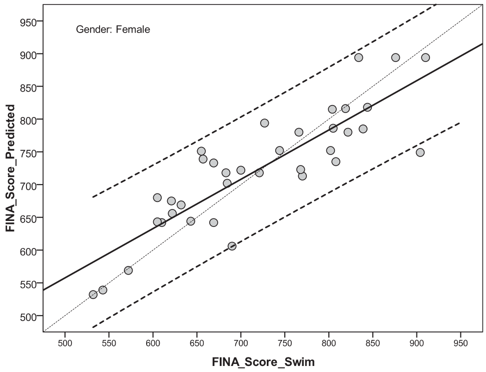

Figure 2. Linear regression for a calculated FINA score and FINA score performance for female sprint swimmers ($y = 181.114 \times \text{FINA\_Score\_Swim}^{0.753}$).

4. Discussion

The results from this cross-selection study confirmed the existence of statistically significant and complex relations between body composition characteristics and performance in international sprint swimmers, both male and female.

Averaged basic anthropo-morphological characteristics (Table 1) indicate that sprint swimmers from this sample are taller, heavier, and larger than male elite swimmers of the past (183.8 cm, 78.4 kg, and 23.21 $kg \cdot m^{-2}$) [35]. Female swimmers are, also, taller, have lower body weight and BMI comparing with the results published previously (171.5 cm, 63.1 kg, and 21.45 $kg \cdot m^{-2}$) [35]. Moreover, the results showed that the international sprint swimmers from the study, both men and women are taller, with less body fat, with lower body mass index, but with a higher level of contractile, i.e., muscle mass, than national level swimmers [30].

This statement is also supported by the results of the correlation (Table 2), on the basis of which it can be claimed that sprint performance of male swimmers have the highest level of statistically significant correlation with variables that define the relative and absolute amount of contractile, i.e., muscle tissue in the body, primarily partialized in relation to the volume of the body, in absolute value, and then in relation to longitudinal characteristics such as: PSMM ($r = 0.353$, $p = 0.016$), SMM ($r = 0.350$, $p = 0.017$), and SMMI ($r = 0.323$, $p = 0.029$). The second set of correlation defined index variables identify quantitative relationship, i.e., balance between fat mass and protein, and body volume and relative fat mass value such as: PFI ($r = 0.392$, $p = 0.007$) and IBC ($r = 0.391$, $p = 0.007$). Identifying the optimal balance between body characteristic as a parameters of lean mass and fat mass is likely to be beneficial and of some importance for maximizing swimming performance [11].

Comparing to the males, different correlation structure was established in female swimmers. The first set represent variables that define the absolute and relative amount of the contractile component in the body in correlation with competitive performance such as: PSMM ($r = 0.732$, $p = 0.000$), SMM ($r = 0.730$, $p = 0.000$), FFM ($r = 0.702$, $p = 0.000$), PFFM ($r = 0.696$, $p = 0.000$). However, the next set of correlations, considering their statistical significance level, is determined in terms of the structure of body fat characteristics such as: PBF ($r = -0.621$, $p = 0.000$), FMI ($r = -0.642$, $p = 0.000$), and BF ($r = -0.566$, $p = 0.000$). Very high statistically significant correlations for index variables were also established, identifying quantitative relationship, i.e., balance between fat mass, and body volume or protein mass: IBC ($r = 0.687$, $p = 0.000$) and PFI ($r = 0.655$, $p = 0.000$). Finally, although not found in the male section, BH, as a basic longitudinal characteristic of the body, has a statistically significant correlation with sprint swimming performance at $r = 0.535$ and $p = 0.001$ (Table 2).

One of the most important papers, which investigates the relation between performance, body composition, and somatotype in competitive collegiate swimmers, was published 25 years ago [14]. At the beginning of the season, the authors have found a statistically significant relation between 100-yard performance and body height ($r = -0.466$, $p < 0.01$), percent of body fat ($r = 0.351$, $p < 0.05$), and fat-free weight ($r = -0.332$, $p < 0.05$) only in female swimmers. At the main period of the season, they found higher correlations for the same variables of performance and body height ($r = -0.766$, $p < 0.001$), fat-free weight ($r = -0.657$, $p < 0.001$), body weight ($r = -0.437$, $p < 0.01$), and the ectomorphic and mesomorphic body type ($r = -0.441$ and 0.392, $p < 0.05$, respectively). These correlations were established only in female swimmers. Authors made a conclusion that the characteristics of body composition and somatotype may serve as performance predictors only in female athletes. The results of our study demonstrated exactly the same structure with almost the same level of correlations considering female sample (Table 2).

Results of Fisher r-to-z transformation demonstrate the existence of a statistically significant body composition variables, which influence more the female performance in comparison with the male performance (Table 2). The greatest differences between male and female characteristics (Table 2) were established between PFFM correlations ($z = 2.74$, $p = 0.006$), PBF ($z = 2.72$, $p = 0.007$) etc. The most pronounced differences were found in four variables: defining the relative values of fat-free, i.e., most likely contractile mass of the body, such as PFFM and PSMM, as a body volume independent variables (averaged *p* difference was 0.011); absolute values for the same variables, FFM and SMM (averaged *p* difference was 0.012); ballast tissue mass variables such as PBF, FMI, and BF (averaged *p* difference was 0.017); and finally, in longitudinal independent variables, defining the amount of fat-free, i.e., most likely contractile mass of the body per body high, such as FFMI and SMMI (averaged *p* difference was 0.031). These results demonstrate a hierarchical structure of gender dependent differences regarding the influence of body structure on sprint swimmers performance.

In general, it has been established (Table 2) that the greatest individual impact on the result of sprint disciplines for female swimmers is represented by body characteristics of the absolute quantity (SMM, r = 0.730, $p = 0.000$) and relative voluminosity (PSMM, r = 0.730, $p = 0.000$) of muscles in the body. Both relations are positive, which imply that for higher FINA point score (better sprint swimming performance) the higher amount muscle mass (absolute and relative) female sprint swimmers need to have.

The indicators of the ratio of ballast (fat) and contractile (protein) mass, which are absolutely related (PFI, r = 0.392, $p = 0.007$), and structurally related (IBC, r = 0.391, $p = 0.007$) have the greatest impact on the result in the sprint has the influence of body structure in males (Table 2).

According to MRA results, it is possible to predict swimming performance in international sprint swimmers with the body composition variables. The models chosen for interpretation were defined according to the criterion of least prediction error. They explained 35.1 and 75.1% of performance relation with standard error of 57.48 and 55.99 FINA score, for male and female swimmers, respectively (Table 3).

Moreover, the results of MRA models demonstrated a complex structure of the variables included in equations. In general, we used 14 variables, six and seven of them were included in equation for male and female athletes, respectively. Four variables were common for both genders (PFI, BM, PSMM, and IBC), and the rest was specific (BH and SMMI for male athletes; BMI, FFMI, and PBF for females' athletes; Table 3).

Strength and power are highly connected with muscle size [21,36]. Thus, an increase in muscle or fat free body mass enables the athletes to produce more muscle force during specific movement efforts, which improve speed, quickness, acceleration, and agility [6,37–39]. Importance of body structure characteristics that strongly contribute to contractile potential of swimmers, regardless of their gender, are determined by following variables: the amount of skeletal muscle mass in the body (SMM, PSMM, and SMMI); the amount of fat-free mass in the body (FFM, PFFM, and FFMI); the structural relations between muscle and fat tissues indexes (PFI and IBC). All of these variables indicate the body potential for strength and power production during sprint swimmers. Strength parameters have been recently proposed as one of the most important specific factors that influence positively swimming performance through the mechanism of increasing stroke pulling force and stroke efficiency mechanisms regardless of age [11,20,37,40–44]. Therefore, the significant improvement of body strength or segmental body strength in swimmers (arms, legs, or trunk) results in a higher maximum peak force per stroke and may influence the sprint speed and turns, or may result in more stable torso during swimming [40,43,45].

Longitudinal anthropometric characteristics such as body height and the length of the upper (arm span) and lower limbs are of primary importance for achieving high results regardless the age of swimmers [13,46,47]. The results of this study have demonstrated that the equation for the prediction of swim performance recognizes basic longitudinal characteristics (BH) as an important factor independent of gender (Table 3). Morales and co-authors [46] established a fundamental fact about the importance of longitudinal characteristics in swimming. In particular, they showed that for the female and male athletes aged 9–22 and involved in 50-m freestyle swimming, the increase in swim velocity is the result of the increase in stroke length and stroke index. Of course, these parameters depend on the efficiency of stroke mechanism (hydrodynamic, energetic, and bio-mechanical dependency). However, longitudinal characteristics of the body represent anatomical and mechanical potential for this phenomenon.

The effect of the body fat variables (BF, PBF, and BFI, Tables 2 and 3) on performance in female swimmers could be explained by body drag characteristics, as well as with active and passive drag efficiency [15,19,48]. Energy in swimming is used for maintaining the body on water surface and generating the muscle force required to overcome water resistance. Generally, swimming speed depends on the interaction of propulsive and resisting forces. Swimming efficiency and speed can improve by increasing propulsive forces and/or minimizing resisting forces that affect the body per se, or body movement at a given speed. The lower body fat probably results in lower body shape drag (frontal area) and skin friction drag, while at the same time, body composition contractile potential, which depends on variables (SMM and FFM...etc.), provides a better propulsion force potential for the faster swimming [36]. Moreover, there are significant evidence that fat reduction contributes to muscular and cardio-respiratory endurance as well as, to the development of speed and agility [11,15,21].

In the sprint swimming, it is necessary to increase stroke frequency to improve the speed of swimming. However, as that increase is not limitless because of neural coordination factors and stroke coordination and because of muscle power limitation, for faster sprint swimming, swimmers adapt their stroke technique pattern, from so-called “S-hape“ to a much more sprint-speed productive, the so-called “I-shaped“ stroke. Using that stroke style, sprint swimmers were able to continue increasing their stroke frequency to gain more speed by using “straight pull“ arm movement pattern [49]. Therefore, compromising the propulsion efficiency to gain extra speed, it seems that particular swimming disciplines are subjected to specific sports evolutionary processes and the search is on for the way to optimize the selection of sprint swimmers, which are consequentlly taller, have less body fat and have more contractile potential.

According to the defined gender performance predictive models (Table 3), body composition elements may represent biological potential for the achievement of better sprint swimming performance. Relation between achieved real performance and estimates by defined models of prediction, showed us that both models underestimate real performance, but much more so in males than in females (intercept for male is 435.143, and for female is 181.114, Figures 1 and 2). Other possible explanations could be connected with a sample structure by stroke, as the majority of swimmers is free-stylers (39.0%), and then breast-strokers (24.4%) and back and fly strokers (by 17.5%, respectively). It was established that, for well trained competitive swimmers, freestyle (front crawl) is the most economic stroke among competitive swimming strokes, followed by the backstroke, the butterfly, and the breaststroke [17,38]. At the same time, swimming forces that can be produced during the 30 s maximal intensity tethered swimming conditions, showed that a breast-stroker can produce more forcefull strokes, than buter-flyers, front crawlers, and back-strokers [43].

The results of the actual study showed us that body composition related to gender specificity characteristics should be considered much more in the future as one of important issues within swimming science [50] along with the known bio-physical determinants related to swimming as: hydrodynamics, kinematics, energetics, and kinetics characteristics [41,49], anthropo-morphological characteristics [13,19,35,47], strength, and power capabilities [20,37,42,45].

This research identified that sprint performance at the male international swimmers level, first and foremost, considering body composition, is associated in male swimmers with specifically balanced presence of proteins and fats (PFI), as well as the balanced ratio between the total body volume and the percentage of fat present in the body (IBC) (Tables 1 and 2). Secondly, the body predisposition to achieve international level results implies a body composition having a high percentage of muscle mass in relation to all three observed parameters: the absolute value (SMM), along with relative value, which is independent of the body volume (PSMM), and body length (SMM). The results have shown that the variables used to define the mass component of the body had a negative but statistically insignificant correlation and, thus, were also without a predictive potential with regards to the sprint-swimmers performance. This is most likely the consequence of the average values of fat percentage in male swimmers being low (PBF = 9.82 ± 3.35 %, Table 1) along with sprint swimmers with higher PBF values having inferior spring performances, and vice versa (Tables 1 and 2).

On the other hand, the body predisposition in female swimmers aiming at international level performance presumes in the first place a high percentage of muscle mass (both for relative and absolute values), and in the second place it presumes high value presence of fat-free body mass (PSMM, SMM, FFM, and PFFM) (Tables 1 and 2).

All the investigated body mass variables, regardless of the specialization method used (relative - PBF, longitudinal independent -FMI, and the absolute -BF), have had a statistically significant and negative correlation with the sprinting performance. In other words, a direct inversely proportional relation has been established between the body mass and the performance of female sprint swimmers, as those with higher PBF values had inferior results, and vice versa (Tables 1 and 2).

Larger multiseason longitudinal cohort study, with body composition measurement in different training periods, would provide new further evidence to assess in-season and off-season specific relations between body characteristics of swimmers and performance, and not only in sprinters, but also in middle and long distance specialists. Moreover, in future research, it is necessary to determine the relationships between body composition and swimming performance in relation to different swimming techniques, which is one of the limiting factors of this study.

5. Conclusions

The current study presents several important and significant correlations between body composition variables and sprint swimming performance in male and female international level sprint swimmers. Generally, quantifying body composition is beneficial for sprint swimmers in order to control the process of training to improve performance. Body composition measurement procedures may be helpful to determine optimal training modalities (methods, volume, and intensity) for continuous improvement of sprint performance.

Considering body composition, sprint-swimming enhanced performance in males is significantly associated with optimal balance between contractile and noncontractile tissue; and with optimally high level of muscle tissue, while for females, optimally high level of muscle tissue with proper low level of fat is primary determinant.

According to MRA results, it could be concluded that it is possible to predict swimming performance in sprint swimmers by body composition variables. Defined models explained 35.1 and 75.1% of the mutual variability of performance, with standard error of 57.48 and 55.99 FINA score, for male and female swimmers, respectively.

This data clearly demonstrates the necessity to monitor body composition characteristics in sprint swimmers, at the international level, even during the preparation training season, as well as during the competition period. This provides a valuable system of collecting information for coaches. Generally speaking, body composition control in international sprint swimmers should be a valuable system of control of the efficiency of body adaptation on training process aimed at optimizing competitive performance potential.

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Article

Effects of Eccentric Single-Leg Decline Squat Exercise on the Morphological and Structural Properties of the Vastus Lateralis and Patellar Tendon

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Abstract: The purpose was to examine the effect of 6-week eccentric single-leg decline squat (SLDSe) training with two technical execution times (3 s or 6 s) on changes related to the structural properties of the vastus lateralis (VL) and patellar tendon (PT). Thirty-six physical active volunteers were randomly divided into three groups: control group (CG, $n = 13$, age = 20.8 ± 1.9 years, no intervention program), experimental group 1 (EG1, $n = 11$, age = 21.6 ± 2.5 years, execution time = 6 s) and experimental group 2 (EG2, $n = 12$, 21.1 ± 1.2 years, execution time = 3 s). Participants completed a 6-week SLDSe training program (80% of 1-RM) three days a week. The structural characteristics of the VL and the PT were measured with ultrasonography before and after 6-week SLDSe training and after 6 weeks of de-training. Our results indicate that EG1 increased ≈21.8% the thickness of the PT and EG2 increased ≈15.7% the thickness of the VL after the 6-week intervention program. EG1 and EG2 showed greater values ($p < 0.05$) of lean mass and lower values ($p < 0.05$) of fat percentage on the thigh after the intervention program. In conclusion, the SLDSe training carried out with the execution time of 6 s had greater effects on the structural and elastic properties of the PT, and the exercise with the execution time of 3 s caused greater structural adaptations in the VL musculature.

Keywords: eccentric exercise; single-leg decline squat; patellar tendon; vastus lateralis

1. Introduction

In recent years, the analysis of the use of eccentric exercises as a prevention and treatment modality for the recovery of injuries, mainly muscle and tendon injuries, has increased in the scientific literature [1,2]. Furthermore, eccentric exercises have been introduced into sports training programs due to the physiological characteristics provided by eccentric contractions [3]. Eccentric training can lead to greater strength gains because it implies a lower energy cost to develop a certain load [4]. In addition, there are several mechanisms by which eccentric exercises can lead to better results than concentric training in hypertrophy [5,6].

The effects of eccentric exercises on the neuromuscular system have been evaluated in different studies [7,8]. Knee extensors are the most frequently studied muscle group due to their clinical importance in human locomotion [9], showing a significant increase in muscle strength and lean mass [10,11]. Long-term eccentric exercise programs are characterized by giving rise to a series of functional adaptations that appear in the muscle. Taken together, these adaptations can have important applications for injured people or for those athletes who want to improve their performance. Since muscle is capable of generating more strength in the eccentric phase of contraction than in the concentric phase [12], one of the goals of eccentric training may be to improve muscle strength. On the other hand, the influence of eccentric exercises on healthy or pathological tendons has been less studied than on muscle tissue.

Several authors [13,14] demonstrated that a training program of 6–12 weeks of duration, performing 2–3 sessions a week of eccentric exercises can provoke enough stimulation to improve muscle function in different types of populations. Previous studies assessed the magnitude of muscle strength retention up to 6 weeks of detraining in subjects with moderate physical activity [15,16], and for these studies, the muscular strength returns to control levels over several weeks of detraining via a reversal of the neuromuscular and hormonal adaptations that occurred during the training phase [17]. However, we have not found any study comparing the effects of single-leg decline squat (SLDS) exercise performed in the eccentric phase (SLDSe) on the knee extensor apparatus and the consequences of 6 weeks of detraining. Therefore, we decided to investigate the effects of the SLDSe with different technical execution times (3 s and 6 s) on the morphological and structural properties of the vastus lateralis (VL) and the patellar tendon (PT), which are very important in the knee extensor apparatus. The purposes of this study were (1) to establish and compare which eccentric technical execution time in the SLDSe (3 s or 6 s) causes greater adaptations in the morphological and structural properties of the VL muscle and PT and on the composition of the thigh; and (2) to assess the effect of six weeks of detraining in the variables analyzed of the VL and PT.

2. Materials and Methods

2.1. Subjects and Inclusion Procedure

Fifty subjects belonging of the University of Castilla-La Mancha were included in the final selection to participate in this study. All of them were tested according to the inclusion and exclusion criteria designed for this investigation. The inclusion criteria were (1) to perform moderate physical activity (3–6 h per week), (2) to be men between 18 and 35 years old, because load response may deteriorate with age (Reeves et al. 2004) and (3) to score > 90 points in the Victorian Institute of Sport Assessment—patellar tendon questionnaire (VISA-P) in its adapted Spanish version (VISA-P-Sp) [18] to rule out symptoms of patellar tendinopathy. Participants were excluded from the study who (1) had had some type of injury to both lower limbs during the 8 weeks prior to start of the study; (2) had performed strength training in the lower body during the 8 weeks prior to start of the study; (3) practiced sports where jumping was a specific action (volleyball, basketball, high jump) more than two hours a week or competitively; (4) consumed supplements aimed at increasing muscle mass and improving strength; and (5) consumed more than 60 mg of caffeine per day (≈1 cup of coffee).

The final sample of the study was composed of 39 participants. All of them participated voluntarily and signed the informed consent before the start of the investigation. The participants were randomly divided into 3 groups: control group (CG) made up of 13 subjects, who did not carry out the intervention program; experimental group 1 (EG1) formed by 13 subjects (2 were lost during the follow-up of the study), who carried out the intervention program for 6 weeks, performing the eccentric repetition of the SLDSe during 6 s; and experimental group 2 (EG2) formed by 13 subjects (1 was lost during the follow-up of the study), who carried out the same eccentric training program as EG1 but eccentric repetition of the SLDSe during 3 s (Figure 1). The sample size was calculated beforehand based on previous research [19], which measured the influence of a 6-week eccentric training program on VL thickness. The minimal number of subjects required to attain a power of 0.9 and a bilateral alpha level of 0.05 was calculated to be 8 participants per group. The descriptive characteristics of the subjects can be seen in Table 1.

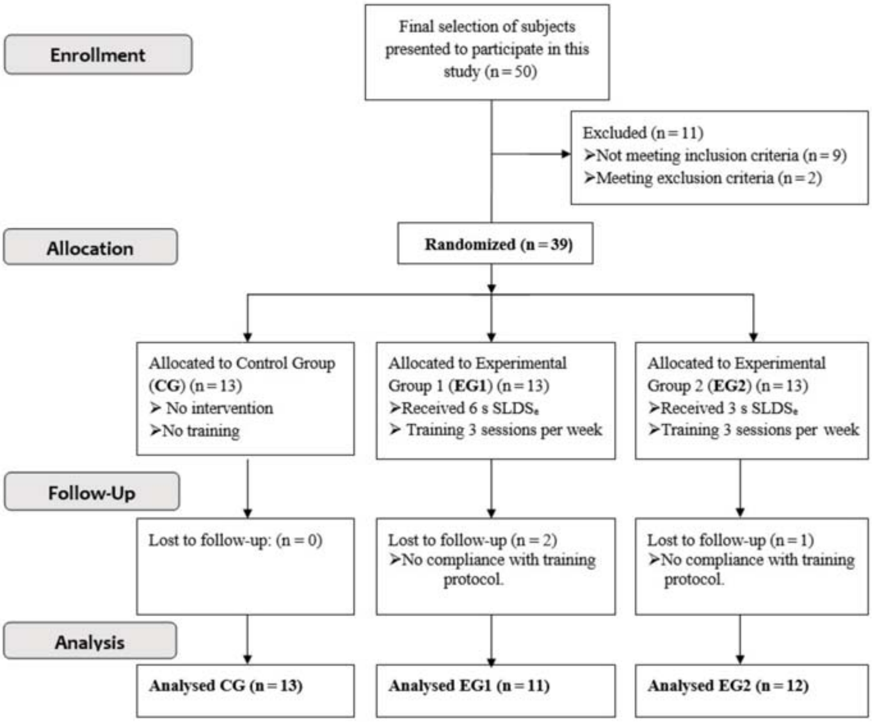

Figure 1. Flow diagram. CG = control group; EG1 = experimental group 1; EG2 = experimental group 2.

Table 1. Descriptive characteristics of the subjects.

	CG (*n* = 13)	EG1 (*n* = 11)	EG2 (*n* = 12)
Age (years)	20.77 ± 1.88	21.55 ± 2.46	21.08 ± 1.24
Weight (kg)	69.84 ± 10.89	71.85 ± 11.82	71.27 ± 8.28
Height (cm)	1.75 ± 0.06	1.76 ± 0.07	1.74 ± 0.07
Fat percentage (%)	18.99 ± 7.02	18.91 ± 05.04	19.54 ± 4.81

Presented as mean ± SD. CG = control group; EG1 = experimental group 1; EG2 = experimental group 2.

This study was approved by the Department of Physical Activity and Sport Sciences of the University of Castilla-La Mancha and by the Clinical Research Ethics Committee of the health area of Toledo (number 62, dated 10 June 2015), according to the principles of the latest version of the Declaration of Helsinki.

2.2. Design and Procedure

Three assessments were carried out with each of the groups studied (CG, EG1, and EG2) in this investigation. The first assessment was carried out before the intervention program began (PRE) (week 0), the second assessment was carried out at the end of the intervention program (POST 1) (week 7), and the third and last assessment was carried out 6 weeks after the intervention ended to evaluate the residual effects after a 6-week non-training process (POST 2). In the non-training process, participants were instructed to continue their normal lives without participating in any additional training programs (week 13) (Figure 2).

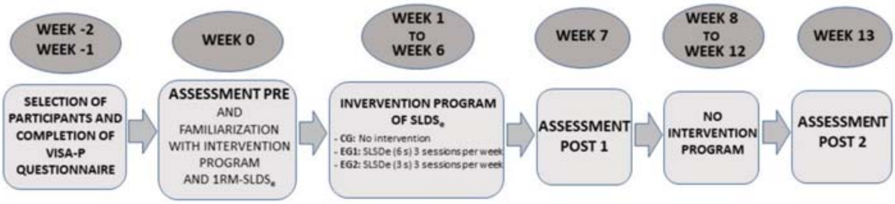

Figure 2. Design of the investigation.

The first session was for data collection (assessment PRE) of the studied variables of body composition by densitometry and of the morphological and elastic variables by ultrasound and sonoelastography so that the sessions of familiarization did not influence these variables. The familiarization sessions and before assessment PRE were to eliminate any learning effects and to inform subjects about the eccentric training program (SLDSe), and 2 days later, the calculation of 1-RM of eccentric exercise (SLDSe) for EG1 and for EG2 in the dominant limb (leg with which the subjects kicked a ball) was carried out in order to determine the training load. The two experimental groups (EG1 and EG2) performed each of the eccentric repetitions in a time of 6 s (EG1) or in a time of 3 s (EG2), resting 6 s between each repetition and reaching up to 90° of knee flexion, which was evaluated with a manual goniometer placed on the joint that followed the bony lines of the femur and fibula. To do this, the participants performed a series of 5 eccentric repetitions of increasing intensity, starting with the weight recorded on the last day of the familiarization pre-testing. With a 2 min rest between each set, the load was increased to establish the 5-RM. In the event that the subject did not maintain the execution speed or did not reach 90° of knee flexion, the repetition was considered null. In the event that there was muscle failure in which the participants could not perform the repetition due to fatigue or inability to tolerate the load, the weight lifted in the last series of 5-RM performed was recorded. If after performing 5 sets it was not possible to obtain the 5-RM, the test was canceled and had to be repeated after 48 h. In this way, we avoided the effect of fatigue on the test result. When the participants reached 90° knee flexion during the exercise, two researchers were placed on each side of the multipower bar, lifted the weight toward the initial position, and the participant returned to the beginning of the exercise using bipodal support. In this way, we reduced the influence of the concentric phase force of the anterior thigh muscles. The time of execution of the eccentric action and the rest time between each repetition were controlled by a metronome (www.webmetronome.net) that emitted acoustic signals indicating the start and end of each contraction. The attainment of 90° of knee flexion was controlled by the principal investigator, placing the goniometer on the knee and determining the end of the repetition.

The 5-RM eccentric test of SLDSe (the same eccentric exercise to be performed in the intervention program) was carried out on a multipower machine (Technogym, Gambettola, Italy). The weight of the 5-RM for each subject was noted, and the calculation of the 1-RM was performed indirectly with the following formula $predicted\ 1 - RM = \frac{Weight\ Lifted}{1.0278 - (0.0278 \times 5)}$ [20]. In the 6 weeks of the intervention program, the 5-RM eccentric test of SLDSe was repeated by EG1 and EG2 every 2 weeks with the specific training execution time (3 or 6 s) in the dominant limb in order to update the training loads.

The intervention program for both groups (EG1 and EG2) lasted 6 weeks. Every week, 3 training sessions were carried out separated by at least 48 h. All sessions were carried out on the same multipower machine and were supervised by the main researcher and two collaborators. In each of the sessions, a 10 min warm-up was performed on a cycle ergometer at an intensity of 100 W and at a cadence of 80–90 rpm (Wattbike cycle-ergometer, Wattbike Pro, Nottingham, UK). After the warm-up, and for both experimental groups, 3 series of 8 repetitions of the SLDSe eccentric exercise described by Purdam, Jonsson [21] were executed. The rest between repetitions was 6 s and between series was 2 min, and the intensity was 80% of the 1-RM, which was calculated as mentioned above for each group in their specific working technical execution time.

2.3. Outcome Measures

An experienced sports traumatologist with extensive musculoskeletal ultrasound training (FJ) carried out all ultrasound examinations on the dominant leg. Morphological examinations of the VL muscle and PT were performed with a Logiq® S8 ultrasound (GE Healthcare, Milwaukee, WI, USA) with a 10 MHz linear probe (ML6-15-D; General Electric Healthcare system). In addition, the elastography index (EI) of the PT was recorded with a probe to measure sonoelastography connected to the same ultrasound machine with which measurements of morphological characteristics were performed (Logiq® S8). Muscle architecture measurements of the VL were performed at two points: 50% of the total thigh length [22] and 4 cm from the distal myotendinous junction. The ultrasound probe was aligned with the fascicle direction to measure thickness and pennation angle of the VL. The assessment of muscle thickness, pennation angle, and fascicle length were performed as described in previous studies [23–25].

The PT was scanned in the sagittal and axial planes, taking care to avoid anisotropy. The thickness and the sonoelastography of the tendon were measured at 50% of the tendon length (distance between the lower pole of the patella to the deep distal insertion in the tibia). Sonoelastography was performed by applying light repetitive compression with the hand-held transducer. The elastogram appeared within a rectangular region of interest (ROI) as a translucent color-coded real-time image superimposed on the B-mode image [26]. The color code indicated the strain of the tissues within the ROI, where red corresponded to soft elasticity, green and yellow indicated medium elasticity, and blue indicated hard elasticity. The B-mode image and elastogram were displayed side-by-side on the screen and the graph that appears on the screen standardized the amount and uniformity of compression. The best cine image derived from at least three compression–relaxation cycles was used for the assessment of the EI [26]. A higher value of EI Is related to a higher stiffness level.

The measurements were always taken with the subjects lying down. The VL and PT were scanned with the subject in a supine position and the knee flexed at 15° (0° corresponding to full extension of the knee) with a pillow underneath [27]. All images were analyzed with Image analysis software (Image J, 1.47v, National Institute of Health, MD, USA) to measure fascicle length, pennation angle, muscle thickness, and tendon thickness. The reliability of this ultrasound technique for measurements of human muscle architecture has been previously studied with intraclass correlation coefficients that ranged from 0.92 to 0.99 [28].

Body composition (fat mass, lean mass on the thigh, and fat mass on the thigh) was assessed by dual emission X-ray absorptiometry (DXA, GE Healthcare, Lunar, Diegem, Belgium) with participants in a supine position as previously stated [29]. Thigh-specific analyses were performed as described by Alegre et al. [30], who found a high reliability of this technique with a coefficient of variation that ranged from 0.4% to 3.9%.

2.4. Statistical Analysis

The statistical analysis was performed with IBM SPSS Statistics 23.0 (SPSS, Chicago, IL, USA). All data were expressed as mean ± standard deviation. The data were tested for normality with the Shapiro–Wilk test. Since the assumption of normality (all variables $p > 0.05$) was verified, a two-way (3×3) repeated measures ANOVA was used to determine the main effects of the two training interventions upon measures of PT variables, VL variables, and the composition of the thigh parameters. One factor was the group (CG, EG1, and EG2) and the other was the timeline (PRE-, POST-1 and POST-2). Effect size statistics were used to quantify the magnitude of the difference in pairwise comparisons, according to the formula proposed by Cohen [31]. The magnitude of the effect size was interpreted using the scale of Cohen [31]: an effect size lower than 0.2 was considered as small, an effect size around 0.5 was considered as medium, and an effect size over 0.8 was considered as large. A probability level of $p < 0.05$ was defined as statistically significant.

3. Results

3.1. Morphological and Elastic Properties of PT

The values recorded for the thickness and EI in PT are shown in Figure 3. Significant time x group interaction (F = 5.28; $p = 0.001$) but no inter-group (CG, EG1 and EG2) differences were found in the PT thickness. The PT thickness was 0.08 ± 0.05 cm (IC 95%, from 0.04 to 0.11 cm, $p < 0.001$, ES = 1.3) greater in EG1 in POST-1 compared to PRE and 0.06 ± 0.07 cm (IC 95%, from 0.01 to 0.11 cm, $p = 0.011$, ES = 0.9) lower in POST-2 compared to POST-1. Significant time x group interaction (F = 3.84; $p = 0.008$) were found in the EI. The EI values of EG1 were greater ($p < 0.05$) in POST-2 compared to both PRE and POST-1. Moreover, EG1 showed greater values ($p < 0.05$) than EG2 and CG in POST-2.

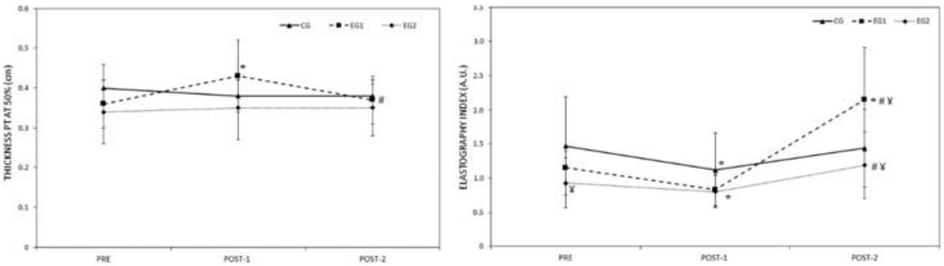

Figure 3. Morphological and elastic properties of patellar tendon (PT) in response to six weeks of eccentric single leg decline squat exercise training (Post-1) with two technical execution times (EG1 = 6 s and EG2 = 3 s) and 6-week follow-up of detraining (Post-2). * = $p < 0.05$ from pre evaluation; # = $p < 0.05$ from post-1 evaluation; ¥ = $p < 0.05$ from CG; PT = patellar tendon; CG = control group; EG1 = experimental group 1; EG2 = experimental group 2.

3.2. Vastus Lateralis Structure

The values obtained for the pennation angle, thickness, and fascicle length in VL are shown in Table 2. No significant differences were found among groups (CG, EG1, and EG2) in any of the assessments (PRE, POST-1, and POST-2) for any of the variables studied except in the pennation angle and the thickness of the VL distal region in POST-1 between EG1 and CG. Differences were observed intra-groups. The pennation angle of the VL distal region in EG1 was 2.25 ± 2.19° (IC 95%, from 0.4 to 4.1°, $p = 0.021$, ES = 0.9) greater in POST-1 compared to PRE, and 3.0° ± 2.19° (IC 95%, from 0.6 to 5.5°, $p = 0.018$, ES = 1.3) lower in POST-2 compared to POST-1. In addition, the thickness in the distal region was 0.22 ± 0.07 cm (IC 95%, from 0.03 to 0.41 cm, $p = 0.026$, ES = 0.9) greater in EG2 in POST-1 compared to PRE. In the same way, EG2 showed an increase of 0.22 ± 0.18 cm (IC 95%, from 0.01 to 0.44 cm, $p = 0.042$, ES = 0.6) in the thickness at 50% after the training and a decrease of 0.17 ± 0.21 cm (IC 95%, from 0.04 to 0.29 cm, $p = 0.013$, ES = 0.5) after the 6 weeks of follow-up without training.

No significant differences were found in the pennation angle at 50% and in the fascicle length at 50% and in the distal region between PRE, POST-1, and POST-2 in any of the groups. No significant differences were found in CG in any of the variables analyzed in the VL between PRE, POST-1, and POST-2.

Table 2. Morphological properties of VL in response to six weeks of eccentric single-leg decline squat exercise training (Post-1) two technical execution times (EG1 = 6 s and EG2 = 3 s) and 6-week follow-up of detraining (Post-2).

	PRE	POST-1	POST-2	*p*-Value	Timeline Effect		Time × Group Interaction	
					F	*p*-Value	F	*p*-Value
Pennation angle VL distal (°)								
CG	15.92 ± 2.69	16.15 ± 3.29	17.15 ± 2.19	0.437	3.94	0.032	3.83	0.034
EG1	17.38 ± 2.39	19.63 ± 2.88 *¥	16.63± 1.77 #	0.025				
EG2	15.50 ± 3.24	17.10 ± 3.96	17.40 ± 2.98	0.108				
Thickness VL distal (cm)								
CG	1.53 ± 0.32	1.39 ± 0.33	1.48 ± 0.27	0.267	1.26	0.299	5.01	0.014
EG1	1.61 ± 0.45	1.79 ± 0.41 ¥	1.68 ± 0.40	0.244				
EG2	1.42 ± 0.24	1.63 ± 0.26 *	1.55 ± 0.36	0.088				
Fascicle length VL distal (cm)								
CG	5.65 ± 1.14	5.45 ± 0.99	5.30 ± 0.63	0.716	0.47	0.628	0.06	0.993
EG1	5.15 ± 2.47	5.10 ± 0.56	4.89 ± 0.85	0.860				
EG2	5.51 ± 1.19	5.57 ± 1.35	5.39 ± 0.90	0.877				
Pennation angle VL 50% (°)								
CG	15.46 ± 3.89	15.62 ± 3.69	15.46 ± 3.13	0.954	2.53	0.099	0.60	0.662
EG1	14.75 ± 2.76	16.38 ± 2.62	16.63 ± 2.07	0.168				
EG2	14.20 ± 2.97	15.40 ± 2.46	15.10 ± 2.51	0.297				
Thickness VL 50% (cm)								
CG	2.09 ± 0.39	2.08 ± 0.33	2.07 ± 0.33	0.961	4.34	0.023	2.72	0.083
EG1	1.90 ± 0.38	2.13 ± 0.36	2.08 ± 1.82	0.164				
EG2	2.02 ± 0.71	2.25 ± 0.30 *	2.08 ± 0.32 #	0.016				
Fascicle length VL 50% (cm)								
CG	8.05 ± 1.74	7.99 ± 1.72	7.98 ± 1.82	0.997	0.78	0.470	0.35	0.843
EG1	7.04 ± 3.56	8.01 ± 1.85	7.62 ± 1.01	0.353				
EG2	7.84 ± 03.40	8.33 ± 1.79	7.92 ± 1.26	0.455				

* = $p < 0.05$ from pre-evaluation; # = $p < 0.05$ from post-1 evaluation; ¥ = $p < 0.05$ from CG; VL = vastus lateralis; CG = control group; EG1 = experimental group 1; EG2 = experimental group 2.

3.3. Composition of the Thigh

The values recorded for the lean mass and the percentage of fat mass of the thigh are shown in Table 3. No significant differences were found in inter-group comparisons (CG, EG1, and EG2) for any of the variables studied. No significant differences were found in CG in any of the variables analyzed in the composition of the thigh between PRE, POST-1, and POST-2. Significant increases ($p < 0.05$) were found after training in lean mass in both experimental groups (EG1: diff = 0.30 ± 0.38 kg; CI 95%: from 85.6 to 520.1 kg, $p = 0.004$, ES = 0.3 and EG2: diff = 0.36 ± 0.27 kg; CI 95%: from 155.4 to 571.4 kg, $p = 0.001$, ES = 0.5). The fat percentage in EG1 and EG2 was lower in POST-1 compared to PRE (EG1: diff = 0.85 ± 0.95%; CI 95%: from 0.1 to 1.6%, $p = 0.016$, ES = 0.1 and EG2: diff = 1.02 ± 1.00%; CI 95%: from 0.3 to 1.7%, $p = 0.002$, ES = 0.2) and for both groups was greater (EG1: $p = 0.019$ and EG2: $p = 0.015$) in POST-2 compared to POST-1.

Table 3. Composition of the thigh in response to six weeks of eccentric single-leg decline squat exercise training (Post-1) with two technical execution times (EG1 = 6 s and EG2 = 3 s) and 6-week follow-up of detraining (Post-2).

	PRE	POST-1	POST-2	*p*-Value	Timeline Effect		Time × Group Interaction	
					F	*p*-Value	F	*p*-Value
Lean mass thigh (kg)								
CG	7.01 ± 1.16	7.03 ± 1.17	7.16 ± 1.21	0.164	10.75	$p < 0.001$	2.89	0.029
EG1	7.43 ± 1.19	7.73 ± 1.21 *	7.51 ± 1.20 #	0.005				
EG2	7.19 ± 0.77	7.55 ± 0.88 *	7.36 ± 0.74	0.001				
Fat percentage thigh (%)								
CG	20.36 ± 8.44	20.36 ± 8.06	20.57 ± 7.95	0.822	17.90	$p < 0.001$	2.88	0.030
EG1	19.30 ± 5.80	18.45 ± 6.21 *	19.42± 6.10 #	$p < 0.001$				
EG2	19.89 ± 5.55	18.88 ± 4.78 *	19.83 ± 4.80 #	$p < 0.001$				

* = $p < 0.05$ from pre evaluation; # = $p < 0.05$ from post-1 evaluation; CG = control group; EG1 = experimental group 1; EG2 = experimental group 2.

4. Discussion

The aim of this study was to determine the effect of the SLDSe with different technical execution times (3 s and 6 s) on the morphological and structural properties of the VL and PT and on the composition of the thigh between EG1 (executing the exercise in a time of 6 s), EG2 (executing the exercise in a time of 3 s), and CG without intervention. The results demonstrated greater values of thickness in PT, pennation angle, thickness, and fascicle length in the distal region of the VL and lean mass on the thigh in EG1 after the 6-week intervention program. EG1 and EG2 showed lower values of fat mass on the thigh after the intervention program. These results suggest that 6 weeks with 3 sessions per week with 3 series of 8 repetitions at 80% of 1-RM of the SLDSe cause an increase in the thickness of the main muscles and tendons of the knee extensor apparatus (VL and PT), considering that after 6 weeks without training, these adaptations tended to return to the initial values measured before the intervention.

Although the tendon is considered an avascular structure, it has been shown to respond to external mechanical loads by altering its biomechanical properties (Young's modulus) and/or morphological characteristics (thickness and CSA) [27,32]. Present data showed a PT hypertrophic response associated with SLDSe performed in 6s evidenced by the greater tendon thickness with an increase of 21.8 ± 15.0% at the end of the 6 weeks of the intervention program. These data coincide with previous investigations that have managed to hypertrophy the PT, although they used at least 12 weeks of intervention programs through eccentric exercise [33,34]. Our results have important clinical relevance because we demonstrate that 6 weeks of SLDSe training performed more slowly (6 s) are enough to increase the thickness of the PT. This circumstance may be due to the fact that the time in which the tendon is subjected to external overload is greater in the repetitions performed more slowly and stimulates the synthesis process of collagen more, causing an increase in the thickness of the tendon [35]. Eccentric exercise has been one of the most widely used conservative treatment modalities for the recovery of tendinopathies in general [36] and PT tendinopathy in particular [34]. The SLDSe performed in our study (carried out on a 25° incline) offered more favorable results than the single-leg squat performed on a flat surface [36] because the load to which the PT is subjected is greater when the SLDSe is performed on a 25° incline than on a flat surface [37].

Sonoelastography has proven to be a reliable and reproducible technique in the exploration of the stiffness index of healthy PT [38]. The data show that after the 6 weeks without training, the EI increased 91.2 ± 71.4% in EG1 with respect to baseline, which seems to indicate that eccentric exercise causes different long-term adaptations in the stiffness of the PT to isometric exercise, since Kubo et al. [39] found a reduction in the stiffness of the tendon two months after completing a training program using isometric contractions. A higher value in the EI of the PT, such as that found in our study after six weeks without training, indicates greater stiffness, which may benefit the rate of force development [40], and it has been associated with better performance in agility tests, pace changes, sports with continuous stretch–shortening cycles and speed/sprint tests [41]. However, at the same time, the risk of muscle injury in the tendons with these characteristics is higher because the high stiffness makes the tendon absorb less energy and increases the forces that are generated in muscle [42].

The VL is a muscle that has a great capacity for structural adaptation to eccentric strength at the distal level [43]. Other studies have shown adaptations of the architecture of the extensor muscles of the knee after four weeks of training with eccentric exercises [44]. The results in this study have shown that the intervention program increased the thickness of the VL in EG2 (group that trained the SLDSe during 3 s) by 15.7 ± 5.9% at the distal level, and by 12.4 ± 11.7% measured at 50% of the thigh length. In addition, we also found a trend to increase VL thickness at the distal level ($p = 0.097$) and 50% of the thigh length ($p = 0.059$) in EG1 in this group to an increase of 13.4 ± 13.2% in the distal pennation angle and a tendency to increase in EG2 ($p = 0.062$). These results indicate that the mechanical stimuli induced by eccentric exercise of high intensity (80% of the 1-RM) and performed with a technical execution time of 3s may be a fundamental mechanism for VL hypertrophy. Therefore, the speed of execution of the eccentric repetition is a determining factor to achieve adaptations in the mentioned variables of the VL [45]. The results of this study have shown that regardless of the technical execution time, after six weeks of detraining, the adaptations were lost both at the tendon and muscle levels. These results are in line with other studies that claim that the muscular strength returns to control levels over several weeks of detraining [15,16].

This study showed that regardless of technical execution time of the SLDSe at the end of the intervention program, both experimental groups increased lean thigh mass (EG1 ≈4.2% and EG2 ≈5.0%). These results are in line with other research that has stated that eccentric exercise is the most effective training mode for promoting muscle growth [46]. In addition, eccentric exercise causes greater gains in muscle mass than concentric exercise because it produces a series of histochemical and metabolic substrates that induce hypertrophy [47]. Moreover, the fat mass in the thigh decreased in both experimental groups at the end of the 6-week intervention program, which indicates the importance of strength training to improve the lipid profile [48]. It was also shown that after six weeks without training, the muscle mass and fat mass values of the thigh tended to return to their initial levels.

There were some limitations to this study that deserve attention. Our data are best extrapolated to young, healthy, and physical active males. Although this population is arguably the most likely to use eccentric training, other populations such as older individuals, females, and rehabilitation patients may or may not respond in the same manner as our study cohort. We used the eccentric single-leg declined squat exercise, and some individuals may perceive this as a less practical exercise and may not extrapolate well to other sports-related activities. The eccentric training and vastus lateralis and patellar tendon adaptations only were performed on the dominant leg of the participants; the reported results may not be similar in the non-dominant leg. Finally, despite the fact that subjects have been instructed not to perform strength training outside the study protocol, their daily physical activity may affect the study results.

5. Conclusions

In conclusion our results indicate that the eccentric exercise of the SLDSe performed at high intensities (80% of the 1-RM) and carried out with the execution time of 6 s caused an increase of ≈21.8% in the thickness of the PT and carried out with the execution time of 3 s caused an increase of ≈15.7% in the thickness of the VL. The slower eccentric exercise of SLDSe had greater effects on the structural and elastic properties of the PT and, conversely, when it was performed more quickly, it caused greater morphological and structural adaptations in the VL musculature. In addition, it has been shown that regardless of the technical execution time, after six weeks without training, adaptations were lost both at the tendon and muscle levels. These findings suggest that SLDSe is effective for both PT and VL hypertrophy as well as helping to lose fat mass in the thigh.

As practical application, the knowledge of the effects of an eccentric training program lasting 6 weeks with different execution times on the morphological and structural properties of VL and PT, which are two of the main protagonists of the knee extensor apparatus, is essential both for the treatment modalities for recovery from injuries and for improving the performance of athletes. Current results suggest that 6 weeks of eccentric training on the dominant leg of the SLDSe exercise produces increases in VL and PT thickness and thigh muscle mass, which could be used when the goal is to improve muscle strength at the performance level or to produce muscle and tendon adaptations in recovery from injuries.

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The Effect of Balance and Sand Training on Postural Control in Elite Beach Volleyball Players

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Abstract: The aim of this work was to evaluate the effectiveness of a 12-week-long balance training program on the postural control of elite male beach volleyball players and the effect on balance when swapping to specific sports training in the sand in the following 12 weeks. Six elite players were tested before and after the balance training program and also 12 weeks after the balance training had finished. To this aim, a pressure platform was used to collect the following center of pressure parameters: path length, speed, mean position, and root-mean-square amplitude in the medial-lateral and anteroposterior planes. Romberg quotients for the center of pressure parameters were also calculated. The results of the present study showed better static postural control after specific balance training: smaller path length and speed under open eyes condition in dominant ($p = 0.015$; $p = 0.009$, respectively) and non-dominant monopedal stances ($p = 0.005$; $p = 0.004$, respectively). Contrastingly, 12 weeks after the balance training program, the path length and speed values under open eyes condition in bipedal stance increased significantly ($p = 0.045$; $p = 0.004$, respectively) for sand training. According to our results, balance training is effective to achieve positive balance test scores. It is speculated, and yet to be proven, that sand training could be effective to improve dynamic and open eyes postural control during beach volleyball practice. In beach volleyball players, a balance training program is effective to develop static balance but the effect of ecological sand training on dynamic performance deserves specific investigation.

Keywords: athletes; posturography; core; stability; proprioception

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1. Introduction

Postural control is a complex function of the central nervous system for detecting sensory stimuli, interpreting information, and responding appropriately in order to maintain an upright position [1,2]. The development of postural control requires integration between sensory systems, i.e., vestibular, visual, and somatosensory systems [3,4]. The two main functional objectives of postural control are postural orientation and postural equilibrium [5].

Postural control depends not only on the health state but also on training capacity through variations in the tone of postural muscles [6,7]. Thus, physical training allows for the acquisition of new skills and strategies for postural control improvements [8]. Balance training is widely used in rehabilitation, sports, and injury prevention programs [9]. Likewise, balance training has been shown to be effective for the improvement of postural control in different populations: active and inactive young people [10,11], athletes [12–14], and the elderly [15,16].

In the sports field, an improvement in postural control is associated with better performance [17], mainly due to an increase in the efficiency of sport-specific actions [18], better force production [19], and reduction of injury risk [20]. Although balance training seems to be effective in improving performance only in specific trained tasks [21], many mechanisms related to the plasticity of postural control and the sport experience are not yet understood [18,22].

There are limited data on the benefits of balance training in elite athletes [18] especially when athletes play on unstable surfaces such as sandy courts, where physiological and biomechanical characteristics differ from firm ground [23–25]. Despite studies addressing balance training in volleyball [12,26–28], no study related to beach volleyball has been found. Therefore, it would be interesting to explore the effect of balance training in a discipline played on unstable surfaces [29], where demands of motor control are higher than on stable surfaces [30]. In the same way, it would be useful to know if sand training can be a substitute for classical balance training in elite beach athletes with advanced motor skills to maintain balance.

The present study aimed to investigate whether a balance training program could improve postural control in male elite beach volleyball players. It was hypothesized that stabilometric variables would be enhanced as a consequence of the balance training program, mainly decreasing total excursion (TE) and speed, and that these variables would return to baseline values after the replacement of balance training with specific sports training. To this end, the effect of balance training, elimination, and replacement for specific sports training in the sand were studied with regards to postural control.

2. Materials and Methods

2.1. Participants

We studied three elite international beach volleyball teams, six players (age 23.6 ± 3.3 years, height 188.2 ± 7.9 cm, body mass 77.7 ± 13.1 kg, BMI 22.3 ± 2.6 kg/m^2, body fat 8.9 ± 2.5% (measured with a Tanita BC-545N body composition analyzer), professional experience 4.9 ± 1.8 years, predominance of arm and leg, right and left, 6/0). Three players played in the blocker position and the other three played in the defense position. The requirements to participate in the present study were to have competed in the national winter league and the National Circuit “Madison Beach Volley Tour” for three years, to have competed during at least the last year in international championships, and previous experience with balance training. A power analysis using G*3-Power (Heinrich Heine Universität, Düsseldorf, Germany) [31] indicated that this sample size provided 80% power to detect effects of $d = 0.6$ (medium effect) in a repeated measurement test with $\alpha = 0.05$. The Ethics Committee at the University of Alicante gave institutional approval to this study, in accordance with the Declaration of Helsinki (UA-2018-12-19), with the consent of athletes duly informed of the procedure and parts of the study.

2.2. Measures

A baropodometric platform (FreeMed, Rome, Italy) was used for the stabilometric analyses, with an active surface of 400 × 400 mm, 8 mm thickness and a sample frequency of 100 Hz. The test was carried out three times with two minutes rest between trials. Stabilometric parameters were assessed three times and the average value was registered. The reference stabilometric values to express deviation of the center of pressure (CoP) were: path length, defined as the length of the total distance of the CoP over the course of the trial duration; speed, defined as total distance traveled by the CoP over time. It is suggested that increases in TE represent a decreased ability by the postural control system to maintain balance. Similarly, increases in CoP velocity could indicate a decreased ability to control posture [32]. Four displacement parameters were also measured regarding the average absolute displacements around the mean CoP: medial-lateral plane, anteroposterior plane, root-mean-square amplitude in medial-lateral plane, and root-mean-square amplitude in anteroposterior plane. The Romberg test was measured for 30 s in the following positions: bipedal stance with eyes open, bipedal stance with eyes closed, dominant monopedal stance with eyes open, dominant monopedal stance with eyes closed, non-dominant monopedal stance with eyes open, and non-dominant monopedal stance with eyes closed [14,33,34]. Traditional Romberg quotients for the CoP parameters were also calculated (eyes closed/eyes open) [35] as an indicator of proprioceptive and visual field contribution to postural stability [36,37]. A Romberg quotient higher than 1.0 denotes a greater postural sway during eyes closed condition [35]. The test order (positions and deprivation conditions) were randomized.

Data collection was initiated after the players were stable in an erect position on the baropodometric platform. The players remained barefoot and positioned with the feet facing forward. The distance between feet was considered according to the hip width. The arms remained relaxed and parallel to the body and the head faced forward. The players were asked to remain in this position during the evaluation process.

2.3. Design and Procedures

An alternating treatment design was used for the present study to explore the alternation of two interventions (balance and sand training) with different proprioceptive stimuli. In this between-series design, the analysis was focused on the comparison of treatment condition outputs [38]. All measurements were taken during the in-season phase. During the pre-season phase (6 weeks) the players had 5 weekly sessions in the sand and 5 weekly sessions in the gym. The volume and intensity of training sessions increased over the weeks. No specific balance training was carried out in this phase.

The competition phase schedule was as follows: 6 weekly sessions in the sand with an approximate duration of 120 min each. Of these, days 1, 3, 5, and 6 were completely devoted to technical and tactical training. The first hour of day 2 and day 4 was devoted to specific physical training and the second hour was also devoted to technical and tactical training. In the same way, there were also 4 weekly sessions of training in the gym with an approximate duration of 120 min each.

The intervention began 3 weeks after the competition phase has started, so that the players could adjust to training. In the first week of intervention, all players underwent a postural control assessment, for the balance baseline level (Test 1 in Figure 1). Then, participants took part in a balance training program over 12 weeks with 48 total sessions (4 sessions per week, 2 sand sessions and 2 gym sessions, 20 min per session). During this first period, the players also completed 6 weekly sessions in the sand with an approximate duration of 120 min each. Of these, 2 weekly sessions had balance training. The first 20 min of the session after the warm-up was devoted to balance training (40 min/week). In the same way, there were also 4 weekly sessions of training in the gym. In two of them, the first 20 min were devoted to balance training (40 min/week). At the end of the intervention, the second evaluation of postural control was carried out (Test 2).

In the 12 weeks following the second period, the training volume (weekly sessions of sand and gym) was the same as in the first period. The balance training was eliminated both in the sand and in the gym and was replaced by specific physical sand training. This training consisted of a selection of similar exercises to the real game to maintain the same work volume and intensity in the sessions as in the first intervention.

Finally, a third measurement (Test 3) was made to compare the data with the two previous tests to explore if the specific work in the sand could substitute balance training with regards to the improvement of the stability of the athletes.

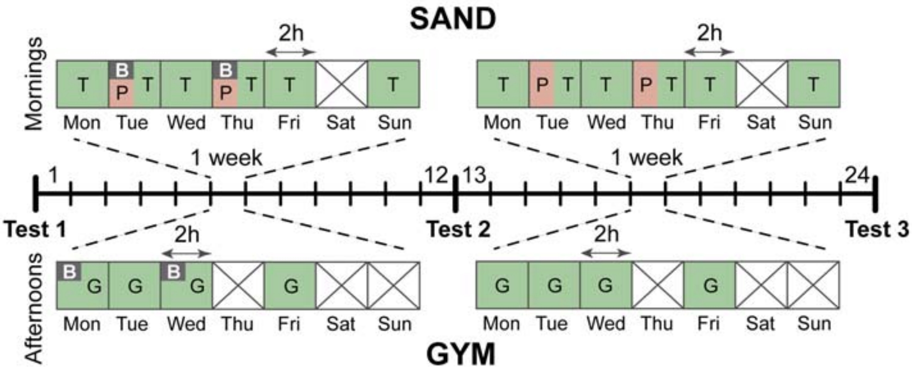

Figure 1. Experimental procedure showing weekly sessions for 24 weeks. Test 1: baseline; Test 2: post-balance training; Test 3: post-sand training. T: technical and tactical training, B: balance training, P: physical training, G: gym training. Days without training are displayed with crossed squares.

2.4. Balance Training

The conventional warm-up consisted of dynamic stretching and moderate aerobic exercises before the regular practice sessions. The time spent to perform balance training did not include the warm-up. The coach conducted the sessions and gave feedback on exercise techniques. Each 20 min session consisted of circuit training with 10 exercises (30 s work/30 s rest between exercises) and 1 min rests between circuits (two laps). Based on previous studies [17,39], the balance training consisted of performing static and dynamic exercises in which the participant's equilibrium was more compromised and the exercises gradually progressed in complexity. In some exercises, the vision of the subject was totally or partially limited, so it was a challenge mainly for the core zone. The exercises were performed in monopedal and bipedal stances and auxiliary materials such as scales coordination, medicine balls, or unstable materials were used to create perturbations. The set of 10 exercises was as follows: bilateral squat on a BOSU ball (one for each leg) and two-hand bump using the wall; single-leg standing on inflated disk progressing to closed eyes execution; lateral medicine ball throwing on a BOSU ball (one for each leg); supine straight leg bridge on a Swiss ball; coordination scale and single leg squat; banded triplanar toe taps progressing to closed eyes execution; paloff press with a slight rotation progressing to monopedal stance; plank with elbows on a Swiss ball progressing to closed eyes execution; on a BOSU ball (one for each leg) the player used the Swiss ball to knock the beach volleyball ball to the coach; attack reception while standing with one or both legs on a BOSU ball; lunge on foam surface progressing to closed eyes execution. The main goal was to make the exercises as similar as possible to the real game actions [40], so that they could be transferred to beach volleyball.

2.5. Statistical Analysis

Descriptive statistics (means and standard deviations), the distribution of normality (Kolgomorov–Smirnov and Lilliefors test), and Levene homogeneity tests were calculated for each stabilometric value. A repeated-measures ANOVA was conducted to compare postural control variables between baseline level, post-balance training, and post sand with post hoc comparisons using the Bonferroni test. A value of $p < 0.05$ was used to identify statistically significant differences. The percentage variation was calculated using the following formula: (Post-Test − Pre-Test)/Pre-Test × 100. Cohen's d was used as a measure of the effect size of differences between tests and interpreted according to Cohen's thresholds as small ($0.00 \leq 0.49$), medium ($0.50 \leq 0.79$), and large (≥ 0.80) [41]. Analyses were performed using the Statistical Package for Social Sciences v.22 (IBM, Armonk, NY, USA).

3. Results

The values of the parameters related to postural control in static bipedal stance are shown in Table 1. The results when baseline level and post-balance training were compared in open eyes condition showed an improvement in the main indicators of postural control after the balance training intervention: 29.4% path length ($p = 0.240$) and 28.7% speed ($p = 0.241$) decreases. The improvements of these variables meant that the participants were more stable and efficient in controlling their posture. Significant worsening for the same variables, i.e., 38.6% path length ($p = 0.045$) and 38.7% speed ($p = 0.004$) increases, resulted between post-balance training and post-sand training, once the replacement training in sand had finished. Similar results were found in closed eyes condition although they were not statistically significant, except for root-mean-square amplitude in the antero-posterior plane ($p = 0.038$) when baseline level and post-sand training were compared.

Table 1. Bipedal stance. Parameters expressed as mean ± SD.

Variable and Condition	Test 1: Baseline	Test 2: Post-Balance	Test 3: Post-Sand	Test 1 vs. 2 Cohen's d	Test 2 vs. 3 Cohen's d	Test 1 vs. 3 Cohen's d
Path length (mm)						
Eyes open	493 ± 132	348 ± 38 †	483 ± 64	1.5 (large)	2.6 (large)	0.1 (small)
Eyes closed	481 ± 129	413 ± 53	524 ± 167	0.7 (medium)	0.9 (large)	0.3 (small)
Romberg	0.98 ± 0.14 *	1.19 ± 0.11	1.10 ± 0.38	1.5 (large)	0.3 (small)	0.3 (small)
Speed (mm/s)						
Eyes open	16.7 ± 4.48	11.8 ± 1.29 †	16.5 ± 2.16	1.5 (large)	2.6 (large)	0.1 (small)
Eyes closed	16.1 ± 4.37	13.8 ± 1.73	17.8 ± 5.85	0.7 (medium)	0.9 (large)	0.3 (small)
Romberg	0.97 ± 0.14 *	1.17 ± 0.10	1.09 ± 0.39	1.6 (large)	0.3 (small)	0.4 (small)
X Mean (mm)						
Eyes open	−11.5 ± 10.1	−10.6 ± 8.9	−8.0 ± 3.6	0.1 (small)	0.4 (small)	0.5 (small)
Eyes closed	−12.2 ± 10.6	−13.1 ± 6.9	−8.9 ± 4.7	0.1 (small)	0.7 (medium)	0.4 (small)
Y Mean (mm)						
Eyes open	−21.6 ± 5.0	−16.1 ± 12.3	−11.7 ± 7.0 #	0.6 (medium)	0.4 (small)	1.6 (large)
Eyes closed	−21.6 ± 7.8	−19.2 ± 8.3	−13.4 ± 6.3	0.3 (small)	0.8 (medium)	1.2 (large)
X RMS (mm)						
Eyes open	0.47 ± 0.25	0.34 ± 0.09 †	0.62 ± 0.11	0.7 (medium)	2.8 (large)	0.8 (medium)
Eyes closed	0.49 ± 0.25	0.45 ± 0.08	0.72 ± 0.24	0.2 (small)	1.5 (large)	0.9 (large)
Y RMS (mm)						
Eyes open	0.60 ± 0.12 *	0.41 ± 0.08	0.43 ± 0.13	1.9 (large)	0.2 (small)	1.4 (large)
Eyes closed	0.61 ± 0.08	0.49 ± 0.13	0.42 ± 0.12 #	1.1 (medium)	0.6 (medium)	1.9 (large)

X Mean: mean position in medial-lateral plane; Y Mean: mean position in antero-posterior plane; X RMS: root-mean-square amplitude in medial-lateral plane; Y RMS: root-mean-square amplitude in the antero-posterior plane; (+/−) sign indicate the direction on X and Y axis; * = significant difference between Test 1 and 2; † = significant difference between Test 2 and 3; # = significant difference between Test 1 and 3. Significance: $p < 0.05$.

Test 3 reported displacements of the center of pressure (CoP) in the medio-lateral and anterior-posterior directions (X and Y mean variables) closer to zero compared with previous tests. This meant that participants had shorter excursions in both planes, especially in the anterior-posterior plane during open eyes condition.

The comparison dominant monopedal variables shown in Table 2 depict significant improvements in path length (23.4% decrease, $p = 0.015$) and speed (30.5% decrease, $p = 0.009$) in open eyes condition for baseline level and post-sand training, which suggest an improvement in postural control after the intervention of a balance training program. As in the previous section, the results got worse once the substitute intervention in the sand was completed. In conditions of visual deprivation, there were improvements in the main indicators of stability after the balance training program and a slight worsening once the replacement intervention in the sand was finished.

The results of the non-dominant monopedal stance (Table 3) also showed an improvement in the main variables related to postural control with open eyes after the intervention of a balance training program: improvement in path length (22.5% decrease, $p = 0.005$) and speed (25.7% decrease, $p = 0.004$). Similar to the bipedal stance, deteriorations in open eyes conditions can be observed after completion of the sand training. A comparison of baseline level and post-balance training in closed eyes conditions showed an improvement in the main variables related to postural control after the intervention of a balance training program.

Table 2. Dominant monopedal stance. Parameters expressed as mean ± SD.

Variable and Condition	Test 1: Baseline	Test 2: Post-Balance	Test 3: Post-Sand	Test 1 vs. 2 Cohen's d	Test 2 vs. 3 Cohen's d	Test 1 vs. 3 Cohen's d
Path length (mm)						
Eyes open	960 ± 194 *	736 ± 113	837 ± 122	1.41 (large)	0.9 (large)	0.8 (medium)
Eyes closed	2107 ± 415	1725 ± 565	1794 ± 503	0.77 (medium)	0.1 (small)	0.7 (medium)
Romberg	2.24 ± 0.48	2.37 ± 0.84	2.20 ± 0.73	0.19 (small)	0.2 (small)	0.1 (small)
Speed (mm/s)						
Eyes open	24.9 ± 5.9 *	17.3 ± 3.3	21.0 ± 4.9	1.58 (large)	0.9 (large)	0.7 (medium)
Eyes closed	64.7 ± 15.7	50.6 ± 18.8	52.9 ± 15.9	0.81 (large)	0.1 (small)	0.7 (medium)
Romberg	2.68 ± 0.70	2.99 ± 1.24	2.67 ± 1.11	0.31 (small)	0.3 (small)	0.0 (small)
X Mean (mm)						
Eyes open	18.8 ± 44.9	−2.78 ± 4.89	−2.77 ± 5.97	0.50 (small)	0.0 (small)	0.5 (medium)
Eyes closed	71.5 ± 55.4	−1.45 ± 4.08	22.2 ± 42.5	1.78 (large)	0.7 (medium)	1.0 (large)
Y Mean (mm)						
Eyes open	−22.4 ± 5.3	−19.2 ± 10.1	−10.2 ±8.2	0.40 (small)	1.0 (large)	1.8 (large)
Eyes closed	−14.4 ± 12.9	−17.9 ± 14.1 †	1.43 ± 16.15	0.26 (small)	1.1 (large)	0.9 (large)
X RMS (mm)						
Eyes open	7.52 ± 1.16	7.87 ± 0.78	7.60 ± 1.53	0.35 (small)	0.2 (small)	0.1 (small)
Eyes closed	6.91 ± 2.61	7.66 ± 0.71	7.80 ± 2.07	0.39 (small)	0.1 (small)	0.4 (small)
Y RMS (mm)						
Eyes open	1.88 ± 1.57	1.04 ± 0.27	1.10 ± 0.39	0.75 (medium)	0.2 (small)	0.7 (medium)
Eyes closed	2.87 ± 0.88	2.66 ± 1.39	2.46 ± 1.06	0.18 (small)	0.2 (small)	0.4 (small)

X Mean: mean position in medial-lateral plane; Y Mean: mean position in antero-posterior plane; X RMS: root-mean-square amplitude in medial-lateral plane; Y RMS: root-mean-square amplitude in the antero-posterior plane; (+/−) sign indicate the direction on X and Y axis; * = significant difference between Test 1 and 2; † = significant difference between Test 2 and 3. Significance: $p < 0.05$.

The Romberg quotient related to bipedal stance showed a significant increase between baseline level and post-balance training for the path length ($p = 0.024$) and mean speed ($p = 0.023$). On the other hand, the Romberg quotient related to non-dominant monopedal stance showed a significant increase between post-balance training and post-sand training for the path length ($p = 0.019$) and mean speed ($p = 0.013$).

Table 3. Non-dominant monopedal stance. Parameters expressed as mean ± SD.

Variable and Condition	Test 1: Baseline	Test 2: Post-Balance	Test 3: Post-Sand	Test 1 vs. 2 Cohen's d	Test 2 vs. 3 Cohen's d	Test 1 vs. 3 Cohen's d
Path length (mm)						
Eyes open	918 ± 102 *	772 ± 59	806 ± 182	1.7 (large)	0.2 (small)	0.8 (medium)
Eyes closed	2435 ± 662	1887 ± 682	1491 ± 332	0.8 (large)	0.7 (medium)	1.8 (large)
Romberg	2.63 ± 0.53	2.66 ± 0.98 †	1.94 ± 0.66	0.1 (small)	0.9 (large)	1.2 (large)
Speed (mm/s)						
Eyes open	27.6 ± 3.4 *	20.5 ± 2.1	23.7 ± 6.2	2.5 (large)	0.7 (medium)	0.8 (medium)
Eyes closed	76.4 ± 23.8	55.5 ± 22.7	43.0 ± 12.1	0.9 (large)	0.7 (medium)	1.8 (large)
Romberg	2.74 ± 0.66	2.73 ± 1.22	1.93 ± 0.76	0.0 (small)	0.8 (medium)	1.1 (large)
X Mean (mm)						
Eyes open	2.95 ± 1.70	1.78 ± 2.78	1.91 ± 1.39	0.5 (small)	0.1 (small)	0.6 (medium)
Eyes closed	−98.6 ± 39.0 *	−18.1 ± 45.8	−2.5 ± 3.8 $^{\#}$	1.9 (large)	0.5 (small)	3.5 (large)
Y Mean (mm)						
Eyes open	−31.4 ± 8.1	−27.7 ± 7.6	−13.8 ± 11.3	0.5 (small)	1.4 (large)	1.8 (large)
Eyes closed	−22.0 ± 14.6	−28.0 ± 6.3 †	−8.14 ± 8.01 $^{\#}$	0.5 (medium)	2.8 (large)	1.2 (large)

Table 3. *Cont.*

Variable and Condition	Test 1: Baseline	Test 2: Post-Balance	Test 3: Post-Sand	Test 1 vs. 2 Cohen's d	Test 2 vs. 3 Cohen's d	Test 1 vs. 3 Cohen's d
X RMS (mm)						
Eyes open	3.44 ± 0.34	3.62 ± 0.42	3.58 ± 0.54	0.6 (medium)	0.0 (small)	0.5 (small)
Eyes closed	8.48 ± 2.00	8.16 ± 0.56	7.56 ± 1.26	0.2 (small)	0.6 (medium)	0.5 (medium)
Y RMS (mm)						
Eyes open	1.48 ± 0.49	1.02 ± 0.16	1.11 ± 0.33	1.3 (large)	0.4 (small)	1.0 (large)
Eyes closed	4.32 ± 2.63	2.93 ± 1.29	1.79 ± 0.45	0.7 (medium)	1.1 (large)	1.3 (large)

X Mean: mean position in medial-lateral plane; Y Mean: mean position in antero-posterior plane; X RMS: root-mean-square amplitude in medial-lateral plane; Y RMS: root-mean-square amplitude in the antero-posterior plane; (+/−) sign indicate the direction on X and Y axis; * = significant difference between Test 1 and 2; † = significant difference between Test 2 and 3; # = significant difference between Test 1 and 3. Significance: $p < 0.05$.

Tests 2 and 3 reported displacements in the center of pressure (CoP) in the medio-lateral and anterior-posterior directions (X and Y mean variables) closer to zero compared with Test 1. These improvements were only significant for closed eyes conditions.

Figure 2 shows the results of the three tables in a graphical way to help the reader visualize the variations in the variables under examination for the three tests.

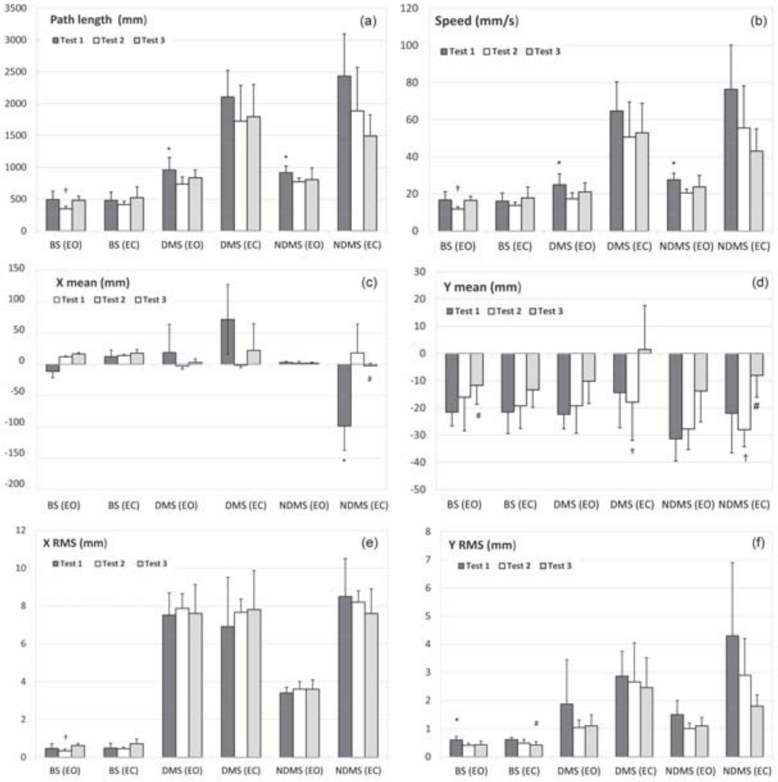

Figure 2. Stabilometric variables compared for bipedal stance (BS), dominant monopedal stance (DMS), and non-dominant monopedal stance (NDMS) for eyes open (EO) and eyes closed (EC) in baseline (Test 1), post-balance (Test 2) and post-sand (Test 3) tests. (**a**) Path length, (**b**) Speed, (**c**) X mean, (**d**) Y mean, (**e**) X RMS and (**f**) Y RMS. * = significant difference between Test 1 and 2; † = significant difference between Test 2 and 3; # = significant difference between Test 1 and 3. Significance: $p < 0.05$.

4. Discussion

The present study was designed to evaluate the effect of a 12-week-long balance training program on the postural control of elite male beach volleyball players and the effect on balance when swapping to specific sports training in sand in the 12 weeks following the program. The results of the present study showed an improvement in postural control, mainly in the monopedal stance, after the implementation of a balance training program. Subsequently, a worsening in postural control values was reported after the elimination and replacement of the balance training. The reference variables such as TE and speed decreased after balance training (BT) intervention, both in bipedal and monopedal stances, although improvements were only significant in the monopedal stance for the open eyes condition. Despite non-significant improvements being found in bipedal stance after BT intervention, it was reported that this condition significantly worsened after the elimination of the BT program. Subsequently, a worsening in postural control values was reported after the elimination and replacement of the balance training.

The ability to maintain or recover balance in any sport is necessary for the correct performance of specific actions [28]. This ability has a relevant role in the performance of volleyball actions, either in contact with the ground (approach to the net, receptions, and placements) or in the air phase (attack and blocking) [42,43]. Several meta-analyses based on the effects of balance training [9,11,17,44,45] revealed that the majority of studies reported a positive effect on postural control.

Consistent with the previous meta-analysis, our results showed an improvement in postural control after an intervention based on balance training. The results indicated an improvement between baseline level and post-balance training in the path length and speed variables, for both eyes open and eyes closed conditions, although improvements were only significant for the open eyes condition. This can be explained by the improvement in both conditions, especially in open eyes conditions, possibly due to the greater volume of training carried out under this condition. In the same way, the Romberg quotients for these variables increased significantly between baseline level and post-balance training in bipedal stances, an indicator of a major contribution of the visual field to postural stability. These findings are in line with the meta-analysis of Kümmel et al. [21], in which the effectiveness of balance training was shown only in those activities carried out under the same training conditions. Similarly, some studies [34,46] comparing different groups of athletes reported improvements mainly for open eyes conditions, suggesting the importance of specificity of training and the stimulation of proprioceptive channels in postural control.

Considering the stance conditions, the improvements found were only significant in dominant and non-dominant monopedal stances for the TE and speed variables. This may be because most actions in volleyball and beach volleyball require high motor control in standing conditions [12,28], so an increase of training in the monopedal stance can be a sufficient stimulus to produce adaptations. Likewise, the fact that there were more parameters enhanced in the non-dominant monopedal stance may be due to the muscular reinforcement of the weakest limb as a consequence of the training program [12].

The results of our study were mainly compared with the study of Pau et al. [12], who reported a potential effect of balance training in postural control of female volleyball players. Conversely to our results, the improvements were obtained for the closed eyes condition. They obtained significant results on sway for both bipedal and non-dominant monopedal stances. However, non-significant results were found in the dominant monopedal stance.

On the other hand, the results of the present study showed a significant decrease in postural control values after removing the balance training program. Therefore, sport-specific training in the sand would not act as a substitute for balance training in players adapted to play on unstable surfaces. This fact was observed in non-athletic populations [47,48] where a process of detraining or loss of postural control could be observed after a period of inactivity or cessation of the stimulus. In the sports field, Dai et al. [27] studied the process of detraining in volleyball players one month after the end of the season. As in the present study, the average speed increased and the CoP speed parameters tended to change in all directions, suggesting a decrease in postural control. Nevertheless, studies that relate postural control and detraining have limitations mainly due to sample characteristics and detraining modality. It should be noted that during the post-sand period, displacements of the center of pressure (CoP) in the medio-lateral and anterior-posterior directions (X and Y mean variables) were closer to zero. This could reveal a specific adaptation to sand training, questioning the real transfer of BT to the beach volleyball balance performance due to the high level of specificity of this training [21]. Therefore, these results could be interpreted as a worsening of postural control for the general population although they could also be due to specific adaptations for beach volleyball sport practice.

In conclusion, the BT program improved the postural control of beach volleyball players. The elimination and replacement of specific beach volleyball training apparently could be interpreted as a worsening of postural control. However, specific adaptations to beach volleyball should not be entirely discarded. Further investigation is needed to investigate the effect of BT on beach volleyball balance performance.

A limitation of the present study is that its participants were all male, viz., with a body center of mass on average higher than females, which could influence different gender balance strategies. Involving females as well would allow larger generalizations of the findings on the whole. Furthermore, it can only be supposed that a sport-specific sand training could effectively develop the dynamic and open eyes postural control acknowledged as required for successful beach volleyball performance. The present study did not specifically investigate this. Therefore, the effect of ecological sand training on dynamic performance should be the object of further research in beach volleyball players. The transfer of dynamic balance management to performance remains unknown and needs to be evaluated during real matches on sand.

5. Conclusions

The results of the present study show that a balance training program in beach volleyball players is effective for acquiring proper static balance control. It was also shown that the same players undergoing their habitual and ecological beach volleyball sand training experienced worsening of their static balance control. It can be hypothesized that beach volleyball sand training might be selectively effective in acquiring a capability different from static balance control, namely a dynamic and open eyes postural control during beach volleyball practice. Such a capability might be acquired at the price of a worsening of static balance control scores. Further studies should investigate relationships between sand training and beach volleyball performance. To date, it can only be hypothesized that beach volleyball players aiming at conditioning specific postural control abilities such as dynamic and open eyes postural balance should favor sand training over gym training.

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Performance and Training Load Profiles in Recreational Male Trail Runners: Analyzing Their Interactions during Competitions

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Abstract: Endurance sports like trail running constitute an extensive individual modality causing numerous physiological changes to occur in the athlete. In this sense, an adequate monitoring of training load appears to be essential to improve competition performance. The aim of this study was two-fold: (i) to analyze trail runners' weekly load variations in the four weeks leading up to a trail running competition, and (ii) to determine the relationship between the runners' pacing in competitions and their physical fitness and workload parameters. Twenty-five amateur male trail runners (age: 36.23 ± 8.30 years old; minimum International Trail Running Association performance index: 600) were monitored daily for the duration of a season (52 weeks). External load (distance covered, pace) and internal load (rate of perceived exertion) were measured daily. Additionally, weekly workload measures of acute:chronic workload ratio (ACWR), training monotony, and training strain were calculated. The runners were also assessed for maximal aerobic speed (MAS) every four months. No significant differences in workload measures ($p > 0.05$) were observed in the four weeks leading up to each short trail competition; however, leading up to the long trail, ultra-trail medium, and ultra-trail long/extra-long competitions, the differences in the runners' workload measures were significant ($p < 0.05$). In the short trail, pace was found to be moderately correlated with the ACWR of total distance (r = −0.334) and with training monotony of rate of perceived exertion (RPE) (r = −0.303). In the ultra-trail, a large correlation was observed between pace and elevation accumulated (r = 0.677). We concluded that significant workload differences from one week to the next only occurred in preparation for longer-distance competitions, with sudden acute load decreases and very low ACWR values reported mainly in weeks 1 and 2 of the taper. Meaningful relationships were found between performance (pace) and MAS for longer trails and between pace and MAS for ultra-trail competitions.

Keywords: sports training; monitoring; endurance sports; trail running; periodization

1. Introduction

Quantifying load contributes to a more precise and individualized training process [1]. It also assists in pinpointing the impacts of exercise on organic and physical functions and maintaining balance in the fitness–fatigue relationship [2]. For these reasons, closely monitoring training load in preparation for competition is common across various modalities [3]. The training load has two components: the external load (the physical and mechanical dimension of the imposed exercise) and the internal load (the biological response to the exercise) [4]. Monitoring training load during training can optimize the sports planning process and ultimately improve an athlete's performance [5]. Different instruments can be used in the implementation of training load monitoring; for example, the global satellite navigation system (GSNS) facilitates external load monitoring by analyzing the distances traveled by an athlete at different speed ranges [6]. Meanwhile, cardio-frequencimeters, which help to control heart rate, are often used in internal load monitoring. Effort scales—which are notable for their validity and reliability [7,8]—may also be used to measure rate of perceived exertion (RPE) as an element of internal load; the RPE scale is multiplied by session time (in minutes) to obtain a global indicator of the designated session-RPE load (s-RPE) [8].

Endurance sports constitute an extensive individual modality (duration and distance), causing numerous physiological changes to occur in the athlete [9]; thus, load monitoring can be crucial to performance improvement [2]. In a study developed on trail running, which employed load monitoring, the RPE values presented in male trail runners for the session are 213.7 ± 223.95 A.U. [10]. These training session values make it possible to quantify the weekly workload and to extrapolate that those weeks with values above 1500 A.U. are associated with an increased occurrence of injury and conditioning performance [11]. Spikes in load must be avoided; the acute:chronic workload ratio (ACWR) makes it possible to assess the chronic load and ensure that it is sufficient for the acute load being imposed on the athlete [12]. In this way, spikes in load create new stimuli and increase the athlete's physical levels and, consequently, their performance [13]. Bearing in mind that the spikes originate variations in the load, they must be weighed across multiple weeks and within individual weeks to allow for appropriate load distribution based on the adaptation and effects on the athlete [14]. Other indices that can be calculated based on external or internal load indicators are the training monotony and training strain—both originally proposed by Foster [15], the training monotony being calculated through the workload's average divided by the standard deviation of the workload and the training strain by monotony multiplied by the workload [15]. Using these two indices (monotony and strain), Matos et al. (2020) developed a study of trail running athletes that demonstrated that monotony values between 0.6 and 0.9 resulted in limitations in the athletes' performance [16].

The primary goal of athletes and coaches being to improve performance in competition, the way to achieve this is through overcompensation caused by load [17]. For this effect to succeed, the training must be planned with great precision to involve phases of overload (e.g., high volumes, great intensity, and diversity of exercises) [18]. A tapering phase—marked by a reduction in load volume, but with sustained frequency and intensity—is used in many sports to incorporate more specific exercises [17,19]. In the tapering phase, athletes and coaches must pay special attention to the balance between the training volume and the length of the phase to ensure that the best performance coincides with the competition's time [20]. In endurance sports, the meta-analysis developed by Bosquet et al. (2007) recommends a two-week tapering phase, involving volume reductions of 41%–60% while training frequency and intensity remain unchanged [21].

In any sport, workload indices are vital to understanding an athlete's adaptations, through an appropriate training prescription, ensuring the best performance in competition [2]. Analyzing the variation in the athlete's response to training is equally essential; the relationship between training and performance is a system of input and output, in which the athlete receives training (input) and generates a final competition performance (output) [22]. In this sense, the relationship can be likened to a dose-response effect in which the athlete's physiological response is derived from their training load—the stimulus [23]. Using the dose-response relationship—which is modeled as an inverted

U-curve—in the planning of an athlete's training, it is possible to adjust the load to target the best performance [24].

Although the load is an essential variable to improve performance, there are intrinsic factors in the athlete that inevitably play an important role. For example, in modalities of extensive character, the maximum volume of oxygen appears to assume a considerable preponderance [25]. In this sense, the literature presents reliable tests such as the Cooper 12-min run test or 5-min field test, making it possible to estimate the maximum oxygen uptake and calculate the aerobic performance [26,27]. Therefore, through the interaction between an athlete's training load and their physical capacity, in trail running also, it is essential to understand this, helping coaches and athletes to predict and plan effectively and contributing to the performance in competition.

However, to the authors' knowledge, no study has been developed on the interaction of training load with performance in competition in trail running athletes. Furthermore, no research has been developed on training load interaction with competition over a trail running season. Thus, the objectives of the present study were (i) to analyze pace and workload indices in the four weeks leading up to different types of competitions, and (ii) to identify correlations between pace, workload indices, and other variables of physical fitness in different types of competitions (short trail (<21 km); long trail (22–42 km); ultra-trail medium (43–69 km); ultra-trail long/extra-long (>70 km).

2. Materials and Methods

2.1. Participants

Twenty-five recreational male trail running athletes participated in the study (age: 36.23 ± 8.30 years old; height: 172.12 ± 5.12 cm; body mass: 67.24 ± 5.97 kg; minimum International Trail Running Association performance index: 600). All were required to participate in the trail running championship (short trail (<21 km), long trail (22–42 km), ultra-trail medium (43–69 km), ultra-trail long/extra-long (>70 km)) in Portugal in the 2018/2019 season. Inclusion criteria included (i) participation in the national trail running championships, (ii) more than three years' experience in the sport, (iii) registration in all training sessions, (iv) registration in all competitions, and (v) not having been injured for more than three consecutive weeks in the 12 months prior (aiming to allow determination of the chronic load for the period). Before the study began, all athletes were informed of the objectives, procedures, and protocol of the study and voluntarily signed an informed consent form. The study was carried out following the Helsinki Declaration's (1964) ethics recommendations for studies on humans.

2.2. Experimental Approach to the Problem

This study followed a cohort study design. Over 52 weeks, all training sessions were monitored using global positioning systems (GPS) that quantified both the total distance covered and, with the use of the Borg CR-10 scale, the RPE and session-RPE. The athletes and their coaches determined the nature and content of the training sessions. A total of 148.12 ± 57.53 training sessions was analyzed for each athlete. Every four months (Figure 1), athletes were assessed for their anthropometry and aerobic performance; for each week of training, load indicators were used to calculate the acute load (the sum of the weekly training loads), the acute:chronic workload ratio, the training monotony, and the training strain. Training weeks were defined as starting on Monday and ending on Sunday. Throughout the testing, pace was calculated and used as the primary performance outcome.

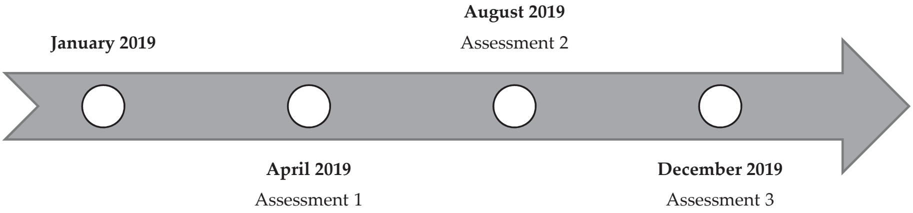

Figure 1. Timeline of assessments during 52 weeks of study.

Per the study's objectives, training load indexes were analyzed and compared in the four weeks leading up to the competition. The weeks were classified as (i) competition week (the week during which the competition occurred), (ii) week −1 (the week of training before the competition), (iii) week −2 (two weeks before the competition), and (iv) week −3 (three weeks before the competition). Meanwhile, the trail running competitions were classified according to the Associação Trail Running Portugal, the organization responsible for trail running in Portugal as ST: short trail (<21 km), LT: long trail (22–42 km); UT-M: ultra-trail medium (43–69 km), and UT-L/XL: ultra-trail long/extra-long (>70 km).

2.3. Periodic Assessment

2.3.1. Anthropometrics Composition

For bodyweight, we used the Tanita BC-601 (Tokyo, Japan, measured to the nearest 0.1 kg), and for height, the stadiometer Seca 217 (Hamburg, Germany, measured to the nearest 0.1 cm).

2.3.2. Aerobic Performance

To assess aerobic performance, we asked all athletes to participate in a 5-min field test (with high validity and reliability), as described by Berthon et al. [27]. Before the test, the athletes performed a standard 5-min warm-up consisting of light running and lower limb mobility. The test was administered on a flat track during morning hours, with temperatures varying from 15 to 25 °C (depending on the hour and day). The athletes were instructed to maintain a constant pace and to avoid resting for the duration of the test to achieve maximal performance and recovery. Their total distance was recorded at the end of the 5 min. Their maximal aerobic speed (MAS) was determined by dividing the total distance in meters by the time in seconds, with the final result expressed in m/s (MAS = total distance/time).

2.4. Training Load Monitoring

2.4.1. Distance Covered

Using the integrated GPS technology in the Polar V800 watch (37 mm × 56 mm × 12.7 mm, weight: 79 g) (Polar, Finland), athletes self-reported the total distance they covered in each of their training sessions. The selected watch model was tested for validity in previous studies and demonstrated acceptable values of accuracy [28].

2.4.2. Rate of Perceived Exertion

Thirty minutes after the training session, athletes were asked the question, "How hard was the training session?" Their response—their RPE—was recorded using the Borg CR-10 scale [29]; this scale was introduced to the athletes two weeks before the study to ensure their familiarity and ability to provide precise responses. Based on the reported RPE value and the duration of the training session in minutes, the session-RPE was calculated and expressed in arbitrary units (A.U.) [6]. This process was repeated for all training sessions to quantify internal load [30].

2.4.3. Workload Indices

Based on the variables of distance, duration, and sRPE, the following indices were determined: (i) weekly training load (the sum of all training loads for the week), (ii) acute:chronic workload ratio (ACWR: calculated by dividing the acute load (the current week's training load) by the chronic workload (the average of the previous four weeks' workloads)) [12], (iii) training monotony (the average of the last seven days' workloads divided by the standard deviation of the last seven days' workloads) [15], and (iv) training strain (monotony multiplied by workload) [15].

2.5. Competition Monitoring

Pace

From the races completed by the athletes and using the different categories, the time of the race and the distance covered were collected, calculating the pace.

2.6. Statistical Analysis

The results were expressed as means and standard deviations. The data were checked for normality ($p > 0.05$) and homogeneity ($p > 0.05$), and an ANOVA of repeated measures was subsequently performed to compare the workload indices (acute load, ACWR, training monotony, and training strain) of the variables (sRPE, total distance, and total time) between the four weeks—that is, the three weeks leading up to the competition and the week of the competition. Bonferroni's post hoc test was used to analyze pairwise variations (week vs. week analysis). The level of statistical significance was set at $p < 0.05$. Additionally, the standardized effect size (ES) of Cohen's d was calculated for pairwise comparisons. The magnitude of the ES was categorized based on the following thresholds: ≤0.2 (trivial), from 0.3 to 0.6 (small), from 0.6 to 1.2 (moderate), from 1.2 to 2.0 (large), and >2.0 (very large) [31]. The associations between pace, physical fitness, and workload indices were made with the Pearson correlation test (r), using the mean values of the four weeks leading up to the competition. The magnitude of the correlation was categorized based on the following thresholds: <0.1 (trivial), from 0.1 to 0.3 (small), from 0.3 to 0.5 (moderate), from 0.5 to 0.7 (large), from 0.7 to 0.9 (very large), and ≥0.9 (nearly perfect). SPSS Statistics software (version 24, IBM Corporation, Armonk, NY, USA) was used for the analysis.

3. Results

The pace in different categories of trail competitions can be found in Table 1, seven athletes having participated in the ST category, seven athletes in LT, six athletes in UT-M, and five athletes in UT-L/XL. The pace (in minutes per kilometer) for short trail (ST) was 6.36 ± 1.97; long trail (LT) was 6.83 ± 1.72; ultra-trail medium was 7.32 ± 1.41; and ultra-trail long or ultra-trail extra-long was 8.48 ± 1.57.

Table 1. Descriptive statistics of pace in different categories of trail competitions.

	ST	LT	UT-M	UT-L/XL
Pace in competition (min/km)	6.36 ± 1.97	6.83 ± 1.72	7.32 ± 1.41	8.48 ± 1.57

ST: short trail (<21 km); LT: long trail (22–42 km); UT-M: ultra-trail medium (43–69 km); UT-L/XL: ultra-trail long/extra-long (>70 km).

Table 2 presents the weekly variations of acute load, acute chronic workload ratio, training monotony and strain of total distance, total time and RPE before the ST competitions. No significant differences ($p > 0.05$) were found between the workload variables before the ST competitions. It was found that in ST competitions, the lowest acute load TD (36.28 km), and sRPE (752.93 A.U.) occurred in the week of competition, and the lowest acute load TT (202.14 min) occurred in week-2, while the greatest occurred in week-1, namely 42.34 km, 254.58 min and in week-3, that is 946.19 A.U. This represents a difference of −14.3% total distance from the week-1 to the week of competition, 25.9% of total time from the week-2 to the week-1, and −20.4% of sRPE from the week-3 to the week of competition.

Table 2. Descriptive statistics of workload measures before short trail (ST) competitions.

	wC	w-1	w-2	w-3	*p*
alTD (km)	36.28 ± 26.28	42.34 ± 33.57	35.97 ± 25.65	42.08 ± 29.67	0.466
alTT (min)	202.50 ± 139.13	254.58 ± 226.67	202.14 ± 145.65	241.53 ± 189.55	0.344
alsRPE (A.U.)	752.93 ± 514.97	877.02 ± 822.97	791.49 ± 633.43	946.19 ± 926.34	0.816
acwrTD (A.U.)	0.95 ± 0.57	0.93 ± 0.66	0.88 ± 0.51	0.96 ± 0.57	0.962
acwrTT (A.U.)	0.95 ± 0.59	0.87 ± 0.62	0.86 ± 0.52	0.96 ± 0.62	0.784
acwrRPE (A.U.)	0.95 ± 0.66	0.97 ± 0.83	0.88 ± 0.63	0.97 ± 0.73	0.940
tmTD (A.U.)	0.66 ± 0.35	0.69 ± 0.41	0.71 ± 0.44	0.75 ± 0.39	0.601
tmTT (A.U.)	0.63 ± 0.31	0.65 ± 0.38	0.68 ± 0.42	0.71 ± 0.35	0.582
tmRPE (A.U.)	0.60 ± 0.29	0.62 ± 0.38	0.63 ± 0.35	0.67 ± 0.31	0.673
tsTD (A.U.)	24.24 ± 20.23	37.38 ± 41.68	35.13 ± 43.72	40.67 ± 40.11	0.203
tsTT (A.U.)	122.00 ± 86.70	206.21 ± 243.19	181.46 ± 213.21	213.02 ± 211.19	0.229
tsRPE (A.U.)	462.59 ± 355.67	787.62 ± 1009.31	531.65 ± 465.86	824.47 ± 933.34	0.207

wC: week of the competition; w-1: one week before the competition; w-2: two weeks before the competition; w-3: three weeks before the competition; alTD: acute load total distance; alTT: acute load total time; alsRPE: acute load session-RPE; acwrTD: acute:chronic workload ratio total distance; acwrTT: acute:chronic workload ratio total time; acwrRPE: acute:chronic workload ratio RPE; tmTD: training monotony total distance; tmTT: training monotony total time; tmRPE: training monotony RPE; tsTD: training strain total distance; tsTT: training strain total time; tsRPE: training strain RPE; ES: effect size.

Table 3 presents the weekly variations of acute load, acute chronic workload ratio, training monotony and strain of total distance, total time and RPE before the LT competitions. It was found that in LT competitions, the lowest acute load TD (34.19 km), TT (166.43 min) and sRPE (673.36 A.U.) occurred in the week of competition, while the greatest occurred in week-1, namely 53.93 km, 301.48 min and 1284.31 A.U. This represents a difference of −36.6% total distance, −44.8% of total time and −47.6% of sRPE from the week-1 to the week of competition.

Table 4 presents the weekly variations of acute load, acute chronic workload ratio, training monotony and strain of total distance, total time and RPE before the UT-M competitions. It was found that in UT-M competitions, the lowest acute load TD (26.23 km), TT (139.98 min) and sRPE (457.08 A.U.) occurred in the week of competition, while the greatest occurred in week-2, namely 56.78 km, 344.14 min and in week-1, that is 1183.43 A.U. This represents a difference of −53.8% total distance, −59.3% of total time to week-2 to the week of competition, and −61.4% of sRPE from the week-1 to the week of competition.

Table 5 presents the weekly variations of acute load, acute chronic workload ratio, training monotony and strain of total distance, total time and RPE before the UT-L/XL competitions. It was found that in UT-L/XL competitions, the lowest acute load TD (19.95 km), TT (113.81 min) and sRPE (326.23 A.U.) occurred in the week of competition, while the greatest occurred in week-3, namely 59.29 km, 355.48 min and 1447.33 A.U. This represents a difference of −66.4% total distance, −68% of total time and −77.5% of sRPE from the week-3 to the week of competition.

Table 3. Descriptive statistics of workload measures before long trail (LT) competitions.

	wC	**Week-1**	**Week-2**	**Week-3**	***p***	**Post Hoc**	**ES**
alTD (km)	34.19 ± 26.00	53.93 ± 32.81	48.17 ± 28.54	45.84 ± 31.14	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.014 *	wC vs. w-1: −0.658 $^{\$}$ wC vs. w-2: −0.491 $^{@}$ wC vs. w-3: −0.392 $^{@}$
alTT (min)	166.43 ± 119.07	301.48 ± 188.15	260.06 ± 173.73	267.01 ± 184.84	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −0.842 $^{\$}$ wC vs. w-2: −0.613 $^{\$}$ wC vs. w-3: −0.628 $^{\$}$
alsRPE (A.U.)	673.36 ± 631.35	1284.31 ± 1081.89	1127.22 ± 992.69	1085.70 ± 974.01	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.003 *	wC vs. w-1: −0.674 $^{\$}$ wC vs. w-2: −0.550 $^{@}$ wC vs. w-3: −0.490 $^{@}$
acwrTD (A.U.)	0.73 ± 0.48	1.11 ± 0.57	1.04 ± 0.58	0.98 ± 0.67	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.045 *	wC vs. w-1: −0.721 $^{\$}$ wC vs. w-2: −0.582 $^{@}$ wC vs. w-3: −0.429 $^{@}$
acwrTT (A.U.)	0.66 ± 0.41	1.09 ± 0.59	1.02 ± 0.62	0.99 ± 0.70	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.003 *	wC vs. w-1: −0.836 $^{\$}$ wC vs. w-2: −0.712 $^{\$}$ wC vs. w-3: −0.575 $^{@}$
acwrRPE (A.U.)	0.65 ± 0.46	1.10 ± 0.66	1.01 ± 0.64	1.00 ± 0.79	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.005 *	wC vs. w-1: −0.790 $^{\$}$ wC vs. w-2: −0.671 $^{\$}$ wC vs. w-3: −0.557 $^{@}$
tmTD (A.U.)	0.64 ± 0.34	0.91 ± 0.47	0.85 ± 0.42	0.77 ± 0.41	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 *	wC vs. w-1: −0.636$^{\$}$ wC vs. w-2: −0.593 $^{@}$
tmTT (A.U.)	0.63 ± 0.33	0.87 ± 0.43	0.79 ± 0.36	0.75 ± 0.41	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 *	wC vs. w-1: −0.627 $^{\$}$ wC vs. w-2: −0.559 $^{@}$
tmRPE (A.U.)	0.59 ± 0.29	0.79 ± 0.35	0.73 ± 0.31	0.67 ± 0.33	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 *	wC vs. w-1: −0.633$^{\$}$ wC vs. w-2: −0.552 $^{@}$
tsTD (A.U.)	21.74 ± 19.45	57.33 ± 52.00	49.23 ± 45.09	41.54 ± 40.37	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −0.855 $^{\$}$ wC vs. w-2: −0.781 $^{\$}$ wC vs. w-3: −0.579 $^{@}$

Table 3. *Cont.*

	wC	Week-1	Week-2	Week-3	p	Post Hoc	ES
tsTT (A.U.)	115.06 ± 103.18	306.57 ± 270.99	249.09 ± 224.28	222.43 ± 213.18	≤0.001	wC vs. w-1: ≤0.001* wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −0.926 $^{\$}$ wC vs. w-2: −0.813 $^{\$}$ wC vs. w-3: −0.642 $^{\$}$
tsRPE (A.U.)	404.46 ± 432.03	1033.02 ± 1003.36	971.43 ± 1007.67	839.89 ± 931.03	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.005 *	wC vs. w-1: −0.806 $^{\$}$ wC vs. w-2: −0.675 $^{\$}$ wC vs. w-3: −0.543 $^{@}$

wC: week of the competition; w-1: one week before the competition; w-2: two weeks before the competition; w-3: three weeks before the competition; alTD: acute load total distance; alTT: acute load total time; alsRPE: acute load session-RPE; acwrTD: acute:chronic workload ratio total distance; acwrTT: acute:chronic workload ratio total time; acwrRPE: acute:chronic workload ratio RPE; tmTD: training monotony total distance; tmTT: training monotony total time; tmRPE: training monotony RPE; tsTD: training strain total distance; tsTT: training strain total time; tsRPE: training strain RPE; ES: effect size; *: statistically significant at a $p < 0.05$; ES: effect size; $^{@}$: small ES; $^{\$}$: moderate ES.

Table 4. Descriptive statistics of workload measures before ultra-trail medium (UT-M) competitions.

	wC	Week-1	Week-2	Week-3	p	Post Hoc	ES
alTD (km)	26.23 ± 21.18	53.79 ± 30.33	56.78 ± 38.87	42.01 ± 33.58	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.008 *	wC vs. w-1: −1.054 $^{\$}$ wC vs. w-2: −0.976 $^{\$}$ wC vs. w-3: −0.562 $^{@}$
alTT (min)	139.98 ± 115.65	328.19 ± 190.74	344.14 ± 253.12	248.00 ± 198.86	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.002 *	wC vs. w-1: −1.193 $^{\$}$ wC vs. w-2: −1.038 $^{\$}$ wC vs. w-3: −0.664 $^{\$}$
alsRPE (A.U.)	457.08 ± 427.23	1183.43 ± 859.84	1142.84 ± 995.41	927.31 ± 909.41	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.002 *	wC vs. w-1: −1.158 $^{\$}$ wC vs. w-2: −0.975 $^{\$}$ wC vs. w-3: −0.764 $^{\$}$
acwrTD (A.U.)	0.56 ± 0.44	1.07 ± 0.49	1.16 ± 0.64	1.05 ± 0.73	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.128 $^{\$}$ wC vs. w-2: −1.119 $^{\$}$ wC vs. w-3: −0.877 $^{\$}$
acwrTT (A.U.)	0.52 ± 0.43	1.08 ± 0.51	1.20 ± 0.67	1.05 ± 0.76	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.220 $^{\#}$ wC vs. w-2: −1.220 $^{\#}$ wC vs. w-3: −0.921 $^{\$}$
acwrRPE (A.U.)	0.44 ± 0.36	1.09 ± 0.67	1.21 ± 0.78	1.06 ± 0.85	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.209 $^{\#}$ wC vs. w-2: −1.301 $^{\#}$ wC vs. w-3: −0.965 $^{\$}$

Table 4. *Cont.*

	wC	Week-1	Week-2	Week-3	p	Post Hoc	ES
tmTD (A.U.)	0.57 ± 0.33	0.83 ± 0.41	0.86 ± 0.46	0.73 ± 0.45	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.016 *	wC vs. w-1: −0.831 $^{\$}$ wC vs. w-2: −0.818 $^{\$}$ wC vs. w-3: −0.520 $^{@}$
tmTT (A.U.)	0.56 ± 0.31	0.78 ± 0.33	0.82 ± 0.43	0.67 ± 0.39	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 *	wC vs. w-1: −0.817 $^{\$}$ wC vs. w-2: −0.769 $^{\$}$
tmRPE (A.U.)	0.53 ± 0.28	0.75 ± 0.33	0.78 ± 0.40	0.65 ± 0.37	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.048 *	wC vs. w-1: −0.820 $^{\$}$ wC vs. w-2: −0.777 $^{\$}$ wC vs. w-3: −0.435 $^{@}$
tsTD (A.U.)	17.53 ± 18.43	51.45 ± 47.81	53.97 ± 50.63	38.19 ± 41.91	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: 0.003 *	wC vs. w-1: −0.965 $^{\$}$ wC vs. w-2: −0.930 $^{\$}$ wC vs. w-3: −0.669 $^{\$}$
tsTT (A.U.)	90.92 ± 94.59	284.57 ± 245.93	300.18 ± 259.90	208.25 ± 218.37	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.120 $^{\$}$ wC vs. w-2: −1.045 $^{\$}$ wC vs. w-3: −0.730 $^{\$}$
tsRPE (A.U.)	282.70 ± 315.00	958.68 ± 907.91	949.92 ± 903.27	740.34 ± 849.62	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.036 $^{\$}$ wC vs. w-2: −1.066 $^{\$}$ wC vs. w-3: −0.817 $^{\$}$

wC: week of the competition; w-1: one week before the competition; w-2: two weeks before the competition; w-3: three weeks before the competition; alTD: acute load total distance; alTT: acute load total time; alsRPE: acute load session-RPE; acwrTD: acute:chronic workload ratio total distance; acwrTT: acute:chronic workload ratio total time; acwrRPE: acute:chronic workload ratio RPE; tmTD: training monotony total distance; tmTT: training monotony total time; tmRPE: training monotony RPE; tsTD: training strain total distance; tsTT: training strain total time; tsRPE: training strain RPE; ES: effect size; *: statistically significant at a $p < 0.05$; ES: effect size; $^{@}$: small ES; $^{\$}$: moderate ES; $^{\#}$: large ES.

Table 5. Descriptive statistics of workload measures before ultra-trail long/extra-long (UT-L/XL) competitions.

	wC	Week-1	Week-2	Week-3	p	Post Hoc	ES
alTD (km)	19.95 ± 12.60	50.00 ± 28.75	50.81 ± 33.68	59.29 ± 39.80	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.354 $^{\#}$ wC vs. w-2: −1.213 $^{\#}$ wC vs. w-3: −1.333 $^{\#}$
alTT (min)	113.81 ± 70.79	313.97 ± 185.75	329.61 ± 243.79	355.48 ± 250.85	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.424 $^{\#}$ wC vs. w-2: −1.202 $^{\#}$ wC vs. w-3: −1.311 $^{\#}$
alsRPE (A.U.)	326.23 ± 260.28	1280.55 ± 876.03	1157.14 ± 945.79	1447.33 ± 1262.24	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: ≤0.001 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.443 $^{\#}$ wC vs. w-2: −1.195 $^{\$}$ wC vs. w-3: −1.256 $^{\#}$

Table 5. *Cont.*

	wC	Week-1	Week-2	Week-3	p	Post Hoc	ES
acwrTD (A.U.)	0.51 ± 0.44	0.88 ± 0.47	0.94 ± 0.55	1.09 ± 0.53	≤0.001	wC vs. w-1: 0.008 * wC vs. w-2: 0.005 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −0.835 $^{\$}$ wC vs. w-2: −0.965 $^{\$}$ wC vs. w-3: −1.218 $^{\#}$
acwrTT (A.U.)	0.49 ± 0.44	0.90 ± 0.47	0.91 ± 0.54	1.07 ± 0.56	≤0.001	wC vs. w-1: 0.007 * wC vs. w-2: 0.009 * wC vs. w-3: 0.002 *	wC vs. w-1: −0.891 $^{\$}$ wC vs. w-2: −0.915 $^{\$}$ wC vs. w-3: −1.130 $^{\$}$
acwrRPE (A.U.)	0.41 ± 0.46	0.98 ± 0.70	0.89 ± 0.63	1.04 ± 0.54	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: 0.020 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −0.962 $^{\$}$ wC vs. w-2: −0.870 $^{\$}$ wC vs. w-3: −1.256 $^{\#}$
tmTD (A.U.)	0.49 ± 0.24	0.77 ± 0.45	0.70 ± 0.33	0.77 ± 0.44	0.002	wC vs. w-1: 0.002 * wC vs. w-2: 0.028 * wC vs. w-3: 0.003 *	wC vs. w-1: −0.744 $^{\$}$ wC vs. w-2: −0.675 $^{\$}$ wC vs. w-3: −0.810 $^{\$}$
tmTT (A.U.)	0.49 ± 0.24	0.73 ± 0.41	0.67 ± 0.31	0.72 ± 0.40	0.004	wC vs. w-1: 0.004 * wC vs. w-3: 0.005 *	wC vs. w-1: −0.666 $^{\$}$ wC vs. w-3: −0.720 $^{\$}$
tmRPE (A.U.)	0.47 ± 0.23	0.70 ± 0.42	0.63 ± 0.29	0.72 ± 0.41	0.006	wC vs. w-1: 0.005 * wC vs. w-2: 0.043 * wC vs. w-3: 0.008 *	wC vs. w-1: −0.679 $^{\$}$ wC vs. w-2: −0.611 $^{\$}$ wC vs. w-3: −0.752 $^{\$}$
tsTD (A.U.)	12.15 ± 10.39	49.15 ± 43.69	44.09 ± 39.35	49.88 ± 51.18	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: 0.002 * wC vs. w-3: 0.003 *	wC vs. w-1: −1.086 $^{\$}$ wC vs. w-2: −1.134 $^{\$}$ wC vs. w-3: −1.031 $^{\$}$
tsTT (A.U.)	67.29 ± 53.84	284.10 ± 253.87	269.68 ± 244.20	269.97 ± 268.55	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: 0.002 * wC vs. w-3: 0.002 *	wC vs. w-1: −1.095 $^{\$}$ wC vs. w-2: −1.152 $^{\$}$ wC vs. w-3: −1.052 $^{\$}$
tsRPE (A.U.)	183.79 ± 161.78	1033.11 ± 881.36	958.90 ± 985.45	695.25 ± 581.55	≤0.001	wC vs. w-1: ≤0.001 * wC vs. w-2: 0.010 * wC vs. w-3: ≤0.001 *	wC vs. w-1: −1.227 $^{\#}$ wC vs. w-2: −1.021 $^{\$}$ wC vs. w-3: −1.275 $^{\#}$

wC: week of the competition; w-1: one week before the competition; w-2: two weeks before the competition; w-3: three weeks before the competition; alTD: acute load total distance; alTT: acute load total time; alsRPE: acute load session-RPE; acwrTD: acute:chronic workload ratio total distance; acwrTT: acute:chronic workload ratio total time; acwrRPE: acute:chronic workload ratio RPE; tmTD: training monotony total distance; tmTT: training monotony total time; tmRPE: training monotony RPE; tsTD: training strain total distance; tsTT: training strain total time; tsRPE: training strain RPE; ES: effect size; *: statistically significant at a $p < 0.05$; ES: effect size; $^{\$}$: moderate ES; $^{\#}$: large ES.

Table 6 presents the correlations between the pace and physical fitness and workload indices in different categories of trail competitions. Moderate correlations were found between the pace in ST with acute:chronic workload ratio of total distance (r = −0.334, 95% confidence interval-CI (−0.58; −0.04)), and pace in ST with training monotony of RPE (r = −0.303, 95% CI (− 0.55; 0)); while, pace in LT had moderate correlation with elevation accumulated (r = 0.425, 95% CI (0.24;0.58)). Concerning the pace in UT-M, small correlations were found with acute load of RPE (r = −0.298, 95% CI(−0.51; −0.05)), acute:chronic workload ratio of total distance (r = −0.253, 95%CI (−0.47; −0.01)), acute:chronic workload ratio of RPE (r = −0.263, 95% CI (−0.48; −0.02)), moderate correlations were found with maximal aerobic speed (r = −0.398, 95% CI (−0.59; −0.17)), training strain of total distance (r = −0.305, 95% CI (−0.51; −0.06)), total time (r = −0.305, 95% CI (−0.51; −0.06)), RPE (r = −0.305, 95% CI (−0.51; −0.06)), and very large correlations were found with elevation accumulated (r = 0.703, 95% CI (0.55; 0.81)). In relation to pace in UT-L/XL large correlation were found with elevation accumulated (r = 0.677, 95% CI (0.42; 0.83)).

Table 6. Correlation coefficients between pace and physical fitness and workload indices.

	Pace in ST	Pace in LT	Pace in UT-M	Pace in UT-L/XL
ELac (mt)	r = 0.177 (−0.13; 0.45) $^{@}$	r = 0.425 (0.24; 0.58) $^{**,\$}$	r = 0.703 (0.55; 0.81) $^{**,\sim}$	r = 0.677 (0.42; 0.83) $^{**,\#}$
MAS (m/s)	r = −0.027 (−0.32; 0.28) $^{\&}$	r = 0.198 (−0.01; 0.39) $^{@}$	r = −0.398 (−0.59; −0.17) $^{**,\$}$	r = −0.229 (−0.54; 0.14) $^{@}$
alTD (km)	r = −0.293 (−0.55; 0.01) $^{@}$	r = 0.058 (−0.15; 0.26) $^{\&}$	r = −0.233 (−0.45; 0.02) $^{@}$	r = 0.014 (−0.34; 0.37) $^{\&}$
alTT (min)	r = −0.293 (−0.55; 0.01) $^{@}$	r = 0.042 (−0.17; 0.25) $^{\&}$	r = −0.244 (−0.46; 0) $^{@}$	r = 0.082 (−0.28; 0.42) $^{\&}$
alsRPE (A.U.)	r = −0.166 (−0.44; 0.14) $^{@}$	r = 0.035 (−0.17; 0.24) $^{\&}$	r = −0.298 (−0.51; −0.05) $^{*,@}$	r = 0.067 (−0.29; 0.41) $^{\&}$
acwrTD (A.U.)	r = −0.334 (−0.58; −0.04) $^{*,\$}$	r = −0.058 (−0.26; 0.15) $^{\&}$	r = −0.253 (−0.47; −0.01) $^{*,@}$	r = −0.004 (−0.36; 0.35) $^{\&}$
acwrTT (A.U.)	r = −0.299 (−0.55; 0) $^{@}$	r = −0.058 (−0.26; 0.15) $^{\&}$	r = −0.237 (−0.46; 0.01) $^{@}$	r = −0.038 (−0.39; 0.32) $^{\&}$
acwrRPE (A.U.)	r = −0.273 (−0.53; 0.03) $^{@}$	r = −0.064 (−0.27; 0.15) $^{\&}$	r = −0.263 (−0.48; −0.02) $^{*,@}$	r = −0.026 (−0.38; 0.33) $^{\&}$
tmTD (A.U.)	r = −0.268 (−0.53; 0.04) $^{@}$	r = 0.064 (−0.15; 0.27) $^{\&}$	r = −0.239 (−0.46; 0.01) $^{@}$	r = −0.062 (−0.41; 0.3) $^{\&}$
tmTT (A.U.)	r = −0.260 (−0.52; 0.04) $^{@}$	r = 0.092 (−0.12; 0.29) $^{\&}$	r = −0.224 (−0.45; 0.03) $^{@}$	r = −0.082 (−0.42; 0.28) $^{\&}$
tmRPE (A.U.)	r = −0.303 (−0.55; 0) $^{*,\$}$	r = 0.075 (−0.14; 0.28) $^{\&}$	r = −0.175 (−0.41; 0.08) $^{@}$	r = −0.053 (−0.4; 0.31) $^{\&}$
tsTD (A.U.)	r = −0.271 (−0.53; 0.03) $^{@}$	r = −0.007 (−0.21; 0.2) $^{\&}$	r = −0.305 (−0.51; −0.06) $^{*,\$}$	r = 0.011 (−0.34; 0.36) $^{\&}$
tsTT (A.U.)	r = −0.269 (−0.53; 0.03) $^{@}$	r = −0.011 (−0.22; 0.2) $^{\&}$	r = −0.305 (−0.51; −0.06) $^{*,\$}$	r = 0.040 (−0.32; 0.39) $^{\&}$
tsRPE (A.U.)	r = −0.203 (−0.47; 0.1) $^{@}$	r = −0.004 (−0.21; 0.2) $^{\&}$	r = −0.305 (−0.51; −0.06) $^{*,\$}$	r = 0.049 (−0.31; 0.40) $^{\&}$

ELac: Elevation accumulated; MAS: Maximal aerobic speed; alTD: acute load total distance; alTT: acute load total time; alsRPE: acute load session-RPE; acwrTD: acute:chronic workload ratio total distance; acwrTT: acute:chronic workload ratio total time; acwrRPE: acute:chronic workload ratio RPE; tmTD: training monotony total distance; tmTT: training monotony total time; tmRPE: training monotony RPE; tsTD: training strain total distance; tsTT: training strain total time; tsRPE: training strain RPE; * Correlation is significant at $p < 0.05$; ** Correlation is significant at ≤0.01; $^{\&}$: trivial magnitude; $^{@}$: small magnitude; $^{\$}$: moderate magnitude; $^{\#}$: large magnitude; $^{\sim}$: very large magnitude.

4. Discussion

The purpose of this study was to analyze training load variations within the four weeks before the competition, as well as the associations between pace, physical fitness, and workload indices for the four running competitions. The main finding was that all competitions except the ST saw significant workload differences in the preceding weeks. Large to very large correlations were found between pace and ELac for UT-M and UT-L/XL competitions, while only UT-M showed associations between pace and physical fitness.

Despite a lack of meaningful differences in workload measures during the weeks leading up to the ST competitions, it was found that the workload indices of the overall measures decreased from week-3 to week-2. From week-2 to week-1, the workload indices increased again; then, they tapered off the week before the competition. The acute load decreases and increases for TD, TT, and sRPE varied from −16% to 26%. Given the considerable variation in workloads, it is important to consider the "10 percent rule" in the association between workload and performance, since when variations between weeks greater than 10% are verified, this can condition performance [32].

The ACWR, TM, and TS of TD, TT, and sRPE values remained in the safe zones in the weeks before competition. Although the TM values reported in the present study are considered "safe", as the "danger" threshold is considered to be at approximately 2.0 A.U. [15], in a study conducted on 25 trail running athletes to analyze their workload indices in the three weeks preceding an injury, it was found that TM values remained consistently low until the last week [16]. These data should be analyzed with some caution, as small increases (<10%) to weekly training load in athletes with low chronic loads may result in a low dose-response to training, while the same increase for athletes with extremely high chronic loads may be too much [33]. Determining whether the ACWR thresholds of other sport contexts are comparable to those expected in trail running may also provide useful insights [11].

Longer distance competitions (LT and UT-M) saw a similar pattern of progressive increase in overall acute loads and workload indices from week-3 to week-2, followed by a 2-week tapering phase that resulted in a decrease of up to −53.8% and −59.3% for TD and TT, respectively. Meanwhile, the extra-long competition (UT-L/XL) saw a "wave-like" workload pattern, similar to the ST competition. The longer the trail running competition, the greater the decrease in load from week-1 of training to the week of the competition (−60%, −64%, and −75% for TD, TT, and sRPE, respectively). There remains no consensus as to which tapering technique is best (i.e., the "optimal duration"), especially when it comes to longer distance competitions [20].

Notwithstanding the "optimal" recovery strategy issue, some authors recommend training volume reductions of 40–80% [20,34]. However, massive reductions in training volume and frequency can also increase a runner's risk of detraining and ultimately reduce their performance [35]. It is worth noting that the data in the present study included significant variation. The standard deviations of the overall workload indices remained high (some even greater than the mean) for all competitions; this indicates that within-subject variation was high and was increasingly accentuated in longer competitions. Also noteworthy were the ACWR values for TD, TT, and sRPE, which dropped well below 0.8 A.U. in the last week of taper (competition week)—a potential danger zone for detraining [13].

Regarding the second objective of the present study, it was found that pacing increased with competition distance. The pace in longer competitions showed large and very large correlations with elevation accumulated (Elac); the longer the competition, the greater the Elac, as more uphill terrain is incorporated into the course. It is possible that athletes feel added pressure and motivation to make up time during such courses, given that their pacing is reduced during hill climbs [36,37]. In fact, this pattern was observed in a study conducted on 50 trail runners competing in trail ultramarathons over a period of two years. The runners' mean pace was 9.23 ± 1.13 min/km—which is similar to the UT-L/XL competition pacing in the present study [38]—and, indeed, the data showed that the athletes slowed their pace (to between 8 and 10 min/km) during longer climbs, and then regained their pace (up to 14 min/km) on downhills and shorter climbs with lower altitudes [38]. Only moderate correlations were found between pace and Elac for UT-M competitions. Based on this data, a similar study conducted on 23 recreational trail runners in a 65 km competition showed a moderate correlation between VO2max and performance; the authors associated higher values of VO2max with the submaximal intensities observed in competitions of longer duration [25]. Coaches should assess and monitor VO2max and MAS values during the training process, as this information may influence performance even in longer distance competitions [39].

The present study showed that the longer the trail running competition, the more the acute loads for TD, TT, and sRPE will decrease in the week before a competition. The shortest (ST) and the longest (UT-L/XL) trail competitions each used a one-week taper, while the LT and UT-M competitions used a two-week taper—revealing large decreases in ACWR values for TD, TT, and sRPE variables in all competitions. Pace showed a large to very large correlation with Elac in longer distance competitions, but only the UT-M competition showed a moderate correlation between pace and MAS.

This study also had its limitations, one of which is quantifying metabolic demands as a function of workload, suggesting that the intake of 120 g/h of carbohydrates may limit metabolic fatigue and exercise-induced muscle damage during ultra-endurance races [40,41]. In terms of the sample size, only male athletes were included. Including female athletes in future studies of this nature would be pertinent, as trail running competitions are seeing an increasing number of female participants. Another limitation was the experience level of the sample; as non-professionals, the athletes were not necessarily accustomed to using sRPE to rate their efforts. Incorporating other GPS metrics into such a study—such as accelerations, decelerations, and impacts—would also offer interesting insights regarding non-linear courses and how they may impact an athlete's risk of injury. Information on carbohydrate intake should also be a variable to include in future studies, which may cause differences in loads' perception. Additionally, collecting data on elevation accumulated—including intra-week analyses of uphill cumulative load during training—would be useful for future studies.

5. Conclusions

In our analysis of trail running competitions, pace was found to increase with the length of the competition. Only the longer distance competitions (from LT to UT-L/XL) showed significant workload differences between weeks, with sudden acute load decreases and very low ACWR values for TD, TT, and sRPE primarily in the one–two weeks of taper. Meaningful correlations were observed between performance (pace) and Elac for longer trails, and between pace and MAS for UT-M competitions only. These results highlight the impact of taper strategies when preparing for a competition and navigating the potential risk of detraining. Exposure to the cumulative elevations encountered in a competition is also an important aspect of an athlete's training, particularly when preparing for a longer distance trail running competition.

Our results suggest that coaches and athletes should pay special attention to variations in workload before the competition and the tapering effect based on the race category, thus enhancing the best performance at the right time (race day). On the other hand, and regarding pace, through this study it becomes possible to understand which indices support each category of race, offering an essential tool in preparing trail running races.

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Effect of the Verbal Encouragement on Psychophysiological and Affective Responses during Small-Sided Games

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Abstract: Verbal encouragement (VE) is considered as external motivation provided by physical education teacher. For this reason, this study aimed to examine the effects of VE on psychophysiological and affective responses during small-sided games (SSG). Sixteen male school students (age: 17.37 ± 0.48 years) completed four sessions of a 4-a-side SSG. Two sessions occurred with VE (SSGE), and two sessions did not include VE (SSGNE). Heart rate was continuously recorded, and rating of perceived exertion (RPE) and blood lactate concentration ([La]b) were measured after each training session. Physical enjoyment was assessed after each protocol. Mood state was recorded before and after each training session using the profile of mood-state. HR max, [La]b, RPE, Physical enjoyment, and vigor were higher in SSGE compared to SSGNE (all, $p < 0.001$). The SSGE and SSGNE resulted in a decreased total mood disturbance (TMD) ($p = 0.001$, ES = 0.60; $p = 0.04$, ES = 0.33, respectively) and tension ($p < 0.001$, ES = 0.91; $p = 0.004$, ES = 0.47, respectively), and the vigor was increased after the SSGE ($p < 0.001$, ES = 0.76). SSGE and SSGNE induce similar improvement in TMD and tension. However, SSGE induced higher physiological responses, RPE, enjoyment, and positive mood than SSGNE. Physical education teachers could use VE during specific soccer sessions to improve physical aspects, enjoyment, and mood in participants.

Keywords: soccer players; encouragement; well-being; recovery state; mood state; enjoyment

1. Introduction

During physical education sessions, sports practitioners are dynamically predisposed to improve their physical, cognitive, and emotional capabilities [1], but continuous stimuli provided by the physical education (PE) teacher are needed to make this happen [2]. Thus, during physical activity, PE teachers can stimulate the natural tendencies of active engagement and positive feelings [3]. This stimulation can be created through verbal encouragement, which could influence intrinsic motivation [4]. This, in turn, increases the desire to exercise leading to technical, physical performance, and emotional improvements [4–6].

The use of verbal encouragement has been shown to improve motivation and physical performance in various settings and activities, including small-sided games (SSGs) [6–8]. SSGs are modified versions of the formal game in which the format of play, pitch dimensions, and rules are constrained to augment the perceptions of players to encourage them to focus on a specific technical action or tactical problem [9–12].Previous literature has reported that SSGs are an excellent training method for simultaneously improving the physical, physiological, technical, and tactical aspects of soccer players [6,13–15].

During integrated training situations, verbal encouragement provided by a coach or a teacher is considered a form of external motivation that positively influences physical engagement, positive behavior, and the desire to train [4,5]. Several studies on SSGs have reported the importance of verbal coach encouragement on the game intensity, as expressed as the perceived exertion (RPE) and physiological responses (i.e., heart rate and lactate concentration) [4,6,9]. For example, Rampinini et al. [7] indicated that during different SSG formats (3 vs. 3, 4 vs. 4, 5 vs. 5, and 6 vs. 6), heart rate (HR), blood lactate concentration ([La]b), and RPE were significantly higher during exercises with encouragement when compared to exercises without encouragement.

Recently, it has been proposed that SSGs are used for affective solicitation [15,16]. In this context, studies have suggested that this training method is more effective pedagogically than other conventional training exercises for reducing the risk of psychological consequences that are associated with a lack of positive affective responses [6,17–19]. Indeed, the motivation resulting from SSGs can be more effective in obtaining a positive mood, great physical enjoyment, and high intensity [15,18,20,21]. According to Selmi et al. [6], verbal encouragement leads to increased motivation and physical enjoyment, thus resulting in improved physiological responses during SSGs among soccer players. In addition, Selmi et al. [18] suggested that the motivation resulting from verbal encouragement ensures mood balance during SSGs, while high-intensity intermittent training produces a mood disturbance in soccer players.

Little is known about soccer-specific training regarding the effects of verbal encouragement on players' performance in civilian team sports. To the best of our knowledge, no studies have addressed the effects of verbal encouragement on psychophysiological and affective responses during SSGs in physical education sessions.

Given the importance of verbal encouragement in athlete motivation, as well as the potential influences of exercise intensity and positive affective responses, research intended to fill this gap in the literature is warranted. Therefore, the aim of this study was to examine the effects of verbal encouragement given by PE teachers during soccer SSGs on the psychophysiological responses, mood state, and physical enjoyment of players. We hypothesized that SSGE would produce higher physiological responses, RPE, enjoyment level, and positive mood than SSGNE.

2. Materials and Methods

2.1. Ethical Approval

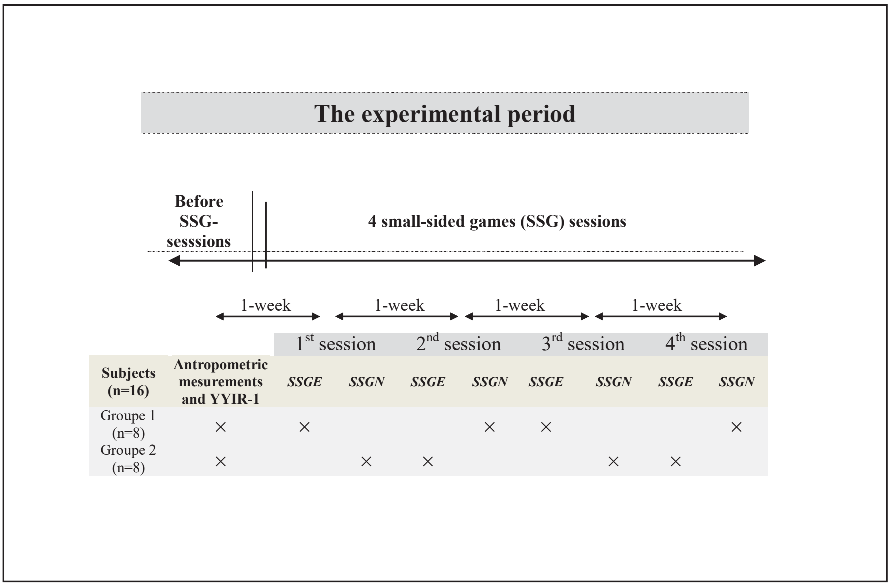
The study was conducted in accordance with the Declaration of Helsinki, and the protocol was fully approved by the research ethics committee of the High Institute of Sports and Physical Education of Kef (ISSEP) (code 2019-0079).

2.2. Participants

Sixteen male students from the same secondary school in Tunisia (participants belong to the same study class) were involved in the study (age: 17.37 ± 0.48 years; experience of physical education: 10.7 ± 0.9 years; height: 176.2 ± 5.9 cm; body mass: 68.1 ± 4.1 kg, %Fat: 12.1 ± 2.5%). The inclusion criteria were: (i) All students competed for the same class; (ii) no injury or illness reported one month before the study and during the study; (iii) no physical or cognitive disease reported. the exclusion criteria were: (i) no regular presence of participants in physical education sessions; (ii) participants fell ill during the study period. During this school season, participants trained three days per week (2 sessions' physical education and 1 session in a school sports club). The participants were familiarized with the experimental protocol and were informed about the procedures. Participants and their parents voluntarily agreed to participate in the study and gave written informed consent after a detailed explanation about the aims and risks involved in the research.

2.3. Procedures

This study followed a cross-sectional design. The research was conducted during the 2019–2020 scholar mid-season (eight weeks after the beginning of the season). Before the beginning of the experimental sessions, anthropometric characteristics were assessed, and the students performed the Yo-Yo intermittent recovery test level1 (YYIR-1) to estimate the maximum heart rate (HRmax) [22]. Four sessions of 4-a-side SSG were performed on separate days, each separated by a one-week interval. Each testing protocol (SSG with the physical education teacher's verbal encouragement (SSGN) and SSG without verbal encouragement (SSGNE)) was repeated twice. During each experimental session, the participants were split into two groups with eight subjects performing SSGE and the other eight performing SSGNE in randomized order. In total, each student performed the SSGE twice, and the SSGNE twice (Figure 1). All measurements were taken on the same synthetic pitch grass at the same time of the day (between 9:00 and 10:30 a.m.) to limit the effects of circadian variations on the measured variables.



		1st session		2nd session		3rd session		4th session	
Subjects (n=16)	**Antropometric mesurements and YYIR-1**	***SSGE***	***SSGN***	***SSGE***	***SSGN***	***SSGE***	***SSGN***	***SSGE***	***SSGN***
Groupe 1 (n=8)	×	×			×	×			×
Groupe 2 (n=8)	×		×	×			×	×	

Figure 1. Experimental design figure. SSG: small-sided game, SSGE: SSG with verbal physical education teacher's encouragement, SSGNE: SSG without verbal physical education teacher's encouragement, YYIR-1: Yo-Yo intermittent recovery test level 1.

HR was continuously monitored during each SSG. Moreover, blood lactate concentration ([La]b), rating of perceived exertion (RPE), and physical activity enjoyment scale (PACES) scores were recorded 5 min after the last bout of the SSG. Participants also had their profile of mood state (POMS) recorded before and after each training session. All participants refrained from strenuous exercise for at least 48 h prior to testing and measurements. Each SSG intervention was preceded by a standardized 15 min warm-up involving low-intensity running, coordination movements, and dynamic stretching and ended with 4 × 8 m sprints; 3 min of recovery separated the warm-up from the first SSG bout [16]. Subjects were allowed to consume drinks during the recovery periods. Subjects were familiarized with the RPE scale, PACES scale, POMS questionnaire, and the SSG regime prior to the beginning of the study. Data were collected by the same sports teacher.

2.4. YO-YO Intermittent Recovery Test (YIRT)

The Yo-Yo intermittent recovery test (YIRT) level 1 was completed according to previously described methods [22]. The YIRT is an incremental intensity test used to evaluate aerobic capacity [22]. This protocol consists of repeated 2 × 20 m runs back and forth between the starting, turning, and finishing lines (180° angle), and at a progressively increased speed, which is controlled by audio beeps from a tape recorder. Between each shuttle, the participants have a 10-s active rest period, consisting of 2 × 5 m of walking. The test was performed on a synthetic grass field. The test was stopped when a participant could no longer maintain the required running speed dictated by the beep for two consecutive occasions or felt that he could not complete the stage. The HR was measured and stored using a Polar Team Sport System (Polar-Electro OY, Kempele, Finland). The highest HR average value over 5 s during the test was recorded as YIRT-HRmax. The validity and reliability of this test to determine the aerobic capacity and maximal HR were tested previously [23].

2.5. Small-Sided Games

A 4 vs. 4 format (without goalkeepers) played on a 35 × 25 m synthetic pitch (~109 m^2 per player) was implemented. The session lasted 25 min (four bouts of 4 min separated by 3 min of passive recovery). The SSGE group played the matches while receiving verbal encouragement from the teacher, while the other group (SSGNE) did not receive verbal encouragement. Teacher encouragement involved moving around the perimeter of the field while encouraging the students using soccer-specific terminology and vocabulary (i.e., Go Go Go, Again Again, move, attack the ball, seek the ball, keep the ball, intercept the ball . . .) and providing new balls when necessary to keep play continuously during the exercise [7,14,19]. The encouragement provided was spontaneous based on the game situation, players' positioning and movement on the field, space occupation, and ball circulation. During the SSGNE, the physical education teacher stood next to the field and provided new balls when necessary but did not provide any verbal encouragement.

The participants were asked to compete at a maximum effort throughout the exercise and to maintain possession of the ball for the longest time possible. The number of ball touches authorized per individual possession was fixed at two touches to ensure all participants engaged in the SSG.

2.6. Physical Activity Enjoyment

The 18-item Physical Activity Enjoyment Scale (PACES) was used to measure positive affect associated with involvement in physical activities in college students [24]. Students were asked to rate "how you feel at the moment about the physical activity you have been doing" using a 7-point bipolar rating scale from 1 (It is very pleasant) to 7 (It is not fun at all). Eleven items are reverse scored. The physical enjoyment score was generated by summing the item scores, which yielded a possible range of 18 through 126. Higher PACES scores reflect greater levels of enjoyment [24]. The PACES scale obtained a Cronbach's α value of 0.90. Players answered individually the questionnaire.

2.7. The Profile of Mood State

Profile of mood states (POMS) [25] questionnaire was used to measure mood disturbance. This self-report questionnaire consists of 65 adjectives designed to assess 6 states (Tension-anxiety, Depression-dejection, Anger-hostility, Vigor-activity, Fatigue-inertia, and Confusion-bewilderment). Responses to each item rated on a 5-point Likert scale (0 indicates "Not at all" and 4 indicates "extremely"). The six subscales of POMS can be combined into a Total Mood Disturbance (TMD) score by summing the T scores for the five negative mood subscales and subtracting the T score for positive mood state, and adding a constant of 100 in order to prevent negative numbers [TMD = ((Anger + Confusion + Depression + Fatigue + Tension) − Vigor)) + 100]. The Cronbach's α ranged from 0.85 to 0.91. Players answered the questionnaire individually.

2.8. Physiological Measures

During the SSG, the HR was continuously monitored using individual HR monitors (Polar Team Sport System, Polar-Electro OY, Kempele, Finland) noted every 5-s intervals. To decrease HR recording error, all students were regularly asked to check their HR monitors throughout the exercise. HR data were, therefore, expressed both as a percentage of HRmax (%HRmax) considering the maximal HR estimated in YYIRT. The mean HR for each bout was calculated to attain the means of the 4 bouts of SSG (HRmean). The %HRmax was calculated by the following formula for each form of SSG, %HRmax: (HRgame/YIRT-HRmax) × 100 [18].

For the determination of [La]b, blood samples were collected 3 min post-training passive recovery [14]. Blood samples, taken from the fingertip of the index finger, were analyzed by a validated portable lactate monitor (Lactate Pro, Arkray, Japan) [14].

2.9. Rating of Perceived Exertion

The RPE of each subject was recorded immediately on completion of each session using the 10-point RPE scale proposed by Foster et al. [26] to assess the internal intensity of each SSG intervention. The RPE was measured using a standardized question "How was, and how did you feel the exercise". This tool has already been used in previous studies [7]. The validity and reliability of this scale to estimate the intensity of effort was also confirmed in previous studies [27]. Players answered individually to avoid hearing the scores of their colleagues. Moreover, players were previously familiarized with the scale to maximize the accuracy of the answers.

2.10. Statistical Procedures

All data are expressed as mean ± standard deviation (SD). Before using parametric tests, the assumption of normality was confirmed using the Kolmogorov–Smirnov test. Moreover, the homogeneity of the sample was also tested and confirmed using Levene's test. Each testing protocol (SSGE and SSGNE) was repeated twice, and the two resulting variables were averaged for analysis. Student's paired t-test was used to compare physiological, physical enjoyment and RPE responses elicited by both filed tests (i.e., [La]b, %HRmax, RPE and PACES scores). The magnitude of change expressed as Cohen's d coefficient was employed to give a rigorous judgment about the differences between SSGE and SSGNE [28]. The scales of magnitude were considered trivial, small, medium, and large, respectively, for values of 0 to 0.20, >0.20 to 0.50, >0.50 to 0.80, and >0.80 [29]. As for the mood state, a two-way analysis of variance (ANOVA) was used to examine the effect of the "Training session" (SSGE or SSGNE), "Time" (pre-and post-training session), and their interaction (training modality x effort) on the POMS score. When a significant interaction effect was found, the analysis was completed with a post hoc test. Analyses were conducted using the Statistical Package for the Social Sciences (v20.0, SPSS, SPSS Inc., Chicago, IL, USA) and the level of significance was set at $p < 0.05$.

3. Results

3.1. Physiological Response

Results presented in Table 1 showed significant differences in the %HRmax, [La]b, and RPE variables between SSGE and SSGNE (all, $p < 0.001$).

Table 1. Comparison of physiological variables and rating of perceived exertion(RPE) between small-sided games with verbal coach encouragement (SSGE) and small-sided games without verbal coach encouragement (SSGNE).

Variables	SSGE	SSGNE	CI95%	ES	Rating
%HRmax (beat.min^{-1})	88.06 ± 2.25	84.31 ± 2.24 ***	−4.39, −3.10	1.73	Large
[la]b (mmol.l^{-1})	4.43 ± 0.91	3.62 ± 0.6 ***	−1.12, −0.50	1.01	Large
RPE	7.62 ± 0.79	6.62 ± 0.81 ***	0.66, 1.33	1.69	Large

%HRmax: percentage of maximal heart rate; [La]b: blood lactate concentration; RPE: rating of perceived exertion; CI95%, confidence interval of the differences; ES: effect size *** $p<0.001$.

3.2. Physical Enjoyment

The physical enjoyment score was significantly higher ($p < 0.001$, ES = 1.45) in SSGE compared to SSGNE (Figure 2).

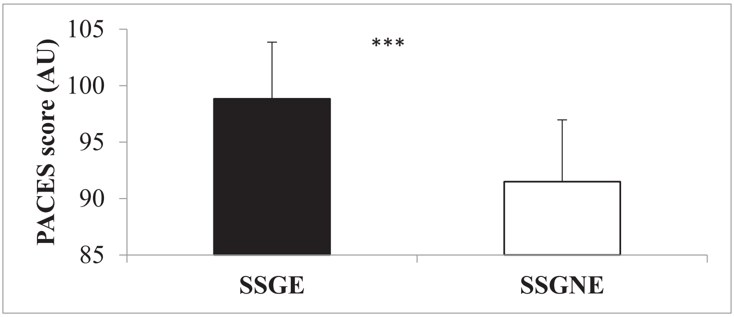


Figure 2. Comparison of perceived physical activity enjoyment between small-sided games with verbal coach encouragement (SSGE) and without verbal coach encouragement (SSGN). *** $p < 0.001$.

3.3. Mood State

Concerning the POMS scores, a significant main effect of Time and a significant main effect of the Training session on TMD (Table 2) was observed. Bonferroni post hoc comparisons revealed that the score of TMD decreased significantly for SSGE and SSGNE (Figure 3).

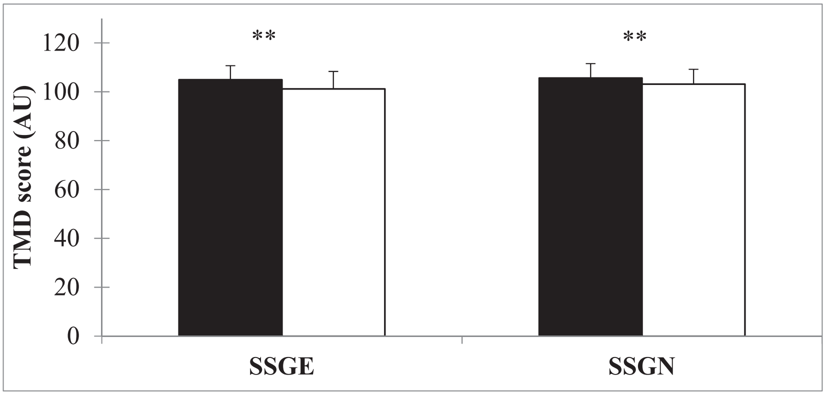


Figure 3. Mean profile of mood states (POMS) score (AU) for both small-sided games training (SSGE) and (SSGNE) collected before (pre-training) and after (post-training) each session. TMD: total mood disturbance. ** $p < 0.01$.

Table 2. Results of the ANOVA with 2 × 2 repeated measures [Training session (SSGE) and (SSGNE)] × Time (pre-and post-test).

Variables	Training Session		Time		Interaction	
	F (1,15)	**ES**	**F (1,15)**	**ES**	**F (1,15)**	**ES**
TMD	5.77 *	0.29	62.89 ***	0.80	1.16	0.07

ES: effect size, * $p < 0.05$; *** $p < 0.001$.

4. Discussion

This study assessed the effect of the sports teacher's verbal encouragement on the physiological responses, mood state, and physical enjoyment of soccer players during SSGs. The main findings of the present study are that (1) SSGE increased physiological and internal intensity to a greater extent than SSGNE, (2) the physical enjoyment was greater after SSGE, and (3) SSGE resulted in a more positive mood state when compared to SSGNE.

The present study showed that the verbal encouragement of a sports teacher positively impacts the physiological responses measured during SSGs. The encouragement variable leads to an increase in the values of %HR max, [La]b, and RPE (4.45%, 22.37%, and 15.10%, respectively). These results suggest that the sports teacher's encouragement can motivate players to provide a high level of physical engagement, maintain a high work rate during the play situations, and keep possession of the ball for the longest time possible.

Several studies have shown that coach encouragement increases physiological responses and internal intensity, especially in young players [4,6,8,12]. For example, Brandes and Elvers [30] mentioned the effectiveness of verbal encouragement in promoting training intensity and training adherence. This result is also in agreement with those of Rampinini et al. [15], who examined the influence of verbal encouragement on various physiological aspects in several SSG formats (3 vs. 3, 4 vs. 4, 5 vs. 5, and 6 vs. 6) on small, medium-sized, and large pitches. They showed that HR, [La]b, and RPE values were significantly greater in SSGs when verbal coach encouragement was given. Moreover, Selmi et al. [6] compared the effects of coach encouragement during 4 vs. 4 SSGs on physiological responses in youth soccer players. Researchers have also indicated that RPE and %HR max were higher in SSGs with encouragement compared to SSGs without encouragement.

In addition, Edwards et al. [31] reported that verbal encouragement in endurance activities resulted in large improvements in performance and motivation, which has important implications for health, adherence, and physical performance using a practical intervention. These findings suggest that the physiological responses and the internal intensity induced by SSGs might vary according to the motivation of the participants, which can be influenced by a sports teacher or coach.

Overall, the present study results demonstrated that SSGE elicited higher aerobic and anaerobic contributions to energetic demands and perceived exertion, thereby confirming the importance of encouragement in the development of students' and athletes' physical fitness.

Physical enjoyment represents a positive affective reaction that allows physical activity to be associated with positive feelings [19,32,33]. Many studies have highlighted the importance of the PACES scale in evaluating physical enjoyment in athletes [19,21,34]. The present study indicates that encouragement has a positive effect on physical enjoyment. The results of the present study are consistent with those reported by Kilit et al. [5], who also indicated that verbal encouragement has a positive effect on participants' physical enjoyment and commitment to engage in physical exercise. We believe that motivational factors associated with the SSGE condition explain the importance of physical enjoyment. Specifically, participants in this study were motivated by the physical education teacher's verbal encouragement. Similarly, Midgley et al. [17] indicated that encouragement during training exercises is expected to increase athletes' motivation and improve positive behavior. In this regard, encouragement during SSG is linked to positive emotional responses to training exercises and is one of the main reasons that motivate participants to contribute to physical activity [4,6,17,31,35].

Another important finding concerns the comparison of mood states (POMS) between the SSGE and SSGNE groups. POMS is commonly used to evaluate the psychological state of participants during physical activity and training [36,37]. Selmi et al. [18] reported that during integrated training, players generally report significant improvements in their mood state. In the present investigation, tension and TMD were positively affected during both SSG conditions (Figure 1). Selmi et al. [18] indicated that high-intensity interval training worsens the mood and is associated with higher scores for anxiety, fatigue, and TMD when compared to SSGs that ensure a mood balance with no change in POMS scores. The stability of the POMS scores in this previous study may be due to the study population (professional soccer players) who participated in the study protocol. Based on the above, it can be suggested that specific soccer training activities decrease negative moods among participants.

Performing SSGE causes a decrease in TMD (as seen in the SSGNE). This indicates that specific training with the ball elicits an increase in positive statements and an increased positive mood. These results are consistent with those presented by Sparkes et al. [38], who examined the influence of SSGs on mood. Decreases in TMD after SSG are usually related to higher vigor and lower tension.

We believe that motivational factors related to the SSGs in the present study may explain the decrease in TMD. Importantly, soccer-specific training leads to mood state improvements. These results suggest that the presence of the ball enhances mood state among participants [6,7,14,18].

The present study indicated that a physical education teacher's behavior could influence students' behavior, exercise intensity, affecting psychophysiological and affective responses. Similarly, Jiménez et al. [39] showed that an autocratic teaching style (characterized by negative coach feedback) negatively affected young swimmers' biological responses, motivational climate, and self-confidence. This kind of teaching style can lead to an affective deterioration. The authors also indicated that swimmers enjoyed training more, were more motivated, and perceived greater personal effort when they were directed by a coach who positively encouraged them. These findings indicate that performance is linked to a positive style of teaching (i.e., teacher's positive feedback) [39].

Several limitations must be taken into account when interpreting the present results. First of all, the study sample was small due to the difficulty of recruiting a large number of homogeneous participants. Moreover, the testing utilized only one SSG format and a single age cohort of soccer specialist students. Future studies comparing the two SSG conditions should use different levels of determining variables of SSG intensity (i.e., different game rules, duration of each bout, pitch size, number of players, and the presence of goalkeepers) and participants of different ages to extend the applicability of the present findings. Finally, it would be interesting to associate these responses with technical aspects of time-motion parameters (i.e., distance covered, run at high intensity, number of sprints, etc.) because physical aspects are important markers of performance.

This study was conducted in a real training area with youth soccer players, thereby providing practical implications. To the best of our knowledge, this study is the first to compare technical aspects, physiological responses, mood state, and physical enjoyment during SSGs among soccer specialist students. The verbal encouragement of a physical education teacher/coach can be considered an essential factor during specific football training sessions, as it induces physiological, psychological, and technical responses. For this reason, physical education teachers and soccer coaches should verbally encourage their students and players during specific training exercises to increase the game intensity, improve players' physiological responses, and create positive psychological states.

5. Conclusions

SSGE and SSGNE induce similar improvements in total mood disturbance and tension, while SSGE induced more intense physiological responses, internal intensity, enjoyment levels, and positive moods than SSGNE. The results suggest that physical education teachers should increasingly deliver verbal encouragement to enhance the motivation and commitment of students to engage in physical training. Verbal encouragement can be considered as an important variable used in order to improve game performance, physiological responses, and psychological states during specific soccer sessions in school students.

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Athlete-Specific Neural Strategies under Pressure: A fNIRS Pilot Study

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Abstract: (1) Background: Stress and pressure during competition and training impair athletes' performance in sports. However, the influence of mental stress on the prefrontal cortex (PFC) functioning in an athlete during the visual simulation task is unknown. The purpose of this pilot study was to investigate hemodynamic responses during the visual-simulation task that induces pressure and stress using functional near-infrared spectroscopy. (2) Methods: Ten archers and ten non-athlete collegiate students performed a visual-simulation task. Participants' current stress levels were collected using a visual analog scale before and after the task. Average oxygenated hemoglobin (HbO), deoxygenated hemoglobin (HbR), and total hemoglobin (HbT) levels and their variability (standard deviation (SD) HbO, SD HbR, and SD HbT) were computed to compare the neural efficiency between athlete and non-athlete. (3) Results: In general, both groups exhibited increased stress levels after the simulation task, and there was no group difference in overall average hemodynamic response from PFC and dorsolateral prefrontal cortex (DLPFC). While the average hemodynamic response level did not differ between groups, variability in hemodynamic responses from the archer group showed a more stable pattern than the non-athlete group. (4) Conclusion: Under this experimental setting, decreasing the variability in hemodynamic responses during the visual simulation, potentially via stabilizing the fluctuation of PFC, was characterized by the stress-related compensatory neural strategy of elite archers.

Keywords: archery; stress; noncontact; simulation training; fNIRS

1. Introduction

Studies investigating human stress responses have received extensive attention, because high levels of stress over long durations relate to the development of mental and physical health problems such as cardiovascular disorders [1,2], anxiety disorder [3], and obesity [4]. Some studies have demonstrated that moderate levels of stress are linked to an improvement in cognitive abilities [5] and physical performance [6], whereas high levels of stress are associated with an impairment of cognitive functions, particularly in cognitive flexibility [7] and executive functions [8]. Moreover, both outstanding physical abilities and cognitive skills that are susceptible to stress levels—such as perception, attention, working memory, and decision-making—are crucial for success at the highest level of sports. However, our understanding of the neurocognitive characteristics of elite athletes in sports is much less understood. In addition, the speed of information processing and movement artifacts still poses many obstacles for studying the neurological and neurophysiological processes regarding the cognitive skills in elite sports. Due to the lack of robust technologies, investigations involving high levels of cognitive processing important for sports performance have to date been restricted to the laboratory, involving responses to stimuli that are not closely related to sports performance [7,9]. Therefore, this study aimed to investigate the cortical activity during archery simulation training using functional near-infrared spectroscopy (fNIRS) to gain a better understanding of the involvement of prefrontal brain regions in stressful situations.

Brain studies associated with sports often use classic expert–novice paradigms to investigate the neurological activity of human beings with a high level of motor skills. Typically, expert–novice paradigm studies adopt two types of experimental tasks: (1) tasks unrelated to motor cognition that determine the effect of exercise training on an individual's brain function and (2) tasks related to motor cognition, including motor execution, motor imagery, and action observation. These techniques are extremely vulnerable to motion artifacts, which poses challenges in investigating cortical activity during performance. To avoid these movements-associated issues, Del Percio and Babiloni [10] conducted a study on elite karate, elite fencing, and non-athletes, comparing the neural activity during their engaging a micro-motor control task (e.g., monopodalic upright standing). Other studies focused on brain activity regarding lower limb movement instructed participants to complete simple motor tasks such as foot or toe swinging and foot-pressing on objects [11,12]. However, these types of tasks are irrelevant to limb movements used in sports settings.

To overcome the impact of motion artifacts on the acquisition of signals, studies focusing on motor imagery or action observation have been conducted instead of motor execution tasks. Specifically, to confirm and extend the evidence for experience-dependent plasticity of cortical activity on action-observation and motor imagery, a study compared the task performance of experienced ballet dancers and non-dancers [13], while another study recruited basketball players, volleyball players, and non-athletes to complete motor imagery tasks [14]. These studies suggested that familiarity and expertise play a crucial role in cortical activity and are important factors that must be considered for task difficulty and engagement [15,16]. In addition, according to the simulation theory [17], there is a functional linkage between motor execution and the simulation of the action. Because the presence of activity in the motor system during the simulation—such as motor imagery and action observation—would place the action representation into the motor execution, the simulation theory postulates that the facilitation of the motor system during motor imagery, action observation, and motor execution are functionally equivalent [18,19].

The brain efficiency hypothesis suggests that an effective task performer exhibits a substantial decrease in brain glucose metabolic rates [20]. In particular, the reduction in neural activity and experience-dependent changes in energy usage indicates an increase in the efficiency of neural function [21]. This hypothesis has been supported by studies focused on the motor cognitive tasks of elite athletes. Previous studies revealed that the amplitude of task-related cortical areas in elite shooters [22] and professional ballet dancers [13] were lesser than the non-athlete control during the task performance. In addition, the brain efficiency hypothesis was also identified by the variability of hemodynamic responses during the tasks requiring a substantial cognitive effort. Specifically, when an individual performed a motor task with an additional cognitive load that induced mental stress, the variability of hemodynamic responses increased compared to the resting state [23–25]. Based on the brain efficiency hypothesis, an elite archer would have a characteristic of cortical activity under pressure and stressful situations. Because elite archers are frequently exposed to stress and pressure during training and completions, we hypothesized that the elite archer would likely show a lower level of hemodynamic responses and a more stable pattern in task-related areas (e.g., prefrontal cortex or dorsolateral prefrontal cortex) during the pressure-inducing simulation task compared to the control group. Because elite archers are frequently exposed to the pressure situation during the match or training, such experiences are expected to contribute to a reduction in cortical activity [21,22].

Regarding the continuous progress of brain imaging technology, fNIRS has attracted much attention to measure and monitor the hemodynamic responses in athletes' cerebral cortexes. It is noninvasive and enables investigations into cortical response during task performance [26]. The experimental task in this study included a pressure-inducing situation that simulates scenarios from the actual competition. This situation also represented internal cognitive processes such as error detection, problem-solving, and motor planning for optimal outcomes. To determine the specific brain activity of elite archers, this pilot study compared the differences in prefrontal cortex (PFC) activation between elite archers and non-athlete collegiate students who participated in the archer class completing the visual-simulation task.

2. Materials and Methods

2.1. Participants

The Institutional Review Board at the Korea Institute of Sport Science approved the procedures (KISS-1905-015-01), and participants provided written informed consent before participation in the study. Ten elite archers (4 males and 6 females, M age = 26.8, SD = 4.1 years) that are currently registered in the Korea Archery Association and ten collegiate students (7 males and 3 females, M age = 23.2, SD = 3.6 years) that participated in the archery class were recruited and were right-hand dominant according to self-reports. None of the participants had a previous history of cardiovascular, pulmonary, neurological psychiatric, or other diseases that might have influenced the experimental results. All participants were asked not to consume caffeinated food or drink at least 24 h before the initiation of the experiment.

2.2. Experimental Equipment and Simulation Film

The simulation film used to create tension and stress for this study was recorded using a digital video camera (Sony hrx-nx30n, SONY, Minato, Japan and GoPro 4, GoPro Inc., San Mateo, CA, USA) set to show the performer's perspective while performing under pressure. Clips were edited using Final Cut Pro software (Macromedia, Inc., San Francisco, CA, USA). Filming was conducted in the archery stadium of the national training center. To emulate the stressful and pressure situation from the performer's perspective as closely as possible, the archer's heartbeat as well as background sounds, including those of spectators, were included in the video clips. A panel of three specialist archery coaches selected the scenes most representative of the stressful situation and behavior of archers while shooting. The simulation film consisted of the following three scenes. (Scene 1) In the first round with the shooting time almost over, the stopwatch was highlighted to increase the participant's pressure and tension to ensure the athlete could not shoot and set the bow down with a buzzer announcing the end of the shooting time. (Scene 2) In the third set of the tournament, the athlete is losing 0-4 and shooting despite the bow sight shaking due to pressure and tension, scoring three points by mistake. (Scene 3) A situation of a 5-5 shoot-off in which only one last shot is left to decide the winner; the archer is shooting in a situation where they can hear the sound of their heartbeat due to great pressure. All scenes were filmed using an action camera to ensure the participants could be perceived from the archer's viewpoint, and high-speed recording was used to show the trajectory of the bow flying. Before the start of each scene, a visual cue was presented for two seconds and then short statements that describe each situation were presented for 7 s. The duration of the simulation film was 140 s.

2.3. Procedures

Prior to the experiment, participants were asked to rate the amount of perceived mental stress before watching the simulation film to investigate the changes in stress levels. The mental stress level was rated with a numerical rating scale with endpoints set at "no mental stress at all" (=0) and "the most intense mental stress imaginable" (=10). Participants performed another stress rating at the end of the experiment.

In the experiment, the participants were asked to rest for 30 s and were then shown a visual cue on a screen for two seconds representing the upcoming task (simulation film). During the simulation task, the athlete group was instructed to consider that they were experiencing a real match, and the collegiate group was instructed to consider that they were going through an intramural game or final test in the archery class. During the task, the participants' hands were placed on the armrest of a chair with their hands pronated, feet close together, and bodies upright. The participants were not allowed to move their bodies or heads during the experiment.

2.4. Channel Configuration

In this study, a near-infrared multi-channel continuous wave system (NIRSIT, OBELAB Inc., Seoul, Korea) with a sampling rate of 8.1138 Hz was employed to measure neural hemodynamic responses of the PFC via 24 emitters and 32 detectors. The device has an active detection sensor with a total capacity of 204 channels—48 of which were used in this study—covering the entire PFC area. In order to ensure that the fNIRS device is located according to the anatomical brain structure of the participants, the frontopolar zone (FPz) was used to assure comparability between all tested participants. The center of the device (Figure 1, red dot) was matched to the subject's FPz and the bottom-most part of the device was right above the eyebrows. Based on the Brodmann area (BA), 48 channels were recorded on both sides of the dorsolateral prefrontal cortex (DLPFC, BA 9, 46), ventrolateral prefrontal cortex (VLPFC, BA 44, 45, 47), frontopolar prefrontal cortex (FPC, BA 10), and orbitofrontal cortex (OFC, BA 11). Figure 1 shows the locations of emitters and detectors with a reference point FPz (red dot) and the eight areas in this study. The wavelengths used for detecting two chromophores (oxygenated hemoglobin; HbO and deoxygenated hemoglobin; HbR) were 780 and 850 nm, respectively. For the best spatial resolution, a 3 cm distance separating the laser and detector pairs was used.

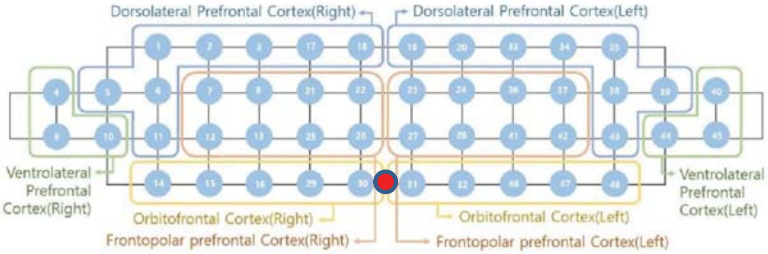

Figure 1. Schematic representation of the functional near-infrared spectroscopy (fNIRS) channel configuration positioned across the prefrontal cortex (PFC). PFC is divided into eight sub-area based on the Brodmann area. The red dot indicates the center of the device that located the participant's frontopolar zone (FPz).

2.5. Data Pre-Processing

Recorded hemodynamic responses were pre-processed and analyzed using the NIRSIT analysis tool. First, the raw NIRS signal was converted into an optical intensity signal using two fourth-order Butterworth filters (low and high-pass) with cutoff frequencies of 0.5 and 0.005 Hz, respectively, to remove systemic responses caused by heartbeat and reduce slow-wave drift caused by the NIRS system [27]. The cut-off frequency of the low pass filter was sufficient to capture the fluctuation of the task-induced hemodynamics which was 20 times higher than the task period (0.025 Hz). The bad quality channels, decided by signal-to-noise ratio as <30 dB, were rejected before extraction of hemodynamics data to prevent misinterpretation and visually inspected. The optical intensity signals were transformed into the time series of HbO and HbR concentration changes using the modified Beer–Lambert law (MBLL) [28]. The concentration in hemodynamic responses (HbO, HbR, and HbT) during the experimental task is normalized to a resting baseline (−5 to 0 s) immediately preceding the onset of the task. Mean HbO levels of the baseline trials were subtracted from the mean HbO levels from the task trials to obtain normalized HbO levels from the bilateral PFC sites. The same processes were applied to HbR and HbT levels.

2.6. Measurements

HbO, HbR, and total hemoglobin (HbT) levels were obtained throughout the experiment. From the fNIRS data, three different types of dependent measures were obtained. To investigate the alteration pattern of HbO, HbR, and HbT levels in the PFC over the entire experimental task, the overall average value and standard deviation of HbO, HbR, and HbT were used for statistical analysis. In addition, the mean HbO, HbR, and HbT level obtained from 0 to 39 s across each scene was used to compute average HbO, HbR, and HbT values. As such, three average values (S1, S2, and S3) were present over 140 s of the experimental task. The average HbO, HbR, and HbT represented the mean hemodynamic response in each area during the experimental trials, whereas the SD HbO, HbR, and HbT represented the variability of that same signal during the simulation trials in each specific area [23]. A visual analog scale (VAS) for the current mental stress level was collected before and after the simulation task to assess the successful manipulation of mental stress levels. The VAS was used to reliably evaluate the effectiveness of the stress protocol [29].

2.7. Data Analysis

All dependent measures were subject to an aligned rank transformation [30] to conduct a mixed model analysis of variance (ANOVA) because of the small sample size and violation of normality assumption. A two-way mixed model ANOVA was used to determine the effect of group (athlete and non-athlete collegiate: a between-subject factor) and test (pre and post) on perceived mental stress level. A two-way mixed model ANOVA was conducted to determine the effects of group and PFC area (left and right DLPFC, VLPFC, FPC, and OFC: within-subject factor, eight levels) on the overall average and SD of HbO, HbR, and HbT values. Finally, a three-way ANOVA was conducted to determine the effects of group, scene (S1, S2, and S3: a within-subject factor, three levels), and hemisphere (left and right: a within-subject factor) on average HbO, HbR, and HbT values of the interest area (DLPFC). Statistical significance was set at $p < 0.05$. A significant main effect and interactions were examined using pairwise comparisons with Bonferroni corrections, as required. Summary data are presented as mean ± standard error (SE). The results of the experiment were visualized using Matlab (R2015b), R (v.4.0.2), and RStudio (v.1.0.136) (R and RStudio, Inc., Boston, MA, USA).

3. Results

The VAS data from pre- and post-simulation tasks were analyzed to test the effectiveness of the stress manipulation. The results revealed that a significant effect of test ($F_{(1,\,18)} = 199.03$, $p < 0.001$, $\eta_p^2 = 0.97$) was found on the VAS stress scores (Figure 2a). The main effect of group and its interaction with test on VAS were not found to be significant (*ps* > 0.29). The VAS values increased across the experiment, and the simulation task was found to successfully increase perceptions of stress in both athlete and non-athlete collegiate students.

To analyze the variability in cortical activity (SD for HbO) during the experimental task, SD HbO values were analyzed with two-way mixed model ANOVA (Figure 2b). The analysis revealed significant group ($F_{(1,\,18)} = 23.48$, $p < 0.01$, $\eta_p^2 = 0.55$) and area effects ($F_{(7,\,126)} = 8.20$, $p < 0.001$, $\eta_p^2 = 0.19$). The non-athlete collegiate group exhibited significantly higher SD HbO levels than the athlete group. Post-hoc analysis of the PFC area revealed that both sides of the DLPFC variabilities were higher than the left side of the OFC (*ps* < 0.003), and the left side of the DLPFC was the least stable area in the PFC (*ps* = 0.001–0.045). Other PFC areas were not significantly different between each other (*ps* = 0.06–0.97). The effect of group interaction with the PFC area was not found to be significant for the variation of HbO ($p > 0.19$). The analysis of SD HbR revealed significant group ($F_{(1,\,18)} = 14.73$, $p < 0.01$, $\eta_p^2 = 0.43$) and area effects ($F_{(7,\,126)} = 8.67$, $p < 0.01$, $\eta_p^2 = 0.32$). The non-athlete collegiate group exhibited significantly higher SD HbR levels than the athlete group (see Supplementary Figure S1a). Post-hoc analysis of the PFC area revealed that both sides of the DLPFC variabilities were higher than the left side of the OFC (*ps* < 0.01), and the left side of the DLPFC variability was higher than right OFC and left

FPC (*ps* = 0.01, 0.04). Other PFC areas were not significantly different from each other (*ps* = 0.06–0.88). The interaction between the group and PFC area was not significant ($p > 0.22$). The analysis of SD HbT revealed a significant group ($F_{(1,\,18)} = 23.48$, $p < 0.01$, $\eta_p^2 = 0.45$) and area effects ($F_{(7,\,126)} = 8.20$, $p < 0.01$, $\eta_p^2 = 0.31$). The non-athlete collegiate group exhibited significantly higher SD HbT levels than the athlete group (Supplementary Figure S1b). Post-hoc analysis of the PFC area revealed that both sides of the DLPFC variabilities were higher than the left side of the OFC (*ps* < 0.01), and the left side of the DLPFC variability was higher than both sides of OFC and FPC (*ps* < 0.01) and left VLPFC ($p = 0.03$). Other PFC areas were not significantly different from each other (*ps* = 0.13–0.98) and the effect of group interaction with the PFC area was not significant for the HbT variability ($p > 0.12$).

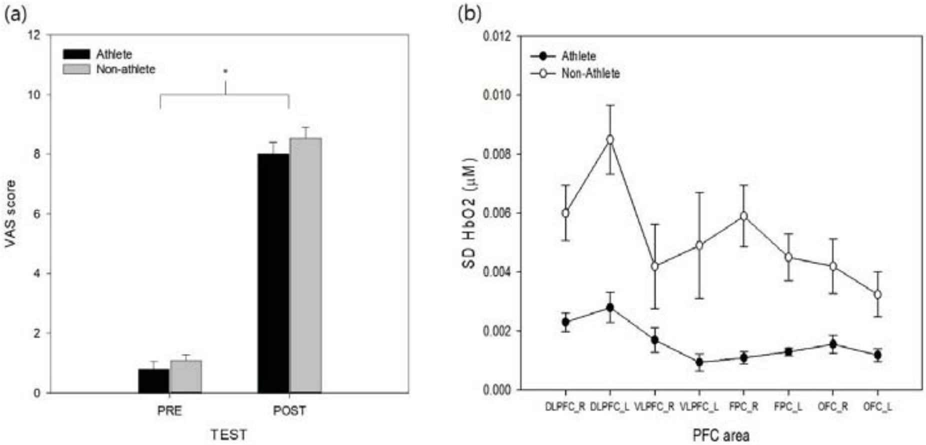

Figure 2. (**a**) Changes in perceived mental stress levels of athlete and non-athlete group before and after the simulation trials. (**b**) Standard deviation of oxygenated hemoglobin (HbO2) level in PFC area from athlete (●) and non-athlete (○) group during the simulation task. Error bar represents SE.

The overall average HbO, HbR, and HbT levels throughout the experimental trial were analyzed with two-way mixed-model ANOVA to explore the differences in mean oxygenation level in each area (Figure 3). The analysis of HbO level showed a significant main effect of the PFC area ($F_{(7,\,126)} = 2.76$, $p < 0.01$, $\eta_p^2 = 0.13$). Post-hoc analysis of the main effect revealed that the cortical activity of the left VLPFC was significantly higher than that of the right VLPFC and right DLPFC ($p < 0.05$). Other cortical areas did not show a significant difference in HbO level (p = 0.06–0.93). The main effect of group ($p > 0.56$) and interaction between group and PFC area were not significant ($p > 0.43$). The analysis of HbR levels did not show a significant main effect of group ($p = 0.58$), PFC area ($p = 0.21$) and its interaction ($p = 0.88$, see Supplementary Figure S2a). The analysis of HbT level, also, did not show a significant main effect of group ($p = 0.97$), PFC area ($p = 0.67$) and its interaction ($p = 0.80$, Supplementary Figure S2b).

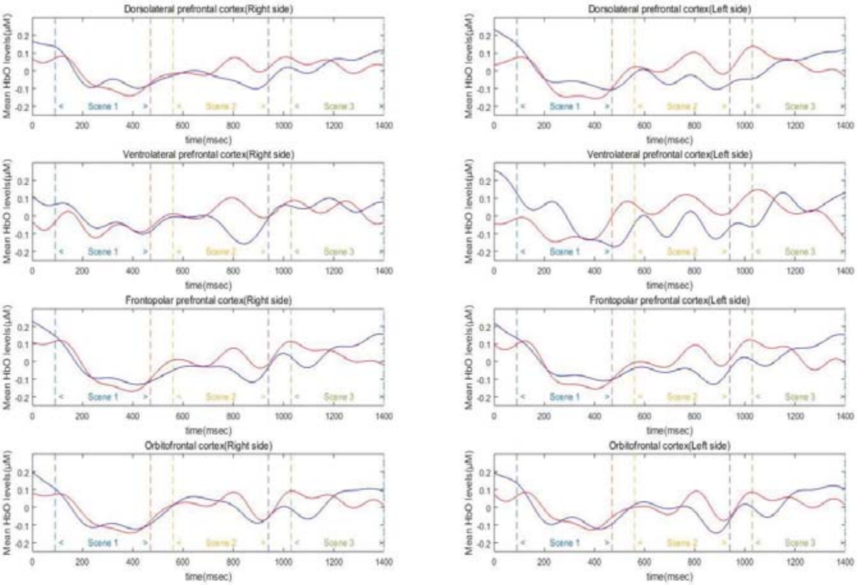

Figure 3. Mean HbO levels in PFC area (both sides of dorsolateral prefrontal cortex (DLPFC), ventrolateral prefrontal cortex (VLPFC), frontopolar prefrontal cortex (FPC), and orbitofrontal cortex (OFC)) of athletes (blue line) and non-athlete collegiate (red line) during the simulation task.

Three-way mixed factor ANOVA was used to test for main and interaction effects of group, hemisphere, and scene on average HbO, HbR, and HbT levels in the DLPFC (Figure 4). The analysis of HbO level in the DLPFC revealed a significant main effect of scene ($F_{(2, 90)} = 35.19$, $p < 0.01$, $\eta_p^2 = 0.89$). Specifically, the DLPFC activation level of Scenes 2 and 3 was significantly higher than that of Scene 1 (*ps* < 0.001), and the difference between Scenes 2 and 3 was not significant ($p > 0.10$). The main effects of group and hemisphere were not significant (*ps* = 0.19–0.71). In addition, the effects of interaction between hemisphere, scene, and group were not found to be significant on the average HbO levels (*ps* = 0.28–0.78). The analysis of HbR level in the DLPFC revealed a significant main effect of scene ($F_{(2, 90)} = 8.08$, $p < 0.01$, $\eta_p^2 = 0.34$). Post-hoc analysis of the scene revealed that HbR level of the Scene 3 was significantly lower than the Scene 1 ($p = 0.01$). Scenes 2 and 3 were not significantly different from each other ($p = 0.91$). The main effect of group ($p > 0.3$), hemisphere ($p > 0.2$) and its interactions were not significant (*ps* > 0.14, Supplementary Figure S3a). Similar to the HbO levels, the analysis of HbT level in the DLPFC revealed a significant main effect of scene ($F_{(2, 90)} = 12.00$, $p < 0.01$, $\eta_p^2 = 0.42$). Post-hoc analysis of the scene revealed that the HbT level of Scenes 2 and 3 was significantly higher than that of Scene 1 (*ps* < 0.01) and Scenes 2 and 3 were not significantly different from each other ($p = 0.92$). The main effect of group ($p > 0.6$), hemisphere ($p > 0.3$) and its interactions were not significant (*ps* > 0.25, Supplementary Figure S3b).

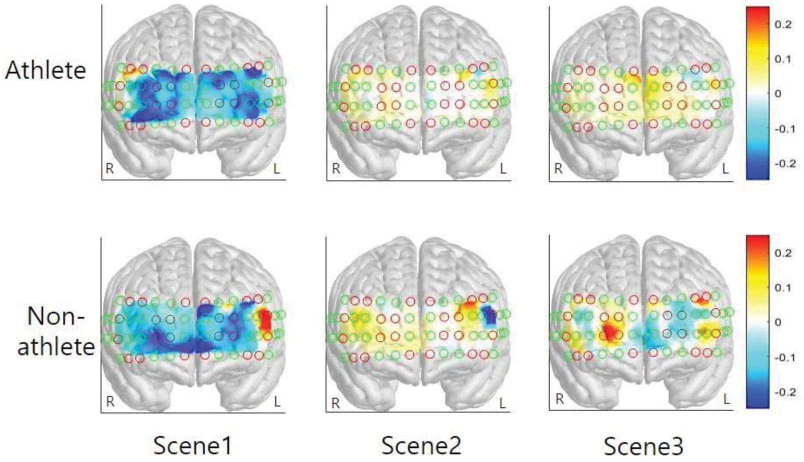

Figure 4. Activation maps illustrate the average HbO level obtained from 0 to 39 s across each scene for the athlete and non-athlete collegiate group. Red and green circles denote the sources and detectors, respectively. R indicates the right hemisphere and L refers to the left hemisphere.

4. Discussion

The present study compared changes in functional hemodynamic responses of the PFC between athletes and non-athletes during pressure-induced visual simulation using fNIRS. Both athletes and non-athletes showed increased mental stress levels after watching the simulation film. The overall average HbO was increased during the simulation in both groups; specifically, activation of the left VLPFC was higher than that of the right VLPFC and right DLPFC. The activation level of other cortical areas (both sides of FPC and OFC and left VLPFC and left DLPFC) were not different to each other. While group differences were not found in the average HbO, HbR, and HbT level, variability in HbO, HbR, and HbT level was found to differ between groups. The cortical activation level of the interest area, both sides of DLPFC, was increased in Scenes 2 and 3. Based on the results of this preliminary study, considerations associated with the main study are discussed in the following section.

The results of the current study associated with average HbO did not show a difference between the athlete and non-athlete groups. According to the brain efficiency hypothesis [20,22], the elite archers were expected to show a neural efficiency compared to the non-athlete. However, the findings are inconsistent with the brain efficiency hypothesis and previous studies. These results imply that both groups experienced a high level of perceived mental stress, as shown in the VAS data in this study. Although athletes are frequently exposed to extreme stress situations and competition, they also have difficulty adapting to these environments. The hemodynamic responses observed in the current study cannot be inferred for all scenarios of the simulation or performance of motor skills. The relatively economic neural activity has been observed as subjects performed a simple key pressing task [22] and a dance sequence imagery task [13]. Some discrepancies between these findings may be explained by differences in the nature of a task, neuroimaging techniques, and training schedules. For instance, stress-inducing photographs are omitted from the process of the event (i.e., injuries, critical coach, and embarrassment due to loss), possibly minimizing the magnitude of prefrontal responses [31]. In contrast, the visual-simulation film in the current investigation was mainly focused on the procedures that induce pressure. Complex and additional cognitive loads modulate the degree of prefrontal activation [32]. Furthermore, enhanced DLPFC activations during Scenes 2 and 3 suggest that these regions are crucial for error detection and online monitoring of performance [32,33]. Unlike Scene 1, participants repeatedly observed performance errors in Scenes 2 and 3. Error detection was likely to be most prominent during Scene 2, scoring 1 point by mistake, in the simulation film possibly accounting for PFC activation. The DLPFC is likely to indirectly contribute to emotion regulation through its interaction with the orbitofrontal cortex and the anterior cingulate cortex, and via these areas, with the amygdala [34]. In addition, the DLPFC is involved in emotion regulation because it plays a key role in the neural network supporting working memory function [35]. Situations such as shooting under pressure that require complex cognitive strategies: multiple sequential error detection processes and conscious control of relevant information are likely to result in profound PFC activations.

The non-athlete collegiate group exhibited significantly greater variability in hemodynamic responses in this study, suggesting that subjects exerted greater cognitive effort during the task. A previous study reported that predominantly greater HbO variability was observed in the PFC during the cognitively demanding task [24]. Subjects in the previous study showed greater HbO variability in the PFC as the reaction time and error rate increased. The authors suggested that between-group differences in neural variability might reflect systematic differences in the level of central nervous system function. In addition, consistent results regarding the variability of HbO from the current study were also reported in the postural control study that assessed the chronic ankle instability (CAI) group and healthy controls' cortical activity in the motor cortex during single-limb stance tasks [23]. The CAI group exhibited greater HbO variability in the supplementary motor area (SMA) than the healthy control group did. The authors interpreted the results that individuals with CAI may use different corticomotor postural strategies, as indicated by the difference in variability of SMA activation. Consequently, the authors asserted that the task required substantial cognitive and motor demand from individuals with CAI, and increased variability in SMA provides evidence for an altered neural mechanism to compensate for the instability [23]. Thus, the variability of HbO values in this study indicated that the archer's cognitive control ability is less susceptible to stress or pressure than the non-athlete counterpart.

The results of the present study's overall average HbO demonstrate profound activation of the left VLPFC. One plausible interpretation of this result is that the subjects viewed the simulation film from the observer's perspective rather than thinking it was what they were experiencing. The VLPFC is instrumental in empathy modulation [36,37]. The VLPFC modulates the activation of emotional systems to support emotion regulation [38]. A previous study suggested that VLPFC laterality is related to emotion processing [39]: enhancement of negative and positive emotions (up-regulations) activates the left VLPFC [40], while inhibition of these emotions (down-regulation) activates the bilateral VLPFC. For example, participants observed others experiencing negative circumstances; the observer might be eligible to empathize with the feelings of others by re-introducing the prevailing negative situation as substantially adverse. Engen and Singer [41] suggested that empathy is modulated by emotion regulation processes via a top-down manner in the VLPFC, which regulates emotional responses. Consistent with this suggestion, a previous fNIRS study reported that left VLPFC activation is directly linked to empathizing with others experiencing negative circumstances [37]. Therefore, this study's participants may have viewed the simulation film from an observer's perspective rather than the performer's perspective.

5. Conclusions

This study is a preliminary study of research to establish an efficient simulation training environment for athletes. In this study, the pressure-induced simulation film was used to investigate the hemodynamic responses of PFC using fNIRS. The participant's mental stress level increased following the observation of the simulation film. Archers showed a lower variability in HbO compared to non-athletes. The overall average HbO level was increased and no group difference was observed during the simulation task. Specifically, the activation level of the DLPFC was higher in Scenes 2 and 3 than in Scene 1. According to the results, the simulation film successfully manipulates the participant's mental stress level and hemodynamic responses. In addition, specific information regarding the stress and pressure-induced situation (e.g., delayed shooting interval, swaying aiming gauge, and missed shot) was identified. While group differences in average HbO levels were not obvious, the variation in

HbO from archers was more stable than that of non-athletes. In addition, the simulation film was created from the performer's perspective as closely as possible. However, hemodynamic responses from the participants were identical to the responses from the observer's viewpoint. To obtain more sound cortical responses as the participants perform the task in the real world, virtual reality (VR) or augmented reality (AR) technique should be considered in future studies. Furthermore, with psychological skill training combined with these techniques, additional positive effects can be expected.

The present study faces some additional limitations, which warrant a comment and which might be improved in follow-up studies. Because this was a preliminary study with a relatively small sample size, the results should be confirmed among larger samples. This study did not use a hemodynamic response function (HRF)-based general linear model (GLM) analysis. To identify a more accurate relative difference between groups or conditions, HRF-GLM analysis is recommended in subsequent studies because fNIRS cannot measure the absolute level of Hb concentration changes. Investigators may also want to explore mental stress responses using physiological factors in subsequent work. Thus, further studies should be aimed at a more diverse sample, as well as different physiological and behavioral responses to improve the generalizability of our results and outcomes.

Supplementary Materials: The followings are available online at http://www.mdpi.com/1660-4601/17/22/8464/s1, Figure S1: Standard deviation of HbR (a) and HbT (b) level in PFC area from athlete (●) and non-athlete (○) group during the simulation task. Error bar represent SE, Figure S2: Mean HbR (a) and HbT (b) levels in PFC area (left and right DLPFC, VLPFC, FPC, and OFC) of athletes (blue line) and non-athlete collegiate (red line) during the simulation task, Figure S3: Activation maps illustrate the average HbR (a) and HbT (b) level obtained from 0 to 39s across each scene for the athlete and non-athlete collegiate group. Red and green circles denote the sources and detectors, respectively. R indicates the right-hemisphere and L refers to the left-hemisphere.

Author Contributions: Conceptualization, I.P. and S.K.K.; methodology, I.P.; software, I.P.; formal analysis, I.P. and Y.K.; data curation, I.P. and Y.K.; writing—original draft preparation, I.P.; writing—review and editing, I.P. and S.K.K.; visualization, I.P.; funding acquisition, I.P. and S.K.K. All authors have read and agreed to the published version of the manuscript.

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Predicting Caregiver Burden in Informal Caregivers for the Elderly in Ecuador

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Abstract: Informal caregivers are the main providers of care for the elderly. The aim of this study is to examine the predictive value of different variables regarding caregivers and their elderly patients with respect to the caregiver's burden. A convenience sample of 688 informal caregivers and 688 elderly people from Ecuador was surveyed. Only households with one caregiver and one elderly person were considered for the study. For informal caregivers, the following standardized measures were obtained: burden (Zarit Burden Interview), neuroticism (Eysenck Personality Questionnaire Revised-Abbreviated, EPQR-A), caregiver's general health (GHQ-12), and social support (modified Duke-UNC Functional Social Support Questionnaire, FSSQ11). For the elderly, we employed standardized measures of cognitive function (short portable mental status questionnaire, SPMSQ), Pfeiffer's test, and functional dependency (Barthel scale/Index, BI). Females were over-represented in caregiving and reported significantly higher burden levels than those of males. In both male and female caregivers, the burden was best predicted by the time of caring, neuroticism, and elderly cognitive impairment. However, some predictors of burden were weighted differently in males and females. The functional independence of the elderly was a significant predictor of burden for male caregivers but not females, while caregiver competence was a significant predictor for females but not males. These variables accounted for more than 88% of the variability in informal caregivers.

Keywords: stress; burden; informal caregivers; burden; elderly; gender differences; neuroticism; competence

1. Introduction

Stress is an intense and unusual stimulus elicited by the presence of a threat, or any other circumstance or event that an individual perceives as adverse [1]. There are some professions that elicit a large stress response, modifying the homeostasis of the natural organism. In professions such as the military, police, firefighting, and even among elite athletes, individuals frequently experience stress responses beyond the body's natural limits [2–4]. However, stress is not only present in these professions; medical personnel, drivers, journalists, and even teachers and students exhibit elevated stress levels as a result of their exposure to their work context [5–7].

As an acute response, stress is designed to maintain the physical integrity of the subject; however, continuous exposure to stress may result in psychopathologies such as anxiety, depression, post-traumatic stress disorder and burnout [8].

In this respect, stress-related health issues as a result of the burden of providing care are well-documented in informal or family caregivers [9–13]. Moreover, the risk in the informal caregivers of the elderly is increasing since the general population age is steadily increasing. Providing care to the elderly is an important source of chronic stress considering the demands of providing care to elderly patients and the lack of resources of informal caregivers, including formal training.

In the context of caregiving, the stress model underlines the importance of the perceived lack of control and the psychological stress involved in the situation of caring compared with the objective amount of expected burden as a result of the degree of mental impairment or dependency of the elderly patient [14]. Some authors suggested that the time spent providing care, the caregiver's age, their lack of social support, the cognitive impairment of the elderly, and the caregiver's neuroticism are important factors [15,16]. Previous studies also found differences in the amount of perceived burden in caregivers [17]. However, most studies focused on caregivers of people with dementia [18–20] or other mental disorders [21].

In sum, the best predictors of burden in informal caregivers of the elderly in Ecuador remain understudied and elusive. The objective of this study is to analyze the predictive value of variables regarding caregivers and their elderly patients with respect to the burden of informal caregivers of the elderly. Furthermore, gender differences were analyzed. To our knowledge, this is the largest study attempting to predict burden in informal caregivers in Ecuador. This research could contribute to the development of interventions aimed at improving the well-being of caregivers of the elderly.

2. Materials and Methods

2.1. Participants

A convenience sample of 688 family caregivers (mean age = 49.1 ± 14.6; 79.8% females) and their respective 688 elderly patients (mean age = 80.8 ± 9.2; 60.5% females) from eight regions of Ecuador were recruited for this study. All participants were selected from the user referral lists of local retirement associations and centers associated with the Ministry of Social and Economic Inclusion (MIES). As inclusion criteria, all caregivers were relatives of their elderly patients, and their role was that of primary caregiver without receiving any compensation for this service, taking on responsibility for any decisions and for their patient's well-being for at least 12 months. None of the caregivers was receiving specialized psychological support at the time of assessment, and they were living at home with the elderly people. All subjects had given their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki and was approved by the local committee at the Public University of Loja (code 05-04/02/2019).

2.2. Measures

All caregivers were interviewed in one session divided into two parts: a sociodemographic questionnaire (age, gender, time providing care, marital status, ethnic, education level), and a psychological protocol using instruments developed and/or validated in Spanish. Specifically, the following standardized measures were included in this survey:

The Zarit Burden Interview [22] consists of 22 items measuring burden in caregivers. Participants respond using a Likert scale, ranging from 1, "never" to 5, "always". Scores range from 22 to 110. One example item is "I think people I am providing care to are asking for more help than they need". The internal consistency for this study was high, with a Cronbach's alpha of 0.92.

CUIDAR [23] consists of 20 items assessing the caregiver's competency to provide care. Participants respond using a Likert scale ranging from 0, "never" to 3, "always or almost always". Scores range from 0 to 60. An example item is "I know how to monitorize the health condition of the person to whom I provide care". The internal consistency for this study was high, with a Cronbach's alpha of 0.88.

The Spanish Version of the Brief COPE (COPE-28) [24] consists of 28 items and assesses different coping scales, including substance use and religion. Participants respond using a four-point scale ranging from 0, “I have not done that at all” to 3, “I have done that very often”. Scores ranged from 0 to 6 for each subscale. An example item related to substance use is “I have taken alcohol or other drugs to feel better”, and an example related to religion is “I have tried to find comfort in my religion or spiritual beliefs”. The internal consistency for this study was high, with a Cronbach’s alpha of 0.83 (substance use $\alpha = 0.62$; religion $\alpha = 0.75$).

Duke-UNC Functional Social Support Questionnaire (FSSQ11) [25,26] consists of 11 items aiming to assess perceived social support. Participants respond using a scale ranging from 0, “never”, to 5, “always”. Scores range from 0 to 55. An example item is “I receive visits from friends and family”. The internal consistency for this study was high, with a Cronbach’s alpha of 0.91.

Personality Questionnaire Revised-Abbreviated (EPQR-A) [27] consists of 24 items aiming to assess personality traits, including neuroticism, one of the big five higher-order personality traits. Scores range from 0 to 6 for each scale. An example item is “I suffer significant changes in my mood”. Internal consistency for this study was good, with a Cronbach’s alpha ranging from 0.63 to 0.78.

The Barthel scale/index (BI) [28] consists of 10 items assessing functional dependency in daily life activities. Participants answer using a scale ranging from 0), dependent, to (5), need help, to (10), independent. Scores range from 0 to 100. An example item is “Help is needed to eat”. The internal consistency for this study was high, with a Cronbach’s alpha of 0.93.

Short portable mental status questionnaire (SPMSQ) [29,30] consists of 10 items aiming to assess cognitive impairment. Scores range from 0 to 10. An example item is “What day is today? (month, day, and year)”. The internal consistency for this study was high, with a Cronbach’s alpha of 0.89.

General Health Questionnaire (GHQ-12) [31] consists of 12 items, each assessing the severity of a mental problem over the past few weeks using a four-point Likert scale (from 0 to 3). An example item is “I have recently felt that I am ill”. The score was used to generate a total score ranging from 0 to 36. The internal consistency for this study was high, with a Cronbach’s alpha of 0.94.

2.3. Design and Procedure

A cross-sectional correlational study was conducted. Data were collected from eight regions in Ecuador in late 2019. For spatial distribution of the areas in Ecuador from which data were collected, see Figure 1. All data were collected via a survey that included standardized sociodemographic scales, which was administered using printed material by a team of psychologists trained by the lead researcher. Data from caregivers and the elderly were collected separately (first from the caregiver, then from the elderly). The duration of the sessions averaged 20–25 min.

2.4. Data Analysis

Statistical analyses were performed using the Statistical Package for Social Sciences application, version 21.0 for Mac (IBM, Madrid, Spain). The sociodemographic and clinical characteristics of the groups were expressed as means (M) and standard deviations (SD). Pearson’s correlation was conducted to examine the relationship between measured informal caregiver burden, and variables pertaining to the caregivers and their elderly patients. Inclusion in the prediction models of burden in informal male and female caregivers (enter method) was based on correlation analysis between burden and target variables. Student’s t-test (independent samples) was used to determine significant gender differences for quantitative variables. Levene’s and Shapiro–Wilk’s tests were used to assess the homogeneity of variance and normality, respectively. Effect size was measured using Cohen’s d. Lastly, independent multiple-regression models were conducted, including measured variables pertaining to caregivers and their elderly patients (predictive variables) and burden as the outcome variables. Significance adopted in analysis was $p < 0.05$.

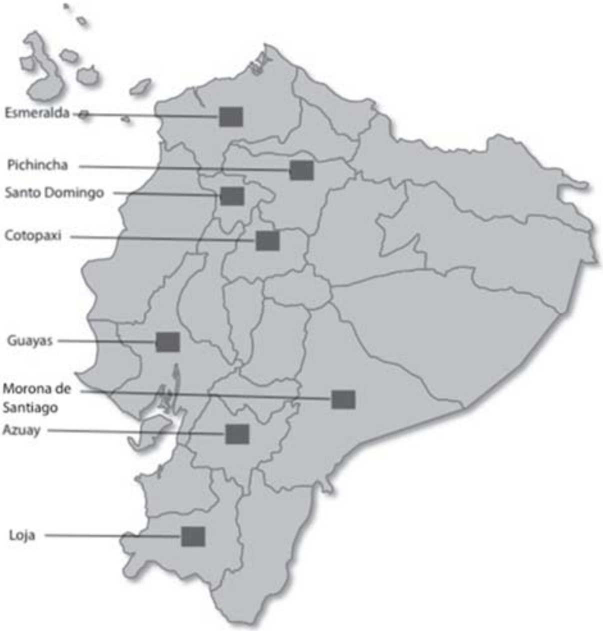

Figure 1. Distribution of the data-collection areas in Ecuador.

3. Results

3.1. Description of Caregiver and Elderly Samples

The final sample was made up of 688 family caregivers and 688 elderly patients from eight regions of Ecuador: 7.7% from Guayas (n = 53), 7.7% from Morona Santiago (n = 53), 2.9% from Esmeraldas (n = 20), 11.9% from Loja (n = 82), 8.7% from Azuay (n = 60), 4.5% from Cotopaxi (n = 31), 7.7% from Santo Domingo (n = 53), and 48.9% from Pichincha (n = 336), the closest area to the capital of the country, Quito. The caregivers' relationship to their elderly patients was as follows: 12.5% (n = 86) were husband/wife, 63.8% (n = 439) were son/daughter, 13.4% (n = 92) were grandson/granddaughter, 5.5% (n = 38) were brother/sister, and 4.8% (n = 33) were daughter/son-in-law. Caregiver age ranged from 17 to 80 years old, and the age of elderly patients (older than 65 years old) ranged from 65 to 104 years old.

Females were over-represented among family caregivers. Both male and female caregivers provided care for the elderly for an average of 6 years. Considering their age at the time of the study, this means that most caregivers of the elderly in Ecuador assume their role in their early 40 s, regardless of their educational level (Table 1).

3.2. Gender Differences in Caregiver and Elderly Variables Associated with Caregiver Burden

Female caregivers reported higher statistically significantly degrees of burden and neuroticism than those of male caregivers. Females mostly cope through religion, and males through substance abuse (Table 2).

Table 1. Sociodemographic information of informal caregivers and elderly.

Informal Caregiver Sociodemographics	**Males M ± SD (*n* = 139)**	**Females M ± SD (*n* = 549)**
Age (years)	47.6 ± 14.8	49.6 ± 14.7
Providing care (years)	6.2 ± 4.5	5.9 ± 4.2
	% (*n*)	% (*n*)
Marital status (S/M/D/W)	41 (57)/52 (71)/11 (6.5)/0	27.1 (149)/56.6 (343)/12.6 (37)/3.6 (20)
Ethnicity (M, W, O)	82 (114)/13.7 (19)/4.3 (6)	93.4 (512)/3.5 (19)/3 (15)
Education level (B, S, C)	8.6 (12)/38.8 (54)/46 (64)	22.4 (123)/41.0 (225)/31.5 (173)
Elderly Sociodemographics	**Males M ± SD (*n* = 272)**	**Females M ± SD (*n* = 416)**
Age (years)	80.6 ± 9.0	81.3 ± 9.3
	% (*n*)	% (*n*)
Marital status (S/M/D/W)	5.5 (15)/60.3 (164)/0	33.7 (140)/49.5 (206)/13.7 (57)/3.1 (13)
Ethnicity (M, W, O)	89.7 (244)/7.4 (20)/3 (8)	90.6 (377)/6 (25)/3.3 (14)
Education level (B, S, C)	59.2 (161)/19.9 (54)/8.8 (24)	15.9 (66)/42.1 (175)/35.8 (149)

Marital status: S = single, M = married, D = divorced, W = widowed; ethnicity: M = mixed racial or ethnic ancestry, W = white, O = other; education level: B = basic, S = secondary, C = college.

Table 2. Gender differences in informal caregiver and elderly variables.

Informal Caregiver Variables	**Males (*n* = 136) M ± SD**	**Females (*n* = 541) M ± SD**	***t***	**df**	***p***	**Cohen's d**
Caregiver's burden (Zarit)	39.4 ± 17.6	45 ± 17.7	−3.300	668	0.001 **	−0.317
Caregiver's competency (CUIDAR)	46 ± 9.7	46.5 ± 8.9	−0.455	666	0.649	−0.044
Caregiver's general health (GHQ12)	17 ± 8.32	17.9 ± 8.9	−1.067	677	0.286	−0.102
Caregiver's social support (DUKE11)	36.9 ± 10.4	36.4 ± 10.8	0.440	682	0.660	0.042
Caregiver's coping style—"substance abuse" (COPE)	0.7 ± 1.4	0.3 ± 0.8	4.843	682	<0.001 **	0.460
Caregiver's coping style—"religion" (COPE)	2.9 ± 2	3.8 ± 2	−4.609	683	<0.001 **	−0.439
Caregiver's neuroticism (EPQRA)	2.5 ± 1.8	3.1 ± 1.8	−3.234	681	0.001 **	−0.310
Caregiver's extraversion (EPQRA)	23.9 ± 1.6	3.7 ± 1.7	1.173	682	0.241	0.112
Elderly Variable	**Males (*n* = 266) M ± SD**	**Females (*n* = 411) M ± SD**	***t***	**df**	***p***	**Cohen's d**
Elderly's cognitive impairment (Pfeiffer)	3.6 ± 3.3	4.7 ± 3.5	−3.892	682	0.001 **	−0.304
Elderly's functional independency (Barthel)	57.8 ± 32.71	59.3 ± 31.9	−0.601	675	0.548	−0.047

Zarit = Zarit Burden Interview score; CUIDAR = Cuidar scale score; GHQ12 = General Health Questionnaire score; COPE = Spanish Version of the Brief COPE score; EPQRA = Personality Questionnaire Revised-Abbreviated score; Pfeiffer = Short portable mental status questionnaire score; Barthel = Barthel index score; df = degrees of freedom; $p < 0.001$ **.

3.3. Pearson's Correlation between Informal Caregivers' Burden and Measured Variables

For informal male caregivers of the elderly, the degree of burden in informal caregivers was positively correlated with time providing care ($r = 0.216$, $p = 0.012$) and neuroticism ($r = 0.316$, $p < 0.001$), and negatively correlated with caregiver's social support ($r = -0.365$, $p < 0.001$) and competence as caregiver ($r = -0.488$, $p > 0.001$). However, burden in informal male caregivers was not significantly associated with the perception of mental-health problems, as measured by the GHQ-12 ($r = 0.026$, $p = 0.768$), which, in fact, was positively and significantly associated with neuroticism ($r = 0.319$, $p < 0.001$). Interestingly, neither functional dependency ($r = -0.0688$, $p = 0.104$) nor cognitive impairment ($r = 0.104$, $p = 0.0234$) were significantly associated with burden, although they were significantly associated with each other ($r = 0.480$, $p < 0.001$).

For informal female caregivers of the elderly, the degree of burden in informal caregivers was positively correlated with time providing care ($r = 0.146$, $p < 0.001$), neuroticism ($r = 0.367$, $p < 0.001$), functional dependency ($r = –0.173$, $p > 0.001$), and cognitive impairment ($r = 0.181$, $p < 0.001$), and negatively correlated with caregiver’s social support ($r = −0.302$, $p < 0.001$) and competence as caregiver ($r = −0.348$, $p > 0.001$). However, burden in informal female caregivers was not significantly associated with the perception of mental-health problems, as measured by the GHQ-12 ($r = 0.027$, $p = 0.537$), which, in fact, was positively and significantly associated with neuroticism ($r = 0.387$, $p < 0.001$).

3.4. Prediction Models of Burden in Informal Caregivers of the Elderly

For informal male caregivers of the elderly, regression-model data indicated that the best predictors of burden were time providing care, the caregiver’s neuroticism, and the degree of the elderly’s functional dependency and cognitive impairment. The model accounted for 87.9% of the variance in burden ($F_{\Delta 8114} = 457.367$, $p > 0.001$). Competence as caregiver and social support failed to significantly predict burden in this model (Table 3).

Table 3. Prediction of burden in informal male caregivers.

Predictor	β	t	p	Lower 95% CI	Upper 95% CI	VIF
Time providing care (years)	0.337	3.872	<0.001 **	0.058	0.179	1.064
Caregiver’s competence (CUIDAR)	0.028	0335	0.739	−0.250	0.351	1.655
Caregiver’s social support (DUKE)	−0.050	−0.484	0.629	−0.415	0.252	1.457
Caregiver’s neuroticism (EPQRA)	0.349	3.798	<0.001 **	1.601	5.090	1.198
Elderly’s functional dependency (Barthel)	0.0416	4.592	<0.001 **	0.128	0.322	1.562
Elderly’s cognitive impairment (Pfeifer)	0.238	2.519	0.013 *	0.268	2.236	1.447

Zarit = Zarit Burden Interview score; CUIDAR= Cuidar scale score; GHQ12= General Health Questionnaire score; COPE = Spanish Version of the Brief COPE score; EPQRA = Personality Questionnaire Revised-Abbreviated score; Pfeiffer = Short portable mental status questionnaire score; Barthel = Barthel index score; β = Unstandardized Beta Coefficient; CI = Confidence Interval; VIF = Variance Inflation Factor; $p < 0.05$ *; $p < 0.001$ **.

For informal female caregivers, regression-model data indicated that the best predictors of burden were time providing care, caregiver’s competence, caregiver’s neuroticism, and elderly’s cognitive impairment. The model accounted for 88.1% of the variance in burden ($F_{\Delta 8496} = 92.816$, $p > 0.001$) (Table 4).

Table 4. Prediction of burden in informal female caregivers.

Predictor	β	t	p	Lower 95% CI	Upper 95% CI	VIF
Time providing care (years)	0.258	5.992	<0.001 **	0.063	0.125	1.035
Caregiver’s competence (CUIDAR)	0.088	2.197	0.029	0.018	0.322	1.530
Caregiver’s social support (DUKE)	0.036	0.715	0.475	−0.102	0.218	1.442
Caregiver’s neuroticism (EPQRA)	0.528	11.218	<0.001 **	4.148	5.910	1.345
Elderly’s functional dependency (Barthel)	0.062	1.353	0.177	−0.015	0.082	1.285
Elderly’s cognitive impairment (Pfeifer)	0.230	4.938	<0.001 **	0.685	1.590	1.262

Zarit = Zarit Burden Interview score; CUIDAR = Cuidar scale score; GHQ12 = General Health Questionnaire score; COPE = Spanish Version of the Brief COPE score; EPQRA = Personality Questionnaire Revised-Abbreviated score; Pfeiffer = Short portable mental status questionnaire score; Barthel = Barthel index score; β = Unstandardized Beta Coefficient; CI = Confidence Interval; VIF = Variance Inflation Factor; $p < 0.001$ **.

4. Discussion

The aim of this study was to examine the predictive value of variables pertaining to caregivers and their elderly patients with respect to caregiver burden. To our knowledge, this is the largest study attempting to predict burden in informal caregivers in Ecuador and contributes to the development of interventions aimed at improving the well-being of caregivers of the elderly.

The results of this study suggest that some gender differences may be relevant in the prediction of burden in informal caregivers of the elderly. For males, objective strains that are directly related with providing care seem to be more relevant, for example, time providing care (duration) and degree of elderly demands (both functional and cognitive) [32,33]. For females, subjective strains, in particular those related with their competence as caregivers, seem to play a more central role in the prediction of burden in this sample. This result is consistent with previous studies focusing on caregiver competence [34]. In both cases, informal male and female caregivers' time providing care and neuroticism remained significant predictors of burden. Indeed, as expected, the duration of the stressor providing care is the most important predictor of burden in informal caregivers. Furthermore, those who were more likely to be moody and to experience feelings such as anxiety, worry, fear, anger, frustration, guilt, depressed mood, and loneliness were more likely to report higher levels of burden. This result is consistent with the previous literature [35,36].

The results of this study indicate that informal female caregivers not only double the percentage of informal caregivers for the elderly, but also report significantly higher levels of burden. This result is consistent with previous research that strongly associated females with the role of care provider [37]. These results are also in line with the previous literature where females presented greater levels of burnout syndrome than those of males in their working environment [38]. Other studies also found higher burnout, perceived stress, and emotional exhaustion in female than in male professors in colleges and high schools [39,40]. Along the same lines, some authors also found cultural differences, since the gender gap regarding emotional exhaustion is greater in females than in males, with this trend being significantly higher in female employees from the United States when compared to that in Europeans [41].

Informal male and female caregivers also differ in their main coping mechanisms [42]. Males tend to use more active stress-management mechanisms, such as those based on substance use, while females tend to rely on more passive mechanisms based on praying and hoping for the best. This result complements previous studies focusing on a higher preference for substance use in males [43]. The absence of effective coping systems in the face of caregivers' highly demanding context on a professional and especially on an emotional level produces nonadaptive responses that can lead to drug abuse or be the basis for other psychological responses based on despair that can lead to psychopathologies such as anxiety and depression [44]. Therefore, it is important to emphasize the role of education in this professional group, not only to offer quality care, but also to preserve themselves from suffering pathologies and antisocial behaviors in the near future.

In sum, these results have potential implications for the development of social policies or recommendations to predict and reduce the burden in caregivers of the elderly or to design gender-specific interventions. This is important because providing care often results in poor health and quality of life [45,46], while at the same time, these consequences have been understudied or overlooked, so caregivers currently remain invisible patients [47]. In general, the results of this study support the importance of monitoring caregivers' period of time providing care (i.e., their exposure to stress), for example, by including resting periods to recover, and the deleterious effects of caregivers' neuroticism, underling the importance of moving towards a more mindful approach. Results also highlight the importance of enhancing elderly people's cognitive function through performing cognitively stimulating tasks (e.g., chess, crosswords), and being socially and physically active. Particularly in female caregivers of the elderly, enhancing their perception of competence as caregivers seems to be an additional element to consider in reducing their burden. However, the results of this study must be taken with caution since the study is based on cross-sectional correlational design on a convenience sample; therefore, there are limitations in terms of generalization to other populations and inference of causality, and more research in this area is needed.

5. Conclusions

Females were over-represented with respect to males among caregivers of the elderly by a ratio of 4:1, and they reported significantly higher levels of burden than those of male caregivers. Overall, time providing care, neuroticism, and cognitive impairment of elderly patients were the best predictors of caregiver burden, and these should be considered for tailored interventions aimed at reducing burden in caregivers of the elderly.

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Article

Hematological and Running Performance Modification of Trained Athletes after Reverse vs. Block Training Periodization

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Abstract: The aim of the present study was to analyze the effect of block (BP) and a reverse training periodization (RP) in the hematological and running performance of amateur trained athletes. Modifications in hematological, aerobic, and anaerobic running performance and countermovement jump before and after twelve weeks of BP vs. RP training programs were analyzed in 16 trained athletes (eight males: 40.0 ± 6.2 years; 179.2 ± 12.8 cm; 73.8 ± 12.2 kg; and eight females: 34.2 ± 4.1 years; 163.4 ± 9.6 cm; 57.0 ± 11.0 kg). A significant decrease in heart rate (HR) at ventilatory threshold (VT1) ($p = 0.031$; ES = 1.40) was observed in RP without changes in BP. In addition, RP increased significantly VO_2max ($p = 0.004$; ES = 0.47), speed at VO_2max ($p = 0.001$; ES = 1.07), HR at VT2 ($p < 0.001$; ES = 1.32) and VT1 ($p = 0.046$; ES = 0.57), while BP improved VO_2max ($p = 0.004$; ES = 0.51), speed at VO_2max ($p = 0.016$; ES = 0.92), and HR at VT2 ($p = 0.023$; ES = 0.78). In addition, only RP increased anaerobic performance in a running-based anaerobic sprint test (RAST) (mean sprint: $p = 0.009$; ES = 0.40, best sprint: $p = 0.019$; ES = 0.30 and total time: $p = 0.009$; ES = 0.40). Moreover, both types of training periodization proposed in this study maintained hematological values and efficiently improved jump performance ($p = 0.044$; ES = 0.6) in RP and $p = 0.001$; ES = 0.75 in BP). Therefore, twelve weeks of either RP or BP is an effective strategy to increase jump and aerobic running performance maintaining hematological values, but only RP increases anaerobic running performance.

Keywords: endurance; heart rate; runners; triglycerides; VO_2max

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1. Introduction

Sports performance is a complex combination of psychological and physiological modifications on the athlete's organism. Specifically, in endurance athletes, the physiological modifications related with success have been clearly defined by previous researchers, in which maximal oxygen uptake (VO_2max), lactate threshold, and efficiency appear to play key roles in endurance performance [1]. Along this line, the VO_2max and lactate threshold interact to determine the maximal oxygen consumption that could be sustained for a given period of time, being the most used predictors of elite performance. As well as the anaerobic threshold, the ventilatory threshold is one of the physiological parameters related specifically with endurance and ultra-endurance performance, being a trainable parameter and having a direct effect on competition performance [2,3].

To reach the physiological adaptations previously mentioned, hematological and chemical modification must occur. Different modifications in red blood cells, blood iron, and iron reserve cells as

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well as in the metabolic substrate are dependent of the performance level reached by the athlete [4,5]. The continuous training and the rational distribution of the training sessions would be the pillars to obtain the correct physiological modifications in athletes. There are numerous training periodization models to reach this aim [6–12]: (i) the traditional periodization models, focused on long distance and low intensity training; (ii) block models, focused on concentrating training load in short time period to increase the organic adaptation, and (iii) the more recent training paradigm based on high intensity and low volume training called reverse periodization (RP).

RP, unlike previous periodization models, begin the macrocycle with high-intensity and low-volume training, while gradually decreasing intensity and increasing volume or, depending on the sport, maintaining intensity and increasing volume during the following training periods [13]. This recent training paradigm has been previously studied in physical fitness, strength training, swimming, triathlon and rowing, obtaining increases in muscular endurance, maximum strength, and endurance performance [10,13,14]. The increases in performance associated with the RP are closely related with the use of high intensity training, especially with methodology like high intensity interval training [7]. These short-term training methodologies have been reported as efficient interventions to increase sympathetic modulation to achieve different physiological adaptions related with aerobic performance such as the increase in muscle buffering capacity, glycogen content, GLUT4 concentration, and maximal glucose transport activity in skeletal muscle [15–18]. In addition, RP improves jump performance in endurance athletes, but traditional periodization affects it negatively [13].

Finally, hematological parameters might be influenced by long-term training and competition periods decreasing during the intense periods of training throughout the season [18]. However, the effect of periodized training, and specifically, the influence of this new training periodization model (i.e., reverse periodization) on chemical parameters and physiological adaptations are still poorly known, especially in the running collective. For this reason, we proposed the present research with the aim to analyze the effect of 12 weeks of a Block Periodization and a Reverse Periodization on hematological parameters, countermovement jump, and aerobic (measured in a treadmill test and a 10 km time trial test) and anaerobic running performance of trained athletes. The initial hypothesis was that reverse training periodization would achieve a higher running performance and a significant modification in hematological parameters than block training periodization.

2. Materials and Methods

2.1. Design

To test the effects of 12 weeks of the two types of periodization training programs (reverse vs. block periodization) on hematological variables, aerobic and anaerobic running performance, and countermovement jump, a single-blinded randomized controlled (participant did not know the periodization model they were performing) trial with a pre- and post-test was conducted. Athletes were randomly divided into two experimental groups: (a) the reverse periodization (rp) group, who performed 4-weeks of high intensity training, 4-weeks of high volume training, and 4-weeks of tapering ($n = 8$); and (b) the block periodization (bp) group, who performed 4-weeks of high volume training (accumulation), 4-weeks of high intensity training (transformation), and 4-weeks of tapering (realization) ($n = 8$).

2.2. Participants

Sixteen amateur athletes (eight males: 40.0 ± 6.2 years; 179.2 ± 12.8 cm; 73.8 ± 12.2 kg; and eight females: 34.2 ± 4.1 years; 163.4 ± 9.6 cm; 57.0 ± 11.0 kg; six training sessions/week; 42.4 ± 12.4 min/session; 4.3 ± 0.4 h of training/week; >6 year of experience on running training; competing at regional and national level in 10 km and half-marathon races) participated in this study. None of the participants had any musculoskeletal disorders. Before the testing sessions, participants were divided, in randomized order, into either the RP ($n = 8$; four males and four females; age: 37.0 ± 9.2 years; height: 170.2 ± 19.2 cm;

weight: 65.8 ± 10.2 kg) or BP (n = 8; four males and four females; age: 37.2 ± 5.7 years; height: 172.4 ± 9.1 cm; weight: 65.1 ± 10.4 kg) groups. The study design and the procedures employed were in accordance with ethical standards and the Declaration of Helsinki (1964). Each participant was fully informed of the risks associated with the study and provided written informed consent before starting the study. The present research was approved by the Catholic University of Murcia Ethics Committee (REF: CE071902).

2.3. Testing Protocol

The assessments were carried out on two different days, separated by 48 h, in both the pre- and post-test and at the same time of the day in both evaluations. Pre- and post-tests were carried out 72 h after the last intense workout to allow complete recovery from training. On the first day, athletes visited the laboratory to conduct a blood test, a running-based anaerobic sprint test (RAST), and a treadmill running test. The second testing session was performed 72 h after the first testing session on an official athletics track and included Countermovement Jump (CMJ) and the 10 km time trial test. On the day after the training program finished, the same testing procedures were applied. In addition, a nutritionist performed an initial prospective 24-h dietary recall to assess the participants' diets. Afterward, a 7-day food record with qualitative and quantitative data, along with a printed guide for proper filling, was given to the participants to calculate their daily average intake through the first week of the training program, and calculated using software (Cronometer Software Inc., https://cronometer.com, V.1.) [19]. This nutritional assessment was performed again when the training program finished and no differences were observed between the periodization models and moments.

2.4. Blood Sample Collection

The blood sample (2.5 mL) was withdrawn from an antecubital vein using a sterile technique to analyze hematological variables. Blood samples were taken before breakfast after an overnight fast. Blood extraction was performed with the subject seated. Erythrocytes ($\times 10^6$/L), hematocrit (%), hemoglobin (g/dL), ferritin (g/L), glucose (mg/dL), and triglycerides (mg/dL) were assessed.

2.5. Running-Based Anaerobic Sprint Test (RAST)

The RAST consisted of six maximal efforts of 35-m, separated by a passive recovery of 10 s. The athlete started 0.5 m behind the start line, which was marked by a photocell (Witty, Microgate, Italy) [20]. Before starting, the athletes were instructed to run as fast as possible to the end of the 35 m course. Before testing, a warm-up consisting of 5 min of jogging followed by active stretching and two short duration submaximal sprints was performed. Following each sprint, athletes decelerated and walked to the starting line ready for the subsequent sprint. The best and mean sprint time were recorded as the performance indices. Verbal encouragement was given to the participants to ensure maximum physical effort.

2.6. Treadmill Running Test

Thirty minutes after RAST test, runners completed an incremental test to exhaustion on a treadmill (Run MedTechnogym, Cessena, Italy) in standard environmental conditions with the grade set at 1%. The tests were performed between 10:30 and 12:00 a.m. in the laboratory with the room temperature set between 20°–22 °C and 45–50% of humidity. The gas analyzer system was calibrated before each test following the manufacturer's recommendations. During testing, gas exchange was measured using a breath-by-breath gas analyzer (Metalyzer 3B; Cortex-medical, Leipzig, Germany). Expired minute volume (VE), oxygen consumption (VO_2), and carbon dioxide production (VCO_2) were continuously recorded and averaged each minute. The respiratory exchange ratio ($R = VCO_2/VO_2$), the O_2 ventilatory equivalent (VE/VO_2) and the CO_2 ventilatory equivalent (VE/VCO_2) were calculated. Athletes started running at 8 km/h for 5 min. Subsequently, the work rate was increased by 0.5 km/h every 30 s in a progressive manner until exhaustion for optimal determination of VT2 and VO_2max.

The corresponding heart rate was also determined by a validated Polar RS800CX heart rate monitor (Polar Electro, Kempele, Finland) [21]. Verbal encouragement was given to ensure maximum physical effort. The test was concluded according to traditional physiological criteria when participants reached volitional fatigue [3]. After the test, Ventilatory Thresholds were determined as follows: T1 was defined as the first increase of VE/VO_2 vs. workload, without a simultaneous increase in VE/VCO_2 vs. workload; and VT2 was defined as the second increase in VE with a concomitant rapid increase in VE/VO_2 and VE/VCO_2 and decrease of end-tidal CO_2 tension ($PETCO_2$) [22].

2.7. Countermovement Jump

During the second testing day, athletes completed a 15 min warm-up, similar to those performed prior to a competitive event, which included the following components: jogging (easy pace-Z1), running technique, and progressive running to race-pace. After warm-up, participants executed two submaximal trials of CMJ to ensure proper execution of the jumps with 1 min of rest in between trials. Two minutes after the specific warm-up to jump, participants started the CMJ test. For the CMJ execution, participants maintained 90° of knee flexion during 5″ while the researcher confirmed the 90° with a square. The CMJ heights were calculated using a contact platform (Ergotester, Globus, Codogne, Italy) [23]. The CMJ was performed at the center of the platform with the feet placed shoulder width apart in the standing position. Participants were asked to jump as high as possible with a rapid self-selected countermovement. Participants were asked to try and land close to the take-off point. Each participant performed two attempts, with 90 s of rest in between attempts. The best trial from each participant was used for data analysis.

2.8. Ten Kilometer Time Trial Test

After the CMJ test, a 10-km time trial test was carried out on an official athletic track. The 10-km time was recorded using a Geonaute chronometer Onstart 710 (Decathlon, Villeneuve-d'Ascq, France) by two of the researchers and the mean of these values was used for analysis.

2.9. Training Program

Two weeks before starting the training program, all participants performed the same two-familiarization weeks. During this stage, athletes completed three running sessions in Z1 and two strength workouts each week. Participants started the familiarization period after three weeks of detraining weeks or off-season period. After this initial training period, participants in both periodizations completed a 12-week training periodization program consisting of two strength workouts, five running session, and one day of total rest per week. The RP group performed a 12-week periodization composed of a 4-week mesocycle based on high intensity and low volume training, a 4-week mesocycle based on high volume and intensity training, and a 4-week mesocycle based on modeling competition and tapering. The training intensity distribution was polarized in the first mesocycle and pyramidal in the second and third. On the other hand, the BP group completed a 12-week periodization composed of 4-weeks of high volume training (accumulation), 4-weeks of high intensity training (transmutation), and 4-weeks of modeling competition and tapering (realization). In addition, we applied a polarized distribution in the second mesocycle and a pyramidal distribution in the first and third. Training zones were classified, according to previous literature, in three training zones: zone 1 (Z1), low intensity training (Rated of Perceived Exertion, RPE ≤ 4); zone 2 (Z2), anaerobic threshold training (RPE 4–7); and zone 3 (Z3), high intensity training (RPE ≥ 7) [24]. To quantify the training load of each session conducted by the athletes during each week, we used the session-RPE method [25]. In this method, the training load is quantified by multiplying the whole training-session RPE using the 10-point Borg scale by its duration. This product represents the training impulse (TRIMP) or the magnitude of the internal training load in arbitrary units. The RPE was recorded thirty minutes after every training session. An example of typical training series in each phase of the 12-week training period for both periodizations are shown in Table 1.

Table 1. Examples of typical training series in each phase of the 12-week training period for both block and reverse training periodizations.

Periodization Model	Weeks 1–4	Weeks 5–8	Weeks 9–12
Block periodization	1 × 50 min Z1 3 × 2000 m/3 min Z2	6 × 1000 m/3 min Z3 2 × (5 × 400 m/90 s)/8 min/Z3	10 × 1000 m/3 min Z3
Reverse periodization	10 × 200 m/2 min Z3 3 × (10 × 100 m/30 s)/3′ Z3	8 × 1000 m/2 min Z2 2 × (10 × 300 m/90 s)/8 min Z3	10 × 1000 m/3 min Z3

Series × (repetition × distance or duration and intensity/recovery between repetitions)/recovery between series; Z1—Low intensity training; Z2—Threshold training; Z3—High intensity training.

2.10. Statistical Analysis

Statistical analysis of data was performed with SPSS v 24.0 (Chicago, IL, USA) in the Windows environment. Descriptive data are presented as mean ± standard deviation (SD). For the inferential analysis, a Shapiro–Wilks W-Test was performed to establish the normality of the sampling distribution and Mauchly's W test to analyze the sphericity between measurements. In addition, a two-way (type of periodization × time) analysis of variance (ANOVA) with repeated measures and Bonferroni post-hoc was used to investigate the differences in the study variables. In addition, an unpaired sample *T*-test was used to compare the training load of both periodization programs. Mean difference and 95% confidence interval (95% CI) were included. The effect size (ES) was calculated using partial ETA squared (η2) in ANOVA. In addition, the d was calculated using Cohen's guidelines to compare the training load in each intervention (BP vs. RP) and to analyze the effect of time using the threshold values of >0.2 (small), >0.6 (moderate), >1.2 (large), and >2.0 (very large) [26]. For all procedures, a level of $p \leq 0.05$ was selected to indicate statistical significance.

3. Results

Table 2 shows the hematological variables. There was no interaction effect of periodization group × time in the hematological variables. There was a main effect of time on triglycerides (F = 5.333; p = 0.037; d = 0.94) in RP.

Table 2. Hematological results in both groups.

Variables	Pre-Training		Post-Training				95% CI for Difference		
	Mean	SD	Mean	SD	ES	p	MD	Lower Bound	Upper Bound
Reverse Periodization									
Erythrocytes ($\times 10^6$/L)	4.5	0.1	4.4	0.1	0.98	0.299	−0.1	−0.3	0.1
Hematocrit (%)	42.9	1.1	42.4	0.8	0.37	0.448	−0.5	−1.8	0.8
Hemoglobin (g/dL)	14.2	0.4	13.8	0.2	0.81	0.129	−0.4	−0.9	0.1
Ferritin (g/L)	129.8	31.8	124.1	19.4	0.16	0.627	−5.7	−30.1	18.5
Glucose (mg/dL)	88.7	3.2	84.7	1.8	1.1	0.077	−4	−8.4	0.4
Triglycerides (mg/dL)	70	5.4	75.7	4.5	−0.94	0.037	5.7	1.1	10.3
Block Periodization									
Erythrocytes ($\times 10^6$/L)	4.5	0	4.5	0	0.89	0.164	−0.1	−0.2	0.1
Hematocrit (%)	42.1	0.9	41	0.6	0.98	0.107	−1.1	−2.4	0.2
Hemoglobin (g/dL)	14.1	0.3	13.6	0.2	1.55	0.1	−0.5	−1	0
Ferritin (g/L)	118.5	19.7	108.3	14.7	0.46	0.396	−10.2	−34.5	14.2
Glucose (mg/dL)	90.7	3.2	87.7	1.8	0.83	0.177	−3.1	−7.4	1.4
Triglycerides (mg/dL)	82.7	5.6	83.6	4.3	−0.14	0.698	0.9	−3.7	5.5

MD: Mean difference. CI: Confident interval; ES: effect size.

Concerning the running treadmill test variables investigated (Table 3), a significant interaction effect of periodization × time was observed in the heart rate at VT1 (F = 7.11; p = 0.018; η^2 = 0.34), showing a significant decrease in RP (F = 4.74; p = 0.031; d = 1.40) but without changes in BP. In addition, there was a main effect of time on HR at VT2 (F = 26.57; p < 0.001; d = 1.32), VT1 (F = 5.74; p = 0.046; d = 0.57), VO_2max (F = 11.93; p = 0.004; d = 0.47), and speed of VO_2max (F = 15.73; p = 0.001; d = 1.07) in RP and on HR at VT2 (F = 5.49; p = 0.023; d = 0.78), VO_2max (F = 11.93; p = 0.004; d = 0.51), and speed of VO_2max (F = 7.56; p= 0.016; d = 0.92) in BP.

Table 3. Treadmill running test results in both groups.

Variables	Pre-Training	SD	Post-Training		ES	p	95% CI for Difference		
	Mean		Mean	SD			MD	Lower Bound	Upper Bound
Reverse Periodization									
HR VT1 (bpm)	140.3	2.0	137.1	2.5	1.40	0.031	−3.3	−6.1	−0.3
Speed VT1 (km/h)	10.8	1.0	11.2	0.9	−0.35	0.278	0.3	−0.3	1
VT1 (% of VO_2max)	59.9	1.6	60.9	1.6	−0.57	0.046	1	0	1.9
HR VT2 (bpm)	177.0	2.8	172.9	2.4	1.32	<0.001	−4.1	−5.7	−2.7
Speed VT2 (km/h)	15.4	0.8	15.7	0.9	−0.27	0.322	0.3	−0.2	0.7
VT2 (% of VO_2max)	71.3	1.4	71.6	1.5	−0.24	0.548	0.4	−0.9	1.6
HR VO_2max (bpm)	187.6	1.7	187.8	1.8	−0.07	0.661	0.1	−0.4	0.7
Speed VO_2max (km/h)	17.5	0.7	18.3	0.6	−1.07	<0.001	0.8	−2.3	−1.2
VO_2max (ml/kg/min)	59.4	2.6	60.8	2.6	−0.47	0.004	1.4	0.5	2.1
Block Periodization									
HR VT1 (bpm)	138.8	2.5	141.2	1.8	−0.83	0.106	2.4	−0.5	5.3
Speed VT1 (km/h)	11.3	0.5	11.2	0.9	0.13	0.364	−0.1	−1	0.3
VT1 (% of VO_2max)	62.6	1.4	62.9	1.2	−0.16	0.602	0.3	−0.7	1.2
HR VT2 (bpm)	170.8	2.1	168.9	1.7	0.78	0.023	−1.9	−3.4	−0.2
Speed VT2 (km/h)	14.4	0.5	14.9	0.6	−0.91	0.056	0.5	0	1
VT2 (% of VO_2max)	72.5	2.1	72.0	1.6	0.2	0.448	−0.4	−1.7	0.8
HR VO_2max (bpm)	183.8	1.6	183.9	1.7	−0.07	0.661	0.1	−0.4	0.7
Speed VO_2max (km/h)	16.8	0.5	17.4	0.6	−0.92	0.016	0.6	−2.5	−1.3
VO_2max (ml/kg/min)	53.6	2.4	55.0	2.1	−0.51	0.004	1.4	0.5	2.1

HR: heart rate; VT: ventilatory threshold; VO_2max: maximum oxygen uptake; MD: mean difference; CI: confident interval; ES: effect size.

Regarding the RAST variables (Table 4), there was a main effect of group × time on mean time in RAST (s) (F = 9.32; p = 0.009; η^2 = 0.40) and on the total time of the RAST test (s) (F = 9.32; p = 0.009; η^2 = 0.40) with a significant decrease in RP. Furthermore, there was a main effect of time on the best sprint in RAST (s) (F = 7.09; p = 0.019; d = 0.30) in RP. Concerning CMJ and the 10,000 m test variables analyzed (Table 3), no main effect of periodization group × time was observed in these variables. However, there was a main effect of time on CMJ height (F = 4.54; p = 0.044; d = 0.6) in RP and in CMJ height (F = 14.72; p= 0.001; d = 0.75) and the 10 km time-trial (F = 9.73; p = 0.008; d = 0.15) in BP.

Table 4. Countermovement jump test, RAST test, and 10,000 m time trial test results in both groups.

Variables	Pre-Training		Post-Training				95% CI for Difference		
	Mean	SD	Mean	SD	ES	p	MD	Lower Bound	Upper Bound
Reverse Periodization									
Mean sprint RAST (s)	5.8	0.3	5.6	0.3	0.59	0.01	−0.2	−0.1	−0.3
Best sprint RAST (s)	5.6	0.3	5.5	0.3	0.3	0.019	−0.1	−0.2	0
Total time sprint RAST (s)	34.8	1.9	33.6	1.8	0.56	0.01	−1.2	−0.6	−1.8
CMJ height (cm)	33	1.5	34	1.2	−0.6	0.044	1	0	1.9
10,000 m (s)	2481.5	369.4	2429.2	363.6	0.13	0.089	−52	−113	9
Block Periodization									
Mean sprint RAST (s)	5.7	0.4	5.7	0.4	0	0.965	0	0.1	−0.1
Best sprint RAST (s)	5.6	0.4	5.6	0.4	0	1	0	0.1	−0.1
Total time sprint RAST (s)	34.2	2.7	34.2	2.4	0	0.965	0	0.6	−0.6
CMJ height (cm)	31.3	2.1	33.1	1.8	−0.75	0.001	1.8	0.8	2.7
10,000 m (s)	2728.8	522.3	2640	418.3	0.15	0.008	−88.8	−149.8	−27.7

RAST: running-based anaerobic sprint test; CMJ: countermovement jump; MD: mean difference; CI: confident interval; ES: effect size.

Concerning training monitoring, significant differences were observed in the total training load (TRIMPS), total training time (min), and in the time spent in each training zone between groups (Table 5 and Figure 1).

Table 5. Training load of each periodization group.

	Total Time (min)	**Time in Z1 (min)**	**Time in Z1 (%)**	**Time in Z2 (min)**	**Time in Z2 (%)**	**Time in Z3 (min)**	**Time in Z3 (%)**	**Training Load (TRIMPS)**
Reverse Periodization	3246.1 ± 38.3	1963.0 ± 30.0	60.5 ± 0.5	715.6 ± 6.1	22.5 ± 0.3	567.6 ± 20.4	17.5 ± 0.5	19,932.1 ± 250.5
Block Periodization	3319.9 ± 37.4	2009.4 ± 25.3	60.5 ± 0.2	774.4 ± 11.5	23.3 ± 0.2	536.1 ± 4.7	16.2 ± 0.1	20,292.3 ± 222.0
p	0.002	0.005	0.780	<0.001	<0.001	0.001	<0.001	0.009
ES (*d*)	1.95	1.58	0	6.04	2.97	2.01	3.41	1.52

Zone 1 (Z1), low intensity training; zone 2 (Z2), anaerobic threshold training; and zone 3 (Z3) high intensity training.

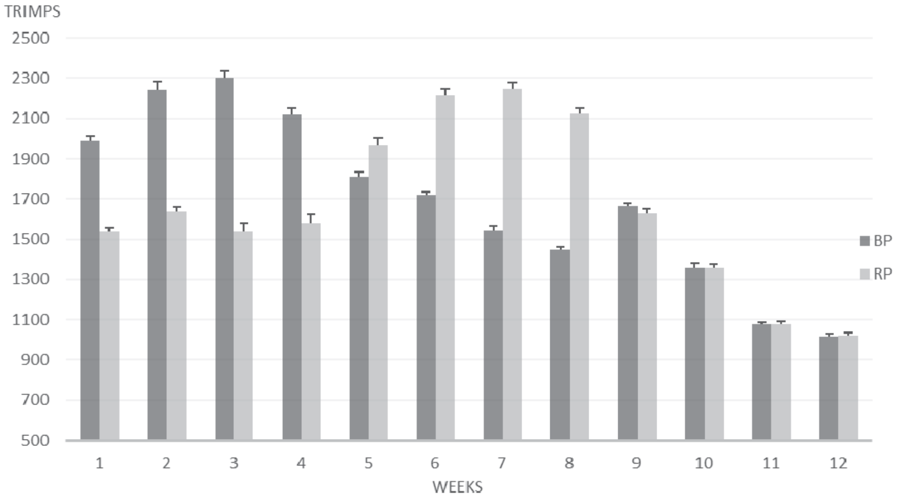

Figure 1. Training load (TRIMPS) in arbitrary units during the 12 weeks of the two periodization models (RP: reverse periodization; BP: block periodization).

4. Discussion

The aim of this study was to analyze the effect of BP vs. RP in the hematological and running performance of amateur trained athletes. The initial hypothesis was partially confirmed since RP achieved an increased running performance as well as BP, with only triglycerides modified by RP. On the other hand, RP achieved higher anaerobic running performance than BP.

Analyzing the training load of each group, we found a significant higher volume in BP than RP. Traditional periodization studies tried to equal the volume of training in the different training groups analyzed, producing a distortion in the reverse training periodization structure in this matching process [13]. Reverse periodization is based on a low training volume combined with high intensity, starting the implementation of high intensity from the beginning of the macrocycle [23]. Previous authors have shown how the equalization of training volumes and intensities when comparing reverse periodization with other models was a limitation to really testing the efficiency of this new training paradigm [6,27,28]. In the present study, we took that fact into account, and the load was distributed according to this premise, obtaining a significant difference in training volumes.

The reverse periodization showed an increase in cardiovascular efficiency in both VT1 and VT2. This modification could be due to the effect of high-intensity training conducted in this periodization model, which led to a hyperactivation of the sympathetic nervous system during the efforts, being a powerful tool to lead this cardiovascular improvement [7,18]. The increases in VO_2max and the speed at VO_2max obtained by reverse and BP highlighted the effectiveness of both systems to reach aerobic physiological markers, but reverse periodization was more efficient since a lower training volume was performed. The increases in VO_2max and speed at VO_2max were in consonance with previous authors in equaled volume training periodization in cycling, triathlon, and swimming [12,13,23], showing how these methods also improve aerobic performance in running. In the same way, the decreased in HR at VT2 was in line with other research conducted with swimmers [6]; nevertheless, in this study, improvement in swimming speed was found, in contrast to the present research, where most likely the different performance level and sport modality could explain this difference.

Both training groups (RP and BP) significantly increased the VO_2max, but only in BP was this increase accompanied by a significant improvement in the running field test (10,000 m).This result was in contrast with previous research where after 10 weeks of reverse periodization, running performance (400 m and 1000 m) increased in amateur athletes [29]. We found how reverse periodization allowed for an increase in physiological laboratory test markers, but in this case failed to increase field performance. This result should take into account for a better design of this specific periodization, trying to improve the transference of training adaptations to final field performance. In this line, the significative improvements of block periodization could be related with the higher volume of training in the Z2 and Z3 intensity zones, specific to improving performance in the field test analyzed [12].

Regarding jump performance results, a previous study reported an interference effect of aerobic endurance exercise on strength and power gains [12]. In addition, a recent study showed that traditional periodization negatively affected jump performance in endurance athletes, but reverse periodization improved it [13]. In this way, our results showed that variations in training periodization could interfere with lower body power in both groups, suggesting that both RP and BP positively improved CMJ height. This fact could be explained by the lower load performed by the two groups during the last weeks of the program (tapering), which is in accordance with a previous study that found an increase in jump performance during the last two weeks of a training program where the load decreased in endurance athletes [30]. Therefore, both RP and BP are effective periodization programs to improve CMJ jump in amateur athletes.

Concerning hematological parameters, hemoglobin and hematocrit might be influenced by long-term training and competition periods decreasing during the intense periods of training throughout the season [31]. According to our results, the type of periodization does not seem to affect the hematological variables analyzed in this study, showing that the two types of periodization reported a non-significant trend to decrease. This fact could be explained by the production of a hemolysis or/and hemodilution, which can contribute to generating sport anemia [32], which in a clinical case can impair athletic performance [33]. Thus, both training periodization of RP focusing more in Z3 and BP with more total training load and more time training in Z1 and Z2 can produce a decreasing trend in hematological parameters. In addition, our results showed a significant increase in blood triglycerides in RP but not in BP. Despite the values of triglycerides in both cases being in a physiological range, this fact could be explained by the training distribution performed in RP. The high-intensity training (Z3) increased post-exercise lipid oxidation, decreased triglycerides levels [34], and a higher amount of training of this type of exercise is performed at the beginning of the RP. In addition, triglycerides and other lipid metabolism parameters are strongly dependent on the training level of athletes, impairing during short-term detraining periods [35]. Therefore, more studies are needed to explain the long-terms effect of RP in fat metabolism and the response of other variables like body composition to this type of periodization.

A previous study showed that RP periodization can optimize anaerobic running performance (i.e., 400 m running) in comparison to BP [29]. We found similar results in the present research, showing that RP, but not BP, significantly improved RAST performance. This fact could be explained by the higher time training performed in Z3 of RP than the BP. The training performed in this zone (i.e., high-intensity interval training) improved anaerobic pathways, muscle buffering capacity, and lactate tolerance [15,35,36], obtaining higher anaerobic performance. Therefore, RP could be a good strategy to improve anaerobic performance in amateur endurance athletes in order to obtain a successful performance in running races where runners sometimes need to perform a sprint at the end of the race, but future research with a larger sample should confirm this point.

The main limitation of the present study was the small sample size analyzed, which limited the generalization of the results. Along this line, we could not perform a randomized controlled crossover because of the impossibility of maintaining participants during a long time of period out of their club training and competitions. In addition, regarding training volumes, we wanted to analyze the real training proposed by coaches following both periodization models, and not only the concept of reverse periodization. Then, a reduced volume was proposed for the reverse training periodization once it was one of the bases of this periodization model. As a consequence, our conclusions should not be extracted regarding only the intense training distribution during the macrocycle, but to the cumulative effect of this with the usual reduction in overall training volume, which is the paradigm of the reverse training periodization: low volume and high intensity since the start of the macrocycle.

In addition, practical recommendations should be restricted to amateur runners. Nevertheless, due to the findings in aerobic performance, it can be reasonably suggested from our data that the findings of this study can apply to other athletes such as endurance athletes who may want to optimize their training program. However, more research with endurance athletes is necessary to obtain more information about the most effective periodization in populations and athletes with different fitness levels. Furthermore, from an applied perspective, the athletes' coaches and research have information to help them in training planification. They can use the results obtained to select the most effective periodization according to the physical demands of the sport modality, given that RP improved both aerobic and anaerobic performance. This periodization, focused on intensity, led us to think that intensified training is a key factor in optimizing endurance athlete performance.

The basis of reverse training periodization is a decrease in low intensity-high volume training and an increase in high intensity-low volume training. As a future line of research, we propose an analysis to address the influence of each individual factor in the final performance of athletes to better understand the principles of this new training paradigm.

5. Conclusions

Reverse and block periodization are an effective strategy to improve physiological variables and aerobic running performance during a treadmill test, but only reverse periodization increased anaerobic running performance in a RAST test. Moreover, both types of training periodization proposed in this study maintained the hematological values of the amateur athletes. In addition, ten weeks of a reverse periodization program increased blood triglyceride values. Moreover, both types of periodization efficiently improved jump performance.

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Article

Post-Exercise Recovery of Ultra-Short-Term Heart Rate Variability after Yo-Yo Intermittent Recovery Test and Repeated Sprint Ability Test

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Abstract: This study aimed to examine the agreement and acceptance of ultra-short-term heart rate (HR) variability (HRV_{UST}) measures during post-exercise recovery in college football players. Twenty-five male college football players (age: 19.80 ± 1.08 years) from the first division of national university championship voluntarily participated in the study. The participants completed both a repeated sprint ability test (RSA) and a Yo-Yo intermittent recovery test level 1 (YYIR1) in a randomized order and separated by 7 days. Electrocardiographic signals (ECG) were recorded in a supine position 10 min before and 30 min after the exercise protocols. The HR and HRV data were analyzed in the time segments of baseline 5~10 min (Baseline), post-exercise 0~5 min (Post 1), post-exercise 5~10 min (Post 2), and post-exercise 25~30 min (Post 3). The natural logarithm of the standard deviation of normal-to-normal intervals (LnSDNN), root mean square of successive normal-to-normal interval differences (LnRMSSD), and LnSDNN:LnRMSSD ratio was compared in the 1st min HRV_{UST} and 5-min criterion ($HRV_{criterion}$) of each time segment. The correlation of time-domain HRV variables to 5-min natural logarithm of low frequency power (LnLF) and high frequency power (LnHF), and LF:HF ratio were calculated. The results showed that the HRV_{UST} of LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio showed trivial to small effect sizes (ES) (−0.00~0.49), very large and nearly perfect interclass correlation coefficients (ICC) (0.74~0.95), and relatively small values of bias (RSA: 0.01~−0.12; YYIR1: −0.01~−0.16) to the $HRV_{criterion}$ in both exercise protocols. In addition, the HRV_{UST} of LnLF, LnHF, and LnLF:LnHF showed trivial to small ES (−0.04~−0.54), small to large ICC (−0.02~0.68), and relatively small values of bias (RSA: −0.02~0.65; YYIR1: 0.03~−0.23) to the $HRV_{criterion}$ in both exercise protocols. Lastly, the 1-min LnSDNN:LnRMSSD ratio was significantly correlated to the 5-min LnLF:LnHF ratio with moderate~high level ($r = 0.43$~0.72; $p < 0.05$) during 30-min post-exercise recovery. The post-exercise 1-min HRV assessment in LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio was acceptable and accurate in the RSA and YYIR1 tests, compared to the 5-min time segment of measurement. The moderate to high correlation coefficient of the HRV_{UST} LnSDNN:LnRMSSD ratio to the $HRV_{criterion}$ LnLF:LnHF ratio indicated the capacity to facilitate the post-exercise shortening duration of HRV measurement after maximal anaerobic or aerobic shuttle

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running. Using ultra-short-term record of LnSDNN:LnRMSSD ratio as a surrogate for standard measure of LnLF:LnHF ratio after short-term bouts of maximal intensity field-based shuttle running is warranted.

Keywords: maximal intermittent exercise; post-exercise recovery; heart rate variability; autonomic nervous system

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1. Introduction

Heart rate (HR) variability (HRV) is a physiological process that reflects the biological fluctuation in cardiac activation that is regulated by the autonomic nervous system (ANS). Heart rate variability assessment requires biosignal recording via non-invasive techniques to detect beat-to-beat intervals of the HR responses in a time series. Patterns of HRV have previously been used to measure the cardiovascular function in response to exercise adaptation [1,2], psychological performance [3], environmental behavior [4], and recovery status in sports training [5,6].

To validate HRV recordings, the Task Force of the European Society of Cardiology and the North American Society of Pacing Electrophysiology recommend a series of 512 R-wave to R-wave intervals (RRI) for HRV data analysis [7]. Alternatively, short-term 5-min HRV recordings after a 5-min stabilization (maintain a fixed posture in a stable manner) can be used as a standard process during HRV measurements [7]. The standardization of testing procedure requires around 10 min to obtain sufficient number of RRI for time-domain, frequency-domain, and non-liner analyses. However, this process is very time-consuming and is mainly limited to clinical application; therefore, this type of analysis is seldom used in applied sports settings. Thus, there is a need for more time-efficient methods for analyzing HRV in applied sports contexts.

Post-exercise recovery of cardiac-related responses play a critical role in the homeostatic functioning of the ANS and cardiovascular system. The capacity to recover from exercise-induced ANS changes is highly related to the exercise intensity [8,9]. Repeated sprint ability tests (RSA), which are anaerobic-based assessments of exercise capacity, may induce post-exercise effects on vagal withdrawal. For example, Abad et al. [10] found that the depression of parasympathetic reactivation may be longer than 2-h during passive recovery after the RSA test. In contrast, aerobic assessments, such as the Yo-Yo intermittent recovery level 1 test (YYIR1), have shown no significant influence on post-exercise modulation of cardiac autonomic function [1]. To understand the potential difference in post-exercise recovery of HRV between anaerobic and aerobic exercise, Nakamura et al.'s study [11] compared the HRV recovery between the 30–15 intermittent fitness test (a similar test of aerobic function to the YYIR1) and the RSA test in 13 national male handball players during the preparatory period of National Championship. The relative change in HRV recovery after 5-min stabilization between the 30–15 intermittent fitness test and RSA test was similarly altered, with fast HRV recovery of the root mean square of successive normal-to-normal interval differences (RMSSD) after 3 min of the RSA test. Due to these contradictory findings, further research is therefore needed to elucidate the differences in HRV between aerobic and anaerobic assessment and training.

To improve the utility of HRV measures in the assessment of exercise recovery in applied sports settings, ultra-short-term HRV (HRV_{UST}) assessment has become attractive to practitioner and researches due to the time efficiency of data collection. HRV_{UST} only requires 10–60 s recording times, depending upon the methodologies and device technology. It is recognized that 1-min HRV_{UST} displays excellent validity, reliability, and limits of agreements as a surrogate to the 5-min criterion of HRV record ($HRV_{criterion}$) [12–17]. The advantage of HRV_{UST} record has been demonstrated in sports training [16,17], exercise testing [12,13,18], cardiovascular medicine [19–21], and metabolic disease [22].

In frequency-domain HRV analysis, the ratio of low-frequency power (LF), and high-frequency power (HF) (LF:HF ratio) is recognized as an essential HRV parameter to indicate the sympathovagal responses [7]. However, most studies examining the validity and accuracy of HRV_{UST} demonstrate inconsistencies in LF, HF, and LF:HF ratio within 60-s time segments, compared to $\text{HRV}_{\text{criterion}}$ [19,23–25]. In contrast, in time-domain HRV analysis, excellent agreement and accuracy of HRV_{UST} has been demonstrated in the standard deviation of normal-to-normal intervals (SDNN) and RMSSD [19,23–25]. Esco and his colleagues [26] reported that 1-min HRV_{UST} of the natural logarithm of SDNN:RMSSD (LnSDNN:LnRMSSD) ratio was significantly correlated (0.72~0.86, very large correlation) to 5-min $\text{HRV}_{\text{criterion}}$ of natural logarithm of the LF:HF (LnLF:LnHF) ratio at both rest and recovery following a maximal graded exercise test in an athletic population. Following their finding, a potential implementation to use the HRV_{UST} of SDNN:RMSSD ratio as a surrogate to integrate the sympathovagal responses after maximal intensity exercise is feasible. Therefore, it is necessary to compare and contrast these two methods in relation to post-exercise evaluation of various exercise modalities.

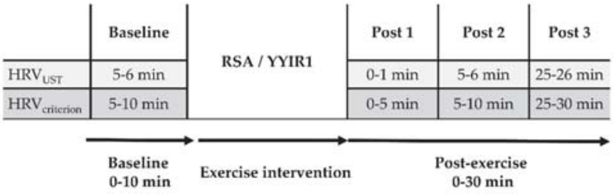
The physical tests, such as RSA and YYIR1, are extensively used to evaluate physical capacities and training adaptations in team sports [27,28]. Considering the lack of information regarding post-exercise HRV_{UST} records after field-based exercises, the primary purpose of this study was to investigate the agreement and acceptance of post-exercise time-domain (LnSDNN and LnRMSSD) and frequency-domain (LnLF and LnHF) HRV_{UST} measures after field-based shuttle running assessments (the RSA and YYIR1) in colleague male football players. The secondary purpose was to determine the relationship of ultra-short-term and short-term recordings of LnSDNN:LnRMSSD ratio to LnLF:LnHF ratio during post-exercise recovery. Based on findings in the previous studies, it was hypothesized that time-domain HRV_{UST} would show better agreement and reproducibility to the 5-min criterion of HRV record ($\text{HRV}_{\text{criterion}}$) than that of frequency-domain HRV. It was also hypothesized that HRV_{UST} of LnSDNN:LnRMSSD ratio would demonstrate significant correlations to $\text{HRV}_{\text{criterion}}$ of LnLF:LnHF ratio during post-exercise recovery in both exercise protocols.

2. Materials and Methods

2.1. Experimental Approach to the Problem

A pretest-posttest crossover design was used to examine the post-exercise HRV_{UST} after the RSA and YYIR1 exercise protocols. The period of HRV assessment was divided into baseline, post-exercise 0~5 min (Post 1), post-exercise 5~10 min (Post 2), and post-exercise 25~30 min (Post 3). The time segments of HRV recordings consisted of the first 60-s (HRV_{UST}) and the 5-min criterion ($\text{HRV}_{\text{criterion}}$). The nLnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio were used in time-domain analysis. In addition, the LnLF,LnHF, and LnLF:LnHF ratio were used in frequency-domain analysis. The relationship of LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio was examined at baseline and post-exerciser time points. Since the RSA and YYIR1 tests are common tools for exercise testing and fitness training, assessment of post-exercise HRV can help to understand the autonomic function during recovery. Comparison of these two methods could help us to improve our current knowledge of post-exercise recovery strategy.

The first 5-min ECG data in the baseline were discarded to prevent orthostatic effect as a standard process of HRV record. Figure 1 presents the experimental procedures in the present study.



	Baseline	RSA / YYIR1	Post 1	Post 2	Post 3
HRV_{UST}	5-6 min		0-1 min	5-6 min	25-26 min
$\text{HRV}_{\text{criterion}}$	5-10 min		0-5 min	5-10 min	25-30 min

Figure 1. Schematic presentation of experimental procedure in the present study. RSA = repeated sprint ability test; YYIR1 = Yo-Yo intermittent recovery test level 1; HRV_{UST} = ultra-short-term heart rate variability; $\text{HRV}_{\text{criterion}}$ = criterion of heart rate variability.

2.2. Experimental Procedure

Participants first visited the exercise performance laboratory for a familiarization. The first visit consisted of an experimental introduction, procedural habituation, and determination of physical characteristics. The second and third visits were experimental visits for the RSA and YYIR1 exercise protocols with 3~7 days apart. The sequence order of the exercise protocols was conducted via an online randomizer website (https://www.randomizer.org/). The participants were asked not to undertake vigorous exercise 24 h before the visits and to avoid caffeine-containing substances and smoking 2 h before the experiments.

Initially, the participants were prepared with a lead II ECG. The skin of the participants were cleaned with alcohol wipes prior to electrode application (Kendall™ 200 Series Foam Electrodes; Covidien, Mansfield, MA, USA). During the baseline measurement (10-min ECG recording), the participants were required to lie supine on a medical bed in a quiet research room. Subsequently, the participants performed a 5-min warm-up cycling exercise, which consisted of 50-watt power output with a pedaling rate of 60 revolutions per minute (Optibike Med; Ergoline, Germany). Afterwards, the participants wore their personal sports shoes and performed the RSA and YYIR1 exercise protocols indoors on artificial surface. After the exercise termination, post-exercise ECG was recorded immediately in a supine position for 30 min. Room temperature was controlled at 25 °C, and humidity was set within the range of 50~60%.

2.3. Participants

The sample size estimation was determined based on a priori type of power analysis using G*Power 3.1.9.4 software (G*Power, Düsseldorf, Germany) [29]. A means differences between two dependent measures with power of 80% and an alpha value of 0.05 in the two-tailed test were set to estimate the minimum number of participants. Based on a previous study [26], it indicated that a minimum of 10 participants was required to approach actual power of 0.85.

Twenty-five male college outfield football players voluntarily participated in the study (mean ± standard deviation: age = 19.80 ± 1.08 years; height = 173.87 ± 5.60 cm; body weight = 67.91 ± 8.17 kg; body fat = 16.04 ± 5.05%; players experience = 10.20 ± 1.66 years). The inclusion criteria included: (1) undertake regular football training at least five times a week; (2) a minimum football training experience of five years; and (3) currently playing in the first division of a national university championship. Exclusion criteria included: (1) any history of severe neuromuscular injury; (2) lower extremity injury within six months; and (3) current neurological or cardiovascular diseases. All players signed informed consent forms and were familiarized with experimental procedures one week before the experiment proper. The study has been approved by the Institute Review Board of the hospital (2014-06-003CC) and was conducted in accordance with the Declaration of Helsinki with its later amendments.

2.4. Heart Rate Variability

ECG lead set (SS2LB, Biopac Inc., Goleta, CA, USA) with a conventional lead II arrangement was used to record cardiac responses in a supine position. A data acquisition system (MP35, Biopac Inc., Goleta, CA, USA) was used to collect resting ECG signals for 10 min before warm up and 30 min immediately after the RSA and YYIR1 exercise protocols. The analog signals of ECG were transformed into digital signals by using an analog-to-digital converter with a sampling rate of 1000 Hz via the Biopac Student Lab system. The ECG waveforms were then filtered using Kubios HRV analysis software Premium version 3.2.0. (Kubios, Kuopio, Finland) to calculate the time-domain and frequent-domain HRV indices. The artefact correction of RRI was set at a threshold of medium level, and window width was set at 300 s, with window overlap of 50%. Smoothing priors set at 500 Lambda were used for the detrending method [30]. The area-under-the-curve of the spectral peaks within the range of

0.04~0.15 Hz and 0.15~0.40 Hz were set for LF and HF, respectively. The power spectra of RR intervals were calculated by means of Fast Fourier Transformation.

2.5. Yo-Yo Intermittent Recovery Test Level 1

The YYIR1 test consisted of 20-m shuttle runs back and forth between two lines, with a gradual incremental increase in speed over time. A jogging distance of 5 m recovery zone was set behind the start line, with participants given 10-s resting recovery after each bout of shuttle running. The running speed was controlled by digital audio bleeps from a laptop. The running speed of the first four bouts (0-m~160-m) was set at 10~13 $km \cdot h^{-1}$, and the subsequent seven bouts (160-m~440-m) were 13.5–14 $km \cdot h^{-1}$. After the 11th bout, the running speed increased by increments of 0.5 $km \cdot h^{-1}$ every eight bouts (i.e., after 760-m, 1080-m, 1400-m, 1720-m, etc.). The total covered distance was recorded when the participants failed to return to the start line a second time (a verbal warning was given to the participants in the first instance that they failed to make the line in time).

The validity of the YYIR1 test to assess aerobic capacity has previously been described [28] with test-retest reliability between 0.78 to 0.98 [31]. Each participant's predicted maximal oxygen consumption was calculated by the equation: distance in meters × 0.0084 + 36.4 [28].

2.6. Repeated Sprint Ability Test

The RSA test consisted of a distance of 20-m sprint back and forth, repeated 6 times with 20-s rest intervals between bouts. The participants were allowed to conducted passive recovery or jogging behind the starting line during the 20-s rest interval. Timing gates (Fusion Sport, Brisbane, Queensland, Australia) were placed 30-cm behind the starting line and set at a height of 1.2-m. Two preliminary trials to familiarize participants with the RSA protocol was given followed by a 5 min rest.

Sprint time for each RSA performance was recorded. The best and worst sprints times were recorded as RSA_{best} and RSA_{worst}, respectively. The mean sprint time of RSA test (RSA_{mean}) was calculated as the average of the six sprints times. The mean of the first, second, and third sprint times were recorded as the $RSA_{1–3mean}$, while the mean of the fourth, fifth, and sixth sprints were recorded as the $RSA_{4–6mean}$. The percent decrement of RSA was calculated by using the equation: $100 - (RSA_{totoal}/(RSA_{best} \times 6) \times 100)$. Fatigue index was calculated by using the formulae: $(RSA_{worst} - RSA_{best})/RSA_{best} \times 100$ [32].

The validity and reliability of the RSA test to assess the 20-m shuttle sprint ability in football players has previously been reported with an interclass correlation coefficients (ICC) value of 0.81 for RSA_{mean} [33].

2.7. Statistical Analyses

Descriptive data of the measured variables are presented as mean and standard deviation (SD). The normality of all variables of interest were assessed via Kolmogorov–Smirnov statistical tests. HRV variables are commonly found to be non-normal distributions. To adjust for this violation, a natural logarithm was used for HRV comparisons. Inter-differences of HRV_{UST} to criterion was analyzed by using effect size (ES) calculations. The level of ES was interpreted as trivial (0.0~0.2), small (0.2~0.6), moderate (0.6~1.2), large (1.2~2.0), and very large (>2.0) [34]. For reliability analysis, ICC with two-way random model and single measures were used to determine relative values of reliability. The level of ICC values were expressed as nearly perfect (0.9~1), very large (0.70~89), large (0.50~69), moderate (0.31~49), and small (0~0.3) [34]. Furthermore, the Bland–Altman plots were used to evaluate the upper and lower limits of agreements between the HRV_{UST} and the $HRV_{criterion}$ among time segments. In addition, the relationship of LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio between HRV_{UST} and $HRV_{criterion}$ during baseline, Post 1, Post 2, and Post 3 in both exercise protocols were assessed by using Pearson product-moment correlation coefficient (r). The threshold level of the correlation coefficient was determined as trivial ($r < 0.1$), small ($0.1 < r < 0.3$), moderate ($0.3 < r < 0.5$), high ($0.5 < r < 0.7$), very high ($0.7 < r < 0.9$), nearly perfect ($r > 0.9$), and perfect ($r = 1$) [34]. Statistical analyses were conducted using by SPSS® Statistics version 25.0 (IBM, Armonk, NY, USA) and Microsoft Excel 2016 (Microsoft Corporation, Redmond, WA, USA).

3. Results

3.1. Exercise Performance

Table 1 shows the descriptive statistics regarding exercise performance during the RSA and YYIR1 exercise protocols.

Table 1. Descriptive data of exercise performance of the participants.

Exercise Performance	Mean ± SD	CV (%)
$RSA_{1\text{–}3mean}$ (s)	7.31 ± 0.29	3.91
$RSA_{4\text{–}6mean}$ (s)	7.64 ± 0.27	3.47
RSA_{mean} (s)	7.48 ± 0.26	3.46
RSA_{total} (s)	44.86 ± 1.55	3.46
RSA_{best} (s)	7.17 ± 0.28	3.85
RSA_{worse} (s)	7.75 ± 0.26	3.30
$RSA_{decrement}$ (%)	4.26 ± 1.94	45.55
RSA fatigue index (%)	8.13 ± 3.38	41.53
YYIR1 distance (m)	1214.40 ± 290.95	23.96
YYIR1 $VO2_{max}$ ($ml{\cdot}kg^{-1}{\cdot}min^{-1}$)	46.60 ± 2.44	5.25

SD = Standard deviation; CV = coefficient of variation; RSA = repeated sprint ability; YYIR1 = Yo-Yo intermittent recovery test level 1; s = seconds; m = meters; $VO2_{max}$ = maximal oxygen consumption; $ml{\cdot}kg^{-1}{\cdot}min^{-1}$ = milliliter per kilogram per minute.

3.2. Heart Rate Variability

The results of the LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio during baseline, Post 1, Post 2, and Post 3 in the RSA and YYIR1 exercise protocols are presented in Table 2. The result of HRV_{UST} and $HRV_{criterion}$ comparison demonstrated trivial and small ES (LnSDNN = RSA: −0.06 trivial~−0.23 small, YYIR1: 0.10 trivial~−0.32 small; LnRMSSD = RSA: −0.07 trivial~−0.16 trivial, YYIR1: −0.17 trivial~0.26 small; and LnSDNN:LnRMSSD ratio = RSA: −0.00 trivial~0.18 trivial, YYIR1: −0.15 trivial~0.49 small). The ICC values showed very large and nearly perfect correlation in all comparisons, except LnSDNN:LnRMSSD ratio in the RSA baseline [0.69 (0.36; 0.83) large] and YYIR1 Post 3 [0.63 (0.27; 0.83) large]. In addition, limits of agreements showed relatively small values of bias in all comparisons [RSA: 0.01 (−0.07; 0.08)~−0.12 (−0.66; 0.41); YYIR1: −0.01 (−0.06; 0.05)~−0.16 (−0.57; 0.26)].

The result of HRV_{UST} and $HRV_{criterion}$ comparison demonstrated trivial and small ES (LnLF = RSA: −0.13 trivial~−0.54 small, YYIR1: 0.18 trivial~−0.47 small; LnHF = RSA: 0.07 trivial~0.52 small, YYIR1: −0.06 trivial~0.33 small; and LnLF:LnHF ratio = RSA: −0.12 trivial~−0.44 small, YYIR1: −0.04 trivial~−0.38 small) (Table 3). The ICC values showed large variations in the RSA test (−0.02 small~0.64 large) and YYIR1 test (0.22 small~0.68 large). In terms of the Bland–Altman analysis, the result showed limits of agreements showed relatively small bias in all comparisons [RSA: −0.02 (−0.60; 0.55)~0.65 (−4.19; 5.53); YYIR1: 0.03 (−0.92; 0.98)~−0.23 (−1.15; 0.67)].

Table 2. Natural logarithm of standard deviation of normal-to-normal intervals (LnSDNN), root mean square differences between adjacent normal R-R intervals (LnRMSSD), and LnSDNN:LnRMSSD ratio during 0–1 min and 0–5 min of time segments of the baseline, post exercise 0–5 min, 5–10 min, and 25–30 min in the repeated sprint ability test and YoY o intermittent recovery test level 1 protocols.

Period	Parameter	HRV_{UST} (Mean ± SD)	$\text{HRV}_{\text{criterion}}$ (Mean ± SD)	ES (95% CI)	ICC (95% CI)	Bias (±1.96 SD)
RSA Baseline	LnSDNN	3.89 ± 0.34	3.91 ± 0.37	−0.06 (0.61; 0.50) *	0.82 (0.64; 0.92) ‡	−0.02 (−0.43; 0.40)
	LnRMSSD	4.06 ± 0.43	4.09 ± 0.42	−0.07 (0.63; 0.48) *	0.90 (0.78; 0.95) §	−0.03 (−0.41; 0.36)
	LnSDNN:LnRMSSD	0.96 ± 0.05	0.96 ± 0.04	−0.00 (−0.55; 0.55) *	0.69 (0.36; 0.83) †	0.01 (−0.07; 0.08)
RSA Post 1	LnSDNN	1.62 ± 0.39	1.69 ± 0.44	−0.17 (−0.72; 0.39) *	0.88 (0.74; 0.94) ‡	−0.07 (−0.46; 0.33)
	LnRMSSD	1.27 ± 0.64	1.36 ± 0.61	−0.14 (−0.70; 0.42) *	0.88 (0.76; 0.95) ‡	−0.08 (−0.66; 0.49)
	LnSDNN:LnRMSSD	1.47 ± 0.48	1.39 ± 0.42	0.18 (−0.38; 0.73) *	0.81 (0.60; 0.91) ‡	0.08 (−0.47; 0.63)
RSA Post 2	LnSDNN	1.69 ± 0.52	1.82 ± 0.52	−0.23 (−0.78; 0.33) #	0.84 (0.65; 0.93) ‡	−0.12 (−0.66; 0.41)
	LnRMSSD	1.34 ± 0.66	1.40 ± 0.68	−0.09 (−0.64; 0.47) *	0.83 (0.65; 0.92) ‡	0.06 (−0.73; 0.84)
	LnSDNN:LnRMSSD	1.45 ± 0.48	1.51 ± 0.61	−0.11 (−0.66; 0.45) *	0.82 (0.60; 0.91) ‡	−0.07 (−0.74; 0.61)
RSA Post 3	LnSDNN	2.81 ± 0.53	2.89 ± 0.51	−0.15 (−0.71; 0.40) *	0.95 (0.85; 0.98) §	−0.09 (−0.39; 0.21)
	LnRMSSD	2.45 ± 0.65	2.55 ± 0.60	−0.16 (−0.71; 0.40) *	0.95 (0.87; 0.98) §	−0.10 (−0.44; 0.25)
	LnSDNN:LnRMSSD	1.17 ± 0.15	1.15 ± 0.11	0.15 (−0.40; 0.71) *	0.90 (0.71; 0.94) §	0.02 (−0.12; 0.16)
YYIR1 Baseline	LnSDNN	3.93 ± 0.34	3.88 ± 0.29	0.16 (−0.40; 0.71) *	0.90 (0.78; 0.95) §	0.05 (−0.22; 0.32)
	LnRMSSD	4.05 ± 0.40	3.98 ± 0.41	0.17 (−0.38; 0.73) *	0.93 (0.82; 0.97) §	0.07 (−0.21; 0.35)
	LnSDNN:LnRMSSD	0.97 ± 0.07	0.98 ± 0.06	−0.15 (−0.71; 0.40) *	0.89 (0.78; 0.95) ‡	−0.01 (−0.06; 0.05)
YYIR1 Post 1	LnSDNN	1.48 ± 0.32	1.53 ± 0.33	−0.15 (−0.71; 0.40) *	0.84 (0.68; 0.93) ‡	−0.05 (−0.41; 0.30)
	LnRMSSD	1.11 ± 0.44	1.23 ± 0.47	−0.26 (−0.82; 0.30) #	0.84 (0.64; 0.93) ‡	−0.12 (−0.59; 0.36)
	LnSDNN:LnRMSSD	1.47 ± 0.45	1.38 ± 0.42	0.20 (−0.35; 0.76) *	0.80 (0.59; 0.91) ‡	0.09 (−0.44; 0.62)
YYIR1 Post 2	LnSDNN	1.42 ± 0.45	1.56 ± 0.40	−0.32 (−0.89; 0.23) #	0.76 (0.48; 0.89) ‡	−0.14 (−0.68; 0.40)
	LnRMSSD	1.03 ± 0.46	1.09 ± 0.47	−0.13 (−0.68; 0.43) *	0.82 (0.64; 0.92) ‡	−0.06 (−0.61; 0.49)
	LnSDNN:LnRMSSD	1.56 ± 0.56	1.65 ± 0.68	−0.14 (−0.70; 0.41) *	0.74 (0.50; 0.88) ‡	−0.09 (−0.96; 0.78)
YYIR1 Post 3	LnSDNN	2.62 ± 0.52	2.67 ± 0.50	−0.10 (−0.65; 0.46) *	0.92 (082; 0.96) §	−0.05 (−0.45; 0.35)
	LnRMSSD	2.01 ± 0.57	2.16 ± 0.59	−0.26 (−0.81; 0.30) #	0.90 (0.66; 0.96) §	−0.16 (−0.57; 0.26)
	LnSDNN:LnRMSSD	1.34 ± 0.17	1.26 ± 0.15	0.49 (−0.07; 1.06) #	0.63 (0.27; 0.83) †	0.07 (−0.17; 0.32)

The level of effect size was symbolled as trivial (0.0–0.2) as *, small (0.2–0.6) as #, moderate (0.6–1.2) as = †, large (1.2–2.0) as ‡, very large (>2.0) as §. The level of interclass correlation coefficients was denoted small (0–0.3) as *, moderate (0.31–49) as #, large (0.50–69) as †, very large (0.70–89) as ‡, and nearly perfect (0.9–1) as §. RSA = repeated sprint ability test; YYIR1 = Yo-Yo intermittent recovery test level 1; HRV_{UST} = ultra-short-term heart rate variability; $\text{HR}_{\text{criterion}}$ = criterion of heart rate variability; SD = standard deviation; CI = confident interval; ES = effect size; ICC = intraclass correlation coefficient.

Table 3. Natural logarithm of low frequency power (LnLF), high frequency power (LnHF), and LnLF:LnHF ratio during 0–5 min of time segments of the baseline, post exercise 0–5 min, 5–10 min, and 25–30 min in the repeated sprint ability test and Yo-Yo intermittent recovery test level 1 protocols.

Period	Parameter	HRV_{UST} (Mean ± SD)	$HRV_{criterion}$ (Mean ± SD)	ES (95% CI)	ICC (95% CI)	Bias (±1.96 SD)
RSA Baseline	LnLF	3.70 ± 0.51	3.81 ± 0.40	−0.24 (−0.80; 0.32) #	0.27 (−0.13; 0.59) *	−0.11 (−1.21; 0.98)
	LnHF	3.90 ± 0.50	3.87 ± 0.40	0.07 (−0.49; 0.62) *	0.36 (−0.04; 0.66) #	−0.03 (−0.99; 1.04)
	LnLF:LnHF	0.98 ± 0.29	1.01 ± 0.22	−0.12 (−0.67; 0.44) *	0.36 (−0.05; 0.66) #	−0.02 (−0.60; 0.55)
RSA Post 1	LnLF	4.35 ± 0.28	4.39 ± 0.15	−0.18 (−0.73; 0.38) *	0.39 (0.01; 0.68) #	−0.04 (−0.53; 0.44)
	LnHF	2.56 ± 0.99	2.70 ± 0.65	−0.17 (−0.72; 0.39) *	0.57 (0.24; 0.78) †	−0.14 (−1.67; 1.38)
	LnLF:LnHF	2.44 ± 2.81	1.77 ± 0.63	0.32 (−0.23; 0.89) #	0.25 (−0.13; 0.58) *	0.65 (−4.19; 5.53)
RSA Post 2	LnLF	4.26 ± 0.28	4.39 ± 0.18	−0.54 (−1.12; 0.02) #	−0.02 (−0.35; 0.35) *	−0.13 (−0.78; 0.52)
	LnHF	3.04 ± 0.74	2.66 ± 0.69	0.52 (−0.04; 1.09) #	0.32 (−0.04; 0.62) #	−0.38 (−1.97; 1.22)
	LnLF:LnHF	1.53 ± 0.58	1.82 ± 0.71	−0.44 (−1.01; 0.12) #	0.56 (0.21; 0.78) †	−0.29 (−1.42; 0.85)
RSA Post 3	LnLF	4.27 ± 0.27	4.30 ± 0.16	−0.13 (−0.69; 0.42) *	0.44 (0.06; 0.71) #	−0.03 (−0.61; 0.55)
	LnHF	3.02 ± 0.73	3.11 ± 0.56	−0.14 (−0.69; 0.42) *	0.58 (0.25; 0.79) †	−0.09 (−1.36; 1.18)
	LnLF:LnHF	1.54 ± 0.58	1.45 ± 0.39	−0.18 (−0.37; 0.74) *	0.64 (0.34; 0.82) †	0.09 (−0.73; 0.91)
YYIR1 Baseline	LnLF	3.64 ± 0.57	3.87 ± 0.37	−0.47 (−1.04; 0.09) #	0.48 (0.12; 0.73) #	−0.23 (−1.15; 0.67)
	LnHF	3.95 ± 0.44	3.81 ± 0.40	0.33 (−0.23; 0.89) #	0.61 (0.30; 0.81) †	0.13 (−0.58; 0.85)
	LnLF:LnHF	0.95 ± 0.26	1.04 ± 0.21	−0.38 (−0.94; 0.18) #	0.58 (0.25; 0.79) †	−0.09 (−0.50; 0.33)
YYIR1 Post 1	LnLF	4.34 ± 0.44	4.43 ± 0.10	−0.27 (−0.84; 0.28) #	0.28 (−0.11; 0.60) *	−0.09 (−0.84; 0.66)
	LnHF	2.46 ± 0.95	2.61 ± 0.57	−0.19 (−0.75; 0.37) *	0.61 (0.30; 0.81) †	−0.14 (−1.49; 1.21)
	LnLF:LnHF	2.49 ± 2.80	1.81 ± 0.55	0.33 (−0.22; 0.89) #	0.26 (−0.13; 0.58) *	0.68 (−4.11; 5.47)
YYIR1 Post 2	LnLF	4.40 ± 0.21	4.47 ± 0.08	−0.43 (−1.00; 0.12) #	0.22 (−0.15; 0.54) *	−0.08 (−0.47; 0.32)
	LnHF	2.55 ± 0.79	2.35 ± 0.53	0.29 (−0.26; 0.85) #	0.47 (0.12; 0.72) #	0.20 (−1.15; 1.55)
	LnLF:LnHF	1.98 ± 0.94	2.01 ± 0.53	−0.04 (−0.60; 0.51) *	0.42 (0.03; 0.70) #	−0.04 (−1.66; 1.58)
YYIR1 Post 3	LnLF	4.33 ± 0.28	4.37 ± 0.14	−0.18 (−0.74; 0.38) *	0.43 (0.05; 0.70) #	−0.05 (−0.51; 0.42)
	LnHF	2.79 ± 0.77	2.83 ± 0.64	−0.06 (−0.61; 0.50) *	0.60 (0.27; 0.80) †	−0.04 (−1.30; 1.22)
	LnLF:LnHF	1.70 ± 0.57	1.67 ± 0.62	0.05 (−0.50; 0.61) *	0.68 (0.39; 0.85) †	0.03 (−0.92; 0.98)

The level of effect size was symbolled as trivial (0.0–0.2) as *, small (0.2–0.6) as #, moderate (0.6–1.2) as = †, large (1.2–2.0) as ‡, very large (>2.0) as §. The level of interclass correlation coefficients was denoted small (0–0.3) as *, moderate (0.31–49) as #, large (0.50–69) as †, very large (0.70–89) as ‡, and nearly perfect (0.9–1) as §. RSA = repeated sprint ability test; YYIR1 = Yo-Yo intermittent recovery test level 1; HRV_{UST} = ultra-short-term heart rate variability; $HRV_{criterion}$ = criterion of heart rate variability; SD = standard deviation; CI = confident interval; ES = effect size; ICC = intraclass correlation coefficient.

The results showed a broad range of Pearson correlation coefficient between the HRV_{UST} and $HRV_{criterion}$ of LnSDNN:LnRMSSD ratio and the HRV_{UST} and $HRV_{criterion}$ LnLF:LnHF ratio at different time points in either the RSA test ($r = 0.34$ moderate, $p = 0.09$~$r = 0.73$ very high, $p < 0.01$) or the YYIR1 test ($r = 0.32$ moderate, $p = 0.12$~$r = 0.89$ very high, $p < 0.01$). The HRV_{UST} LnSDNN:LnRMSSD ratio and the $HRV_{criterion}$ LnLF:LnHF ratio was significantly highly correlated with RSA Post 3 and YYIR1 Baseline. Moderate significant correlations were found between RSA Post 1 and 2, and YYIR1 Post 1, 2, and 3. In addition, the $HRV_{criterion}$ comparison between the LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio was significantly very highly correlated with RSA Baseline, YYIR1 Baseline, and YYIR1 Post 3. High significant correlations were found between RSA Post 1, 2, and 3, and YYIR1 Post 1. A moderate significant correlation was found at YYIR1 Post 2 (Figures 2 and 3).

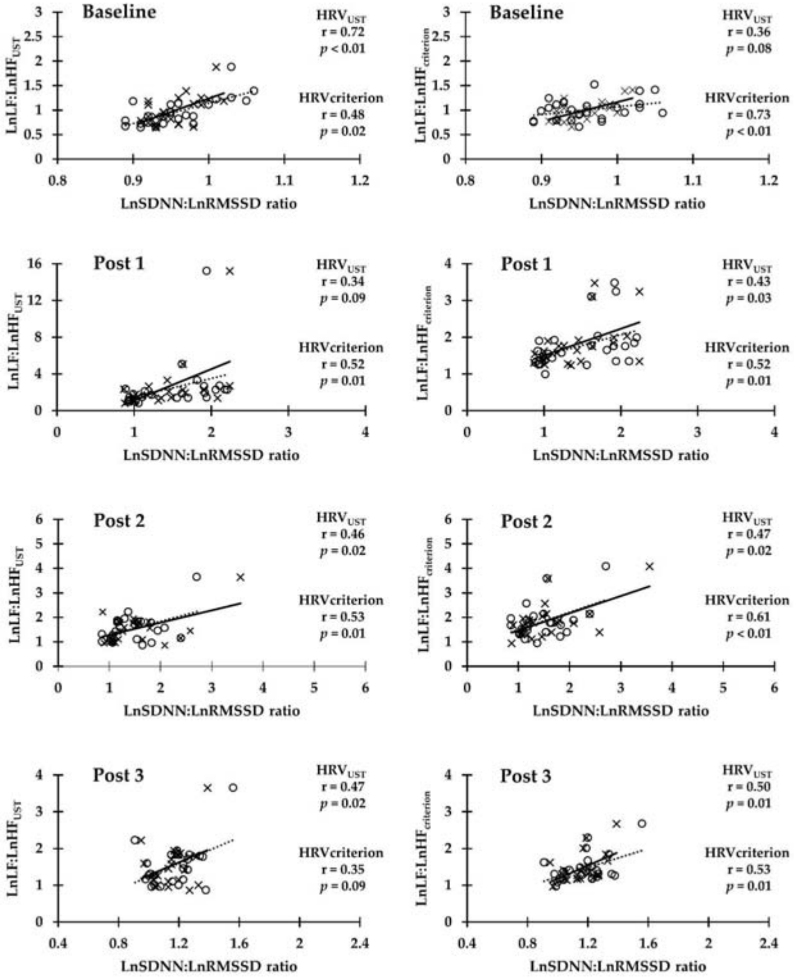


Figure 2. Pearson correlation coefficient between LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio (1 min and criterion) at baseline, post exercise 0–5 min (Post 1), post exercise 5–10 min (Post 2), and post exercise 25–30 min (Post 3) in repeated sprint ability test. Scatter plots between 1 min LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio are presented as open circles (dotted line). Scatter plots between criterion of LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio are presented as cross marks (solid line).

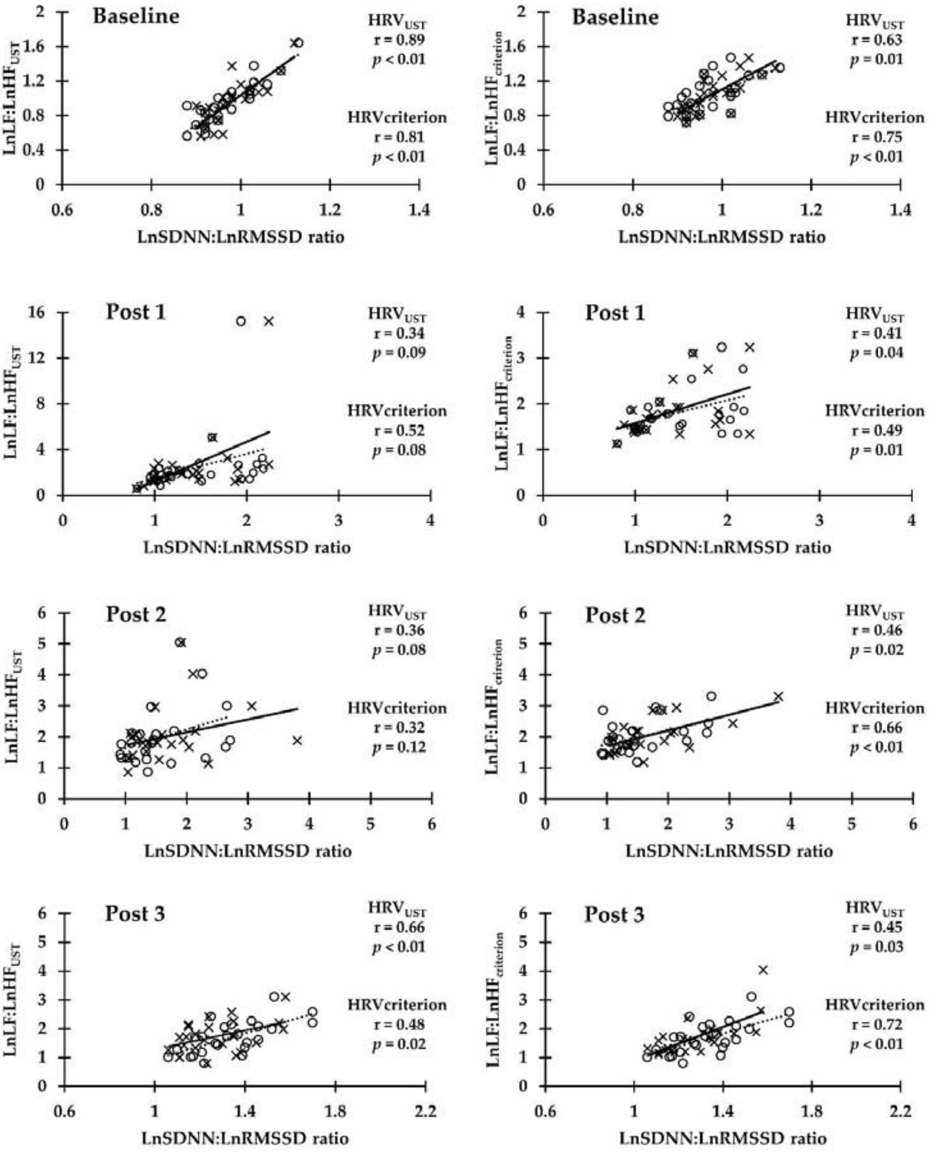


Figure 3. Pearson correlation coefficient of the relationship between LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio (1 min and criterion) at baseline, post exercise 0–5 min (Post 1), post exercise 5–10 min (Post 2), and post exercise 25–30 min (Post 3) in Yo-Yo intermittent recovery test level 1. Scatter plots between 1 min LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio are presented as open circles (dotted line). Scatter plots between criterion of LnSDNN:LnRMSSD ratio and LnLF:LnHF ratio are presented as cross marks (solid line).

4. Discussion

The primary purpose of this study was to examine the agreement and acceptance of post-exercise HRV$_{UST}$ parameters in LnSDNN, LnRMSSD, LnLF, and LnHF after field-based RSA and YYIR1 exercises.

The secondary purpose was to determine the correlation of ultra-short-term and short-term measures of the LnSDNN:LnRMSSD ratio to the LnLF:LnHF ratio during 30-min post-exercise recovery. The main findings of this study included: (1) 1-min HRV_{UST} in time-domain variables (LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio) showed excellent validity and reliability to the standard 5-min HRV assessment after the short-term bouts of anaerobic-based and aerobic-based intermittent running exercises. (2) A large variation of ICC values and correlation outputs in frequency-domain HRV variables (i.e., LnLF, LnHF, and LnLF:LnHF ratio) indicated an inconsistence of post-exercise HRV assessment in ultra-short-term and short-term evaluation. (3) One-minute and 5-min LnSDNN:LnRMSSD ratio scores were moderately to highly correlated with the 5-min LnLF:LnHF ratio scores during passive post-exercise recovery after maximal intensity short-term shuttle running.

4.1. Time-Domain Analysis

The findings in the present study supported our hypothesis that time-domain HRV_{UST} would demonstrate excellent acceptance and reproducibility to the $HRV_{criterion}$. The accuracy and agreement of 1-min LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD to the traditional 5-min $HRV_{criterion}$ measure was observed despite maximal exercise intensity after the RSA and YYIR1 tests. Moreover, our data revealed a gradual enhancement of the LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio when recovery time progressed after the RSA and YYIR1 tests. The augmentation of these time-domain variables indicates an increase in parasympathetic reactivation and a decrease in sympathetic activity during recovery [35]. Specifically, a rebound of LnSDNN, LnRMSSD, and LnSDNN:RMSSD ratio were observed at Post 3 in both exercise protocols. Previously, feasibility and agreement of 1-min HRV_{UST} to the standard 5-min HRV record was based on observations during resting measurement [12,13,15–17,36]. However, current knowledge regarding the methodology of the post-exercise HRV_{UST} is not well understood and has not been extensively explored. Esco et al. [12] investigated the reliability and limits of agreement of 10-s, 30-s, and 1-min HRV_{UST} during resting and after a maximal graded exercise test (Bruce protocol) in 23 college athletes. The results showed that 10-s and 30-s shorter time segments of HRV records decrease the agreement and hence the validity of the measurement compared to the standard 5-min $HRV_{criterion}$. However, acceptable levels of validity, and tight limits of agreement, were found in the 1-min HRV_{UST} during resting and post-exercise recovery. Another recent study revealed high reproducibility of test-retest 30-s HRV_{UST} recordings via LnSDNN, LnRMSSD parameters at the beginning of passive or active recovery after maximal graded cycling test [37]. As demonstrated in our findings, the HRV_{UST} measures showed trivial and small ES, very large and nearly perfect ICC values, and a very high level of correlation to $HRV_{criterion}$, even at the beginning of the post-exercise recovery. This observation may support the time efficient use of HRV assessment after maximal intensity of field-based shuttle running exercise. Since the SDNN and RMSSD are vagal-related HRV indices, the application of HRV_{UST} assessment to evaluated post-exercise parasympathetic reactivation may be considered in future research [35].

4.2. Frequency-Domain Analysis

It is interesting to note that the HRV_{UST} of LnLF, LnHF, and LnLF:LnHF showed trivial to small ES, small to large ICC, and trivial to very high correlation when compared to the $HRV_{criterion}$ in both exercise protocols. Our observations were in agreement with previous studies showing inaccurate measures of LF, HF, and LF:HF ratio in less than 60 s [19,23–25]. It appears that the large variation and inconsistent outcomes could limit the implementation of frequency-domain analysis in post-exercise assessment. One of the possible explanations to our finding is that the LF, HF, and LF:HF ratio is thought of as a sensitive measure to the respiratory frequency and thus could potentially undermine its validity and accuracy in HRV_{UST} measurement in our study [38]. In addition, the underlying mechanisms to determine post-exercise HRV modulation include sympathetic withdraw, parasympathetic reactivation, metaboreflex stimulation, baroreflex activity, and vascular regulation [35]. These mechanical factors may profoundly affect the frequency-domain variables in our study. It is also important to note that the length of data acquisition can influence the frequency-domain analysis [7]. Collectively, our findings suggest not to use the ultra-short-term records of LF, HF, and LF:HF ratio to evaluate HRV recovery.

4.3. Correlation Coefficient

The HRV_{UST} of LnSDNN:LnRMSSD ratio demonstrates significant correlations with $HRV_{criterion}$ of LnLF:LnHF ratio during post-exercise recovery in both exercise protocols. The 1-min LnSDNN:LnRMSSD ratio was significantly correlated to 5-min LnLF:LnHF ratio with moderate~high level during 30-min post-exercise recovery. The correlation coefficient between the HRV_{UST} of LnSDNN:LnRMSSD ratio and the $HRV_{criterion}$ of LnLF:LnHF ratio found in the present study led us to accept our secondary hypothesis. The advantage in using time-domain HRV indices is that it allows for high reproducibility when compared to that of frequency-domain HRV indices during post-exercise recovery [37]. The ratio of SDNN:RMSSD as an alternative of LF:HF ratio has previously been recommended in a longitudinal observation [39] and a cross-sectional study [26]. Esco and colleagues [26] reported that 1-min HRV_{UST} of the LnSDNN:LnRMSSD ratio was significantly correlated (0.72~0.86) to 5-min $HRV_{criterion}$ of LnLF:LnHF ratio at rest and recovery following a maximal graded exercise test in athletic population. Although the level of correlation was lower than those observed in Esco et al.'s study, an alternative of using LnSDNN:LnRMSSD ratio to frequency computations for evaluating post-exercise recovery in relation to sympathovagal balance is warranted.

4.4. Limitation

The limitations of this study were that: (1) the time window between study participation and training periodization varied among the participants. The fitness level and psychological status among the participants could be a potentials bias in this study. (2) Transition between the exercise termination and post-exercise HRV measurement was required in our experimental setting. The transition period may lead to a time delay of the true measurement at Post 1 point. (3) A homogeneous group of college male football players participated in this study. Application to general population and different sports athletes might be beyond the scope of the present study.

4.5. Practical Implications

The functional implication of the present study included that HRV_{UST} measures were acceptable to use immediately after shot-term shuttle running when the exercise intensity approached maximal intensity. The HRV is recognized as a convenient tool to assess the cardiac-related heath and recovery capacities in sports sciences and medicine. Implementation of 1-min post-exercise HRV assessment could help the profession of strength and conditioning to manage recovery duration when multiple rounds of maximal intensity exercise bouts are used.

Furthermore, strength and conditioning practitioners attempt to manage the efficiency of physical testing schedule in elite sports training. In fact, maximal intensity of aerobic or anaerobic tests are usually conducted in a separate day (i.e., RSA and YYIR1) in order to avoid potential fatigue to affect outcome of measurement. Monitoring post-exercise recovery via HRV_{UST} measures might be an alternative to understand the optimal time allocation among testing protocols (resting interval between the tests). Future studies are needed to investigate the relationship of HRV recovery and aerobic-based and anaerobic-based performance associated with rest duration.

5. Conclusions

In conclusion, for accuracy of HRV measurements, the post-exercise 1-min HRV assessment in LnSDNN, LnRMSSD, and LnSDNN:LnRMSSD ratio was a valid and reliable alternative to the 5-min HRV in either the RSA test or the YYIR1 test. Using the 1-min HRV assessment as a surrogate to the 5-min HRV should be cautioned in the RSA and the YYIR1 exercise protocols due to a wide range of ICC values during resting states and post-exercise recovery. The moderate to high correlation coefficient of 1-min and 5-min LnSDNN:LnRMSSD ratios to the 5-min LnLF:LnHF ratios indicate a potential ability to utilize the shortened HRV measurements after short-term anaerobic or aerobic shuttle running.

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Can the Performance Gap between Women and Men be Reduced in Ultra-Cycling?

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Abstract: This study examined a large dataset of ultra-cycling race results to investigate the sex difference in ultra-cycling performance (100 to 500 miles) according to age and race distance. Data from the time period 1996–2018 were obtained from online available database of the ultra-cycling marathon association (UMCA), including distance-limited ultra-cycling races (100, 200, 400, and 500 miles). A total of 12,716 race results were analyzed to compare the performance between men and women by calendar year, age group (18–34, 35–44, 45–59, and 60+ years), and race distance. Men were faster than women in 100 and 200 mile races, but no sex differences were identified for the 400 and 500 mile races. The performance ratio (average cycling speed$_{men}$/average cycling speed$_{women}$) was smaller in the 200 mile races compared to the 100 mile races and remained stable in the 400 and 500 mile races. In all race distances, the difference in average cycling speed between women and men decreased with increasing age. The gender gap in performance was closed in several distance-limited ultra-cycling races, such as the 400 and 500 mile races.

Keywords: ultra-endurance; performance; gender; cycling speed

1. Introduction

Ultra-endurance events (e.g., ultra-running, ultra-cycling) have gained increasing popularity over the last 25 years, with a rising number of female and master athlete participants in particular [1,2]. By definition, and according to the Ultra Marathon Cycling Association (UMCA, www.ultracycling.com), an ultra-cycling race is a race of more than 100 miles or lasting more than six hours in duration [3]. In ultra-cycling races, race distances spread from six-hour challenges up to 5000 km, such as the well-known Race Across America (RAAM). In this sports discipline, the participation of women ranges from 3% to 11%, depending on the race distance [2]. The majority of successful finishers in ultra-cycling events are master athletes in the age group of 35–49 years [1].

Sex differences in sports science have been well investigated [4–7]. The extent of the performance sex difference depends on the sport modality [8]. In ultra-cycling, women are slower than men overall [9]. Regarding an analysis of the "RAAM" (5000 km), the "Furnace Creek" 508 (800 km), and the "Swiss Cycling Marathon" (715 km), the average sex difference in ultra-cycling represents ~18%–28% [2]. An investigation of performance trends in the "Swiss Cycling Marathon" (62–560 miles) showed that sex differences decreased over the years 2001 to 2012, reaching ~14% in 2012 [10].

Sex differences in ultra-endurance performance were also investigated for other sports disciplines. Performance sex differences in running vary by race distance, where the longer the race distance, the smaller the sex difference [11]. According to iaaf.org, the gender gap in the 100 m sprint lies at ~10%, which decreases to ~9% at a marathon distance and to ~6% in ultra-marathon running (iau-ultramarathon.org). In ultra-running, the sex difference lies at ~11%–12% [12–14]. In long-distance triathlons, specifically the Ironman record in "Ironman Hawaii", an even smaller sex difference of ~10.3% in performance was identified [5]. Furthermore, the gender gap was able to be closed in several time-limited ultra-marathons over the past 40 years, such as 6, 72, 144, and 24 h races [11]. Several studies in swimming demonstrated that the gender gap could be closed in ultra-endurance sports with increasing age [15,16]. In ultra-cycling, however, there is still a lack of knowledge regarding whether sex differences can be reduced with increasing age [12].

Aging is accompanied by a decline in physical function. This decline in performance is due to physiological mechanisms, such as a decrease in maximum aerobic capacity (VO_{2max}), lactate threshold, exercise economy [17], and skeletal muscle mass [18]. Interestingly, the age-related performance decline depends on the mode of locomotion. The smallest age-related decline in performance was found in cycling when comparing swimming, running, and cycling [19]. Tanaka and Seals [17] demonstrated a curvilinear decrease in age-related performance up to the age of 60 years, whereupon performance decreased exponentially thereafter when analyzing results in running and swimming for both genders. Ransdell et al. [8] analyzed cycling results (i.e., 200 m and 500 m track and a 40 km road time-trial) and showed that this exponential decline after the age of 55 years occurred to a greater extent in female athletes. Spina et al. [20] found that both the cardiovascular response and adaption to endurance exercise training are better in elderly men than in elderly women.

The age at peak performance and the sex differences in ultra-endurance performance vary between sports. In ultra-cycling, the age at peak performance seemed to be similar for both sexes, specifically ~37 for women and ~38 for men, according to an analysis of a 24-hour ultra-cycling draft-legal event held in Switzerland [10]. In 50 km ultra-running races, men achieved peak performance at a younger age (30–39 years) than women (40–49 years) [21]. This was in contrast to ultra-swimming, where no sex difference in peak performance was demonstrated in the elderly age groups (30–39 years, 40–49 years, and 50–59 years) [22,23].

Based on existing literature, we know that female endurance performance generally declines to a greater extent compared to male endurance performance [8]. This work relied on an enormous number of ultra-cycling races to analyze trends in ultra-cycling with special regard to the gender gap according to race distance and age. This is the first study to include such a large number of participants throughout different race distances (100–500 miles) [10].

2. Materials and Methods

2.1. Ethical Approval

All procedures used in this study were approved by the Institutional Review Board of Kanton St. Gallen, Switzerland, with a waiver of the requirement for informed consent of participants given the fact that the study involved the analysis of publicly available data.

2.2. Data Sampling

Data were collected from publicly available data (World Ultra Cycling Association) [24]. We used the calendar of the Ultramarathon Cycling Association to include the most common races in ultra-cycling all over the world. Data from 1996 to 2018 were obtained from the official race websites. Required information in the original dataset included year of competition, gender, age group, race distance, and racing time. Other data were excluded due to vague age grouping. Based on the different race distances, cycling speed was used as a comparable variable. Data of 12,716 athletes throughout 45 different events was analyzed (Table A1). The whole dataset was cleaned for double coding results. Unfortunately, much of the European data was removed due to missing necessary information. For example, known races such as the Race across Italy, Tortour in Switzerland, the Race across Germany, and Slo24 Ultra had to be excluded. We did not distinguish between nationalities in this study.

2.3. Statistical Analysis

Data were tested for normality and homogeneity using the Shapiro–Wilk and Levene's tests, respectively. Data were expressed as means and standard deviations (±SD). An analysis of variance (one-way ANOVA) compared the performance between different age groups and different race distances. Additionally, Student's t-test for independent samples was applied to compare performances between men and women. Three general linear models were performed (two-way ANOVA) according the following interactions: model 1 = gender × year; model 2 = gender × race distance; model 3 = gender × age group. Performance ratios were calculated according to male and female performance (average cycling speed$_{men}$/average cycling speed$_{women}$), with a higher performance ratio indicating a greater sec difference. Statistical significance was set at 5% ($p < 0.05$). The Statistical Software for the Social Sciences (SPSS v23.0 IBM, Chicago, IL, USA) and GraphPad Prism v.6.0 (GraphPad Software, San Diego, CA, USA) were used for all analyses.

3. Results

The number of participating athletes by calendar year for men and women showed greater male participation in all distance races, with 11,347 (91.05%) men and 1116 (8.95%) women (Figure 1).

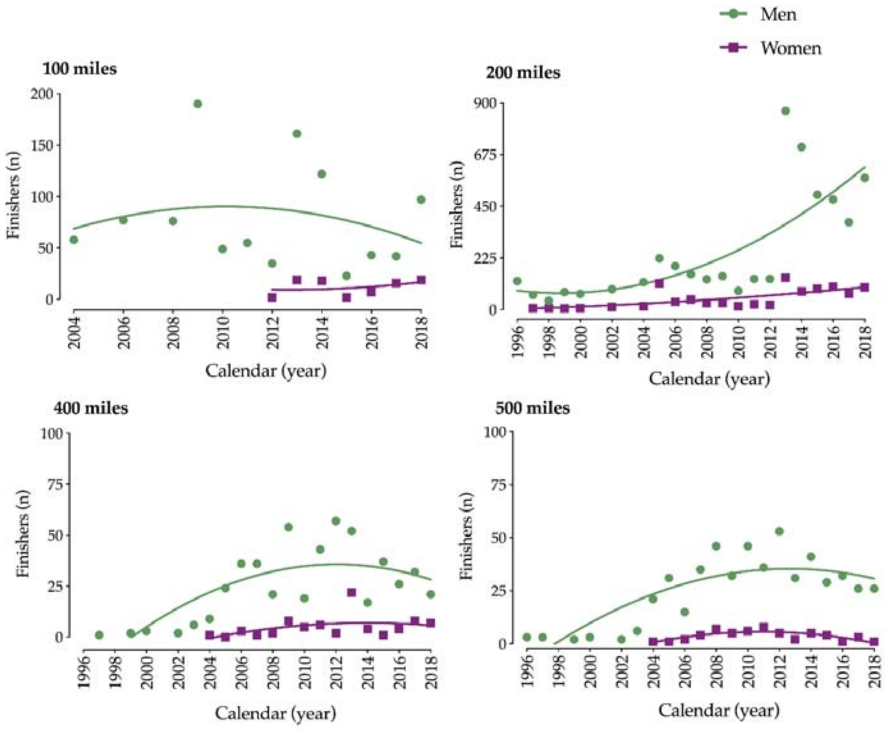

Figure 1. Number of male and female participants in ultra-distance cycling in 100–500 mile races from 1996 to 2018. * Outlier point (n = 190).

In the first model (gender × year), a gender-effect was identified in 200 (F = 22.0, $p < 0.001$ and 400 mile (F = 7.4, $p = 0.010$) races. Moreover, a year effect and interaction between gender and year were found for 200 (F = 9.2, $p < 0.001$; F = 2.3, $p < 0.001$) and 400 (F = 3.3, $p = 0.014$; F = 1.9, $p = 0.025$) mile races, respectively (Figure 2).

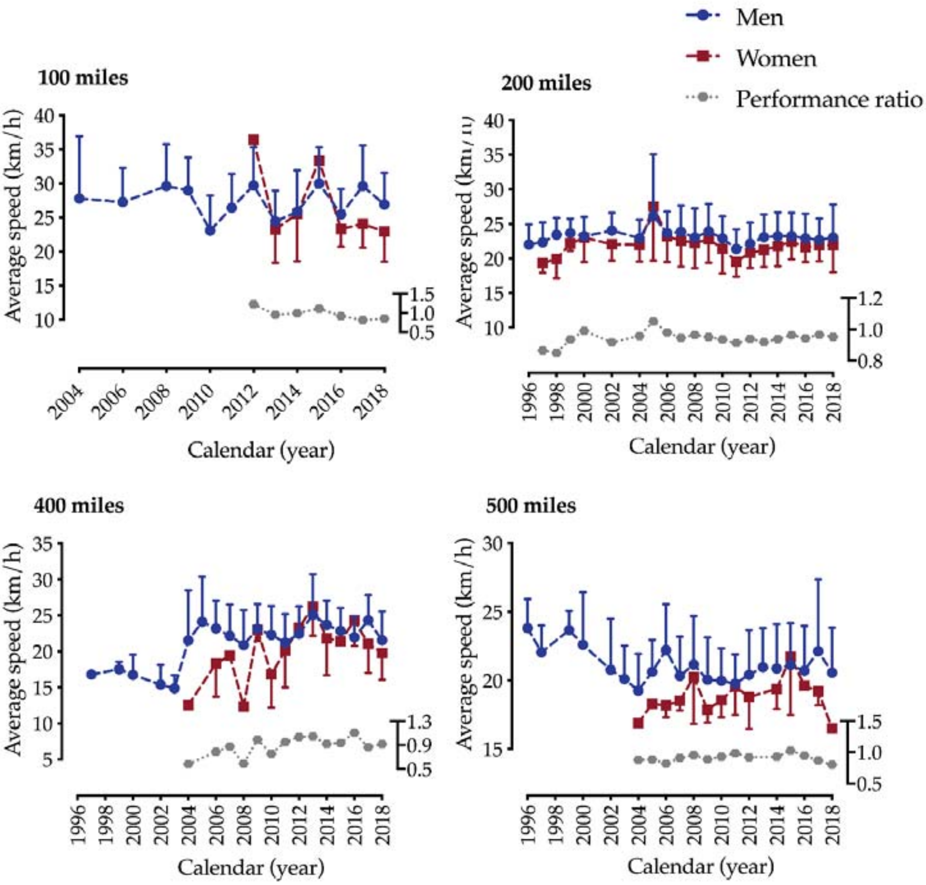

Figure 2. Performance trends of male and female participants in ultra-distance cycling in 100–500 mile races from 1996 to 2018. Data expressed as means and standard deviations.

The second model (gender and race distance) showed a trend for a race distance effect (F = 6.0, $p = 0.087$) and an interaction effect (F = 15.4, $p < 0.001$); pairwise comparisons showed that men were faster than women in 100 mile and 200 mile races (Figure 3). The third model (gender and age group) showed a significant gender effect for 100 mile races (F = 9.1, $p = 0.026$), and age group effects were identified in 200 mile races (F = 9.1, $p = 0.028$). Interaction effects were shown in 100 mile (F = 7.1, $p < 0.001$) and 200 mile (F = 4.1, $p = 0.002$) races. Post-hoc analyses showed a trend regarding men in the younger age group being faster ($p < 0.05$) than the adjacent older age group in 100 mile and 200 mile races, but no differences were observed between the 18–34 and 35–44 age groups in 400 mile and 500 mile races. For the women, the fastest age group in the 100 miles races was 45–59 and 35–44 in the 400 mile races. For the 200 miles races, the female younger age group was faster ($p < 0.05$) than the adjacent older age group (Figure 4). Moreover, post-hoc analyses showed that men were faster in the 18–34, 35–44, and 60+ age groups in 100 miles races; in 200 mile races, men were faster only in the 45–59 age group, and in 400 and 500 mile races men were faster only in the 18–34 age group.

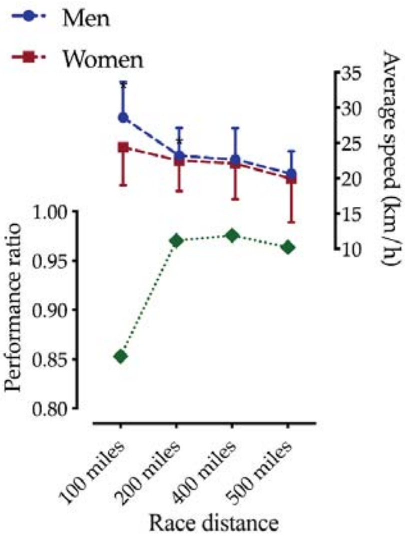

Figure 3. Comparison of performance level between men and women participating in ultra-distance cycling (100–500 mile races). * Statistical difference between men and women. Data expressed as means and standard deviations.

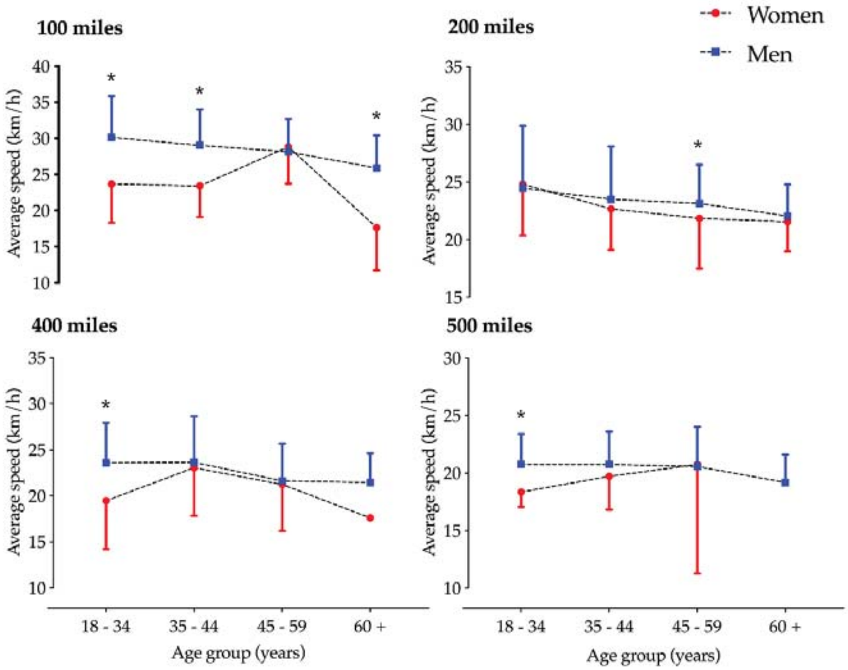

Figure 4. Performance trend by gender and age groups of participants in ultra-distance cycling (100–500 mile races). * Significant difference between men and women ($p < 0.05$) in post-hoc analysis.

4. Discussion

We investigated the impact of age, gender, and race distance on performance in ultra-cycling by analyzing a large dataset including 12,716 results from 45 different ultra-cycling races from 1996 to 2018. We hypothesized a decrease in the gender gap with increasing age and increasing race distance. The main findings were that (i) men had the highest participation in ultra-cycling races over all distances, (ii) men were faster than women over all distances in the majority of the years, (iii) men were faster than women in 100 and 200 mile races, (iv) no sex differences were identified for the 400 and 500 mile races, (v) the performance ratio decreased from the 100 mile races to the 200 mile races and remained stable in the 400 and 500 mile races, and (vi) men showed a higher average cycling speed than women over all race distances (100–500 mile races), with the difference in average cycling speed between women and men decreasing with increasing age. To sum up, the gender gap in ultra-cycling narrowed with increasing age and increasing racing distance.

4.1. The Gender Gap was Reduced in Longer Ultra-Cycling Race Distances

An important finding was that the gender gap was able to be reduced in longer distance-limited ultra-cycling races (200, 400, and 500 mile races). Previous studies struggled to examine whether women could outperform men in the future [4]. Physiological, hormonal, and genetic factors responsible for gender differences must be considered [25,26], such as women having smaller hearts, lower bone density, shorter stride length, lower cardiac output, smaller lungs, lower lung capacity, smaller muscles in relation to body size [19], lower testosterone levels resulting in higher body fat percentage, and lower hemoglobin concentrations resulting in lower anaerobic capacity [25,27,28].

However, in ultra-endurance sports, some of the above-mentioned anthropometric and physiological characteristics can be of advantage, for example, a higher percentage of body fat serving as an energy store [29]. Furthermore, ultra-endurance sports are less dependent on anaerobic energy supply, which is preserved by the impact of testosterone. Physiological conditions result in a gender gap that is the highest over short distances and drops with increasing running distance (according to iaaf.org and iau-ultramarathon.org). Similar findings in this study were that men were faster than women in shorter ultra-cycling distances, such as 100 and 200 mile races, but no significant differences were identified in the 400 and 500 mile races. Thus, gender has an influence on performance, with the performance ratio (PR, average cycling speed$_{\text{men}}$/average cycling speed$_{\text{women}}$) dropping from 100 mile (PR: 1.1633) to 200 mile races (PR: 1.0285) and remaining stable in 400 (PR: 1.0217) and 500 mile (PR: 1.0403) races; therefore the gender gap was greatest in the 100 mile races.

4.2. Participation Patterns in Ultra-Cycling According to Gender

The number of participating athletes in distance-limited ultra-cycling races was unstable, with the highest participation of men over all race distances; there were 11,347 (91.04%) men and only 1116 (8.95%) women (Figure 1). This finding was congruent with other investigations regarding participation trends in ultra-cycling events [1,2] where ~3–11% were female finishers. This lag in participation could partially be explained by the fact that women were not allowed to participate in sports competitions for many years due to previous emancipation, historical, and social reasons [2]. Furthermore, the gap in participation could be explained by different exercise behaviors between women and men, whereby men seem to seek more strenuous exercise involvement [30].

4.3. Average Cycling Speed and Performance Ratio According to Gender and Age

The average cycling speed dropped from one age group to another in 100 and 200 mile races, but there was no similar trend in the 400 and 500 mile races. A similar conclusion was drawn in a former study investigating ultra-cycling races, indicating that the fastest cyclists were younger and participated in shorter races [31]. The performance ratio was the lowest for the age category 34–44 years (PR: 0.8163), increasing for the age group 45–59 years (PR: 0.8818), and even more so for the 60+ age group (PR: 0.9178), thereby showing the narrowing of the gender gap in ultra-cycling according to increasing age.

4.4. Limitations, Strengths, and Implications for Future Research

The strength of this study lies in the large dataset including different race distances, different races, and a huge number of athletes of different performance levels studied over a longer period of time (1996–2018). The difference and the ratio between men and women should be interpreted with caution since there were significantly more data (participants) for men, which may have inappropriately skewed performance data in the small sub-analyses. This cross-sectional study was limited because we were unable to take into consideration other aspects influencing cycling performance, such as previous training, equipment, weight, weather, topographic characteristics, nutrition, and motivation. Nevertheless, this additional information would not have changed our results because we compared the performance gap between men and women. In this study, we demonstrated the influence of age on the gender gap. The fact that both the performance ratio and the gender gap were smallest in the longest race distances and the oldest athletes may empower athletes older than 45 years to compete in longer race distances.

5. Conclusions

In distance-limited ultra-cycling races (100–500 miles) held from 1996 to 2018, men were faster than women for all race distances. Men showed greater participation than women. The gender gap was the highest in the 100 mile races and narrowed in longer race distances (200, 400, and 500 miles). The gender gap decreased in distance-limited ultra-cycling races when race distance and age increased. This finding may empower women to compete in longer race distances to keep up with male competitors. Further studies are necessary to investigate the high participation lag in ultra-cycling races between men and women. Furthermore, it would be of interest to know whether the gender gap equally narrows in time-limited ultra-cycling races and in ultra-cycling races longer than 500 miles, such as the RAAM.

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Appendix A

Table A1. Supplemental table with all included distance-limited ultra-cycling races.

Race	N (Women)	N (Men)	N (Total)
Adirondack540	9	72	81
Bessies Creek	2	10	12
Bharat Alacrity	2	4	6
Bike Sebring	11	39	50
Black and Blue Double Century	3	31	34
Calvins Challenge	0	59	59
Camino Real Double Century	37	205	242
Colorado Triple Crown	15	182	197
Deccan Cliffhanger	1	6	7
Dolomitica	4	12	16
Eastern Sierra Double Century	75	342	417
Fireweed - Race across Alaska	48	452	500

Table A1. *Cont.*

Race	N (Women)	N (Men)	N (Total)
Glocknerman	1	141	142
Gran Fondo la Fortuna	4	46	50
Gran Fondo Las Vegas	5	200	205
Grand Loop	9	45	54
Great Alaska Double Century	162	322	484
Great Alaska Double Century F	31	90	121
Heart of the South	6	69	75
Hemet Double Century	305	2046	2351
Hoodoo 500	7	77	84
Joe Bar 500	0	2	2
Lake Taupo	7	87	94
Mid Atlantic	8	49	57
Mid-Atlantic 100 mile TT	7	35	42
Minnesota RAAM Challenge	9	9	18
Mulholland Double Century	11	86	97
NCOM Ultra Bicycle Race	4	26	30
No country for old men	0	5	5
Ohio RAAM Challenge	4	27	31
RAAM Challenge Minnesota	1	22	23
RAAM Challenge North California	2	4	6
RAAM Challenge North Florida	2	11	13
RAAM Challenge Ohio	2	25	27
RAAM Challenge Oregon	0	5	5
RAAM Challenge Texas	1	20	21
Race across Oregon	13	26	39
Saratoga	9	58	67
SoCal RAAM Challenge	3	14	17
Solvang Double Century	287	1721	2008
Swiss Cycling Marathon	6	125	131
Taupo Bike Race	0	4374	4374
Terrible Two	10	167	177
Texas RAAM Challenge	9	42	51
Texas Time Trial	18	140	158
Texas Time Trials	5	31	36
Total	**1155**	**11,561**	**12,716**

RAAM = Race Across America.

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Article

The Age-Related Performance Decline in Ironman 70.3

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Abstract: Although the age-related decline in sport events has been well studied, little is known on such a decline in recreational triathletes for the Half Ironman distance. Indeed, the few existing studies concentrated on specific aspects such as top events, elite groups, some consecutive years, single locations, or age categories instead of analyzing all the data available. Therefore, the aim of the present study was to examine recreational triathletes' performance in three split disciplines (swimming, cycling, and running) as well as in overall race time by analyzing all data of Half Ironman finishers found on ironman.com (i.e., 690 races; years 2004 through 2018; 206,524 women (24.6%) and 633,576 men (75.4%), in total 840,100 athletes). The age-dependent decline in Half Ironman started earliest in swimming (from the very first age group on) with a smallest age group delta between 35–49 years in men and 40–54 years in women. The performance decline started at 26 and 28 years in men and women for running; at 34 years for men and 35 years for women in cycling; and at 32 years for men and 31 years for women with regard to overall race time. The results may be used by coaches and recreational athletes alike to plan a triathlon career.

Keywords: half ironman; ironman 70.3; age-dependent performance decline; masters athletes; triathlon; endurance; multi-sports; split disciplines

1. Introduction

Triathlon, a multidiscipline endurance sport (i.e., swimming, cycling, and running), has several distances: the Olympic distance (1.5 km swimming, 40 km cycling, and 10 km running), the Half Ironman distance (1.9 km/1.2 miles swimming, 90 km/56 miles cycling, and 21.1 km/13.1 miles running, the full Ironman distance (3.8 km swimming, 180 km cycling, and 42.2. km running), and distances longer than the Ironman distance, so-called ultra-triathlon events [1,2]. Triathlon performance is influenced by different factors such as personal best time, previous experience, sex, training, nationality, anthropometrical and physiological characteristics, pacing, performance in split disciplines, and age, which is considered the most important predictor of performance [3–6].

Peak performance in many sports—except the most explosive ones, which peak much earlier—is observed just before 30 years [7]. In Ironman triathlon, though, performance seems to plateau until approximately 35 years to be followed by a moderate negative slope for the next two decades and afterward thought to be steep [4,8–11]. This decline seems to be more prominent in women than in men, in longer distances than in shorter ones [9] as well as in off-road races than in road-based events [8,11]. It differs between split disciplines, which is due to their specific demands and because they differently challenge an aging athlete's physiology, whereby the swim decline seems to be highest (when the focus is laid on recreational athletes) [12], followed by running decline, with cycling being easiest to keep performance in, as a study on the top 20% of non-elite athletes shows [11].

The age, at which age groups in each split discipline start to deteriorate, depends on the cohort investigated. For example, Lepers et al. [10,13] investigated swimming, cycling, running, and overall race times of the top ten male triathletes between 20 and 70 years of age (in 5-year intervals) for two consecutive Ironman World Championships (2006 and 2007). Their results showed a smaller age-related decline in cycling performance compared with that in running and swimming after 50 years of age for the Ironman distance. With advancing age, the performance decline was less pronounced in cycling (>55 years) and running (>50 years) [14]. On the other hand, according to another study on split and overall race times of 329,066 male and 81,815 female athletes from over 253 different Ironman triathlon races, the female age-related swim decline started much earlier, in the age group 25–29 years, and in the age group 30–34 years for cycling, running, and overall race time, whereas for men it started at the age group 25–29 years in swimming as well as in the age group 35–39 years in cycling, running, and overall race times [12]. Obviously, the selection of top athletes in elite races led to a considerable bias, which is revealed when considering all finishers of all races held worldwide.

The age of the fastest finishers in the “Ironman Hawaii” (i.e., the Ironman World Championship) has been increasing within the last decades. Meanwhile, these athletes’ overall race time improved [4,9,15]. Similarly, non-elite elderly age group (so-called masters) athletes have improved their performance in the “Ironman Hawaii” [9,15]. Masters athletes are typically defined as being older than 35–40 years [15,16] or as athletes of any age older than when world records are usually won [4,17]. There is a large increase in masters athlete participation rates over the last three to four decades [18].

Masters athletes gain increasing scientific interest. Their ability to maintain performance might help us understand health promotion better. Although a lot of research has been performed with respect to how age influences athletic performance in general and endurance events in particular, little data exist on its impact on performance in the Half Ironman triathlon, a relatively young sports event. Thus far, three studies have investigated age-related Half Ironman triathlon performance declines [9,18,19], focusing on aspects such as top events (i.e., World Championships or Olympic events [18]), elite groups (i.e., the top ten [9]), some consecutive years [18], or single locations [11,18,19]. Another one uses 10-year instead of 5-year age group categories [11]. This gap in turn means that we do not know how the normal recreational athlete might perform. Non-elite athletes are highly heterogenous and differ in many aspects—training status [20], nutritional status [21], experience [22,23]. Although the performance density of elite athletes is higher and therefore more reliable than that of recreational athletes, to our understanding we have no better data to extrapolate future recreational performance. This is due to the fact that elite and non-elite athletes cannot be compared due to several differences among the groups, most of all that elite athletes are younger than masters athletes, but that there are also differences in nutritional and training status as well as in mental preparation and strength (see also Section 4.10—limitations, practical applications, and implications for future research).

To the best of our knowledge no study exists so far that investigated the data of all Half Ironman races available on the official website ironman.com. In mass endurance events, only about 1% of the athletes are elitist. This number is taken from an Ironman study [12] but might serve as a rule for Half Ironman as well. As almost every athlete of our dataset taken from ironman.com is non-elitist (i.e., recreational), and many herein are amongst the group of masters athletes, we decided to study this important group. This approach might offer coaches and athletes the ability to plan a career and extrapolate the time as to when to change to a longer distance since it is known that older athletes place better in longer races [9].

We expected the age-related performance decline in Half Ironman races to start earlier than in Ironman triathletes according to Knechtle et al. who investigated the Olympic, Half Ironman, and Ironman distance; Wu et al., who report results for the same distances; and Käch et al. studying the decline pattern in triathlon [9,11,12]. Furthermore, it was assumed that recreational Half Ironman performance would follow these patterns: (i) swimming decline starting first, followed by running with cycling coming last (referring to the study by Käch et al. on recreational triathletes [12]); and (ii) the respective decline curves in women starting earlier and being more prominent than those of men [12]. This study might serve to increase our understanding of the performance decline in recreational Half Ironman triathlon that comes with aging.

2. Materials and Methods

2.1. Ethical Approval

This study was approved by the Institutional Review Board of Kanton St. Gallen, Switzerland, with a waiver of the requirement for informed consent of the participants as the study involved the analysis of publicly available data.

2.2. Data Sampling

We analyzed successful finishers of all Ironman 70.3 races recorded on the Ironman's website (www.ironman.com/im703-races) between 2004—the first year under the official Ironman label—and 2018. The complete dataset of races with the official Ironman 70.3 label has, to the best of our knowledge, not yet been investigated. A total of 690 races (327 in North America, 67 in South America, 144 in Europe, 71 in Asia, 53 in Oceania, 7 in the Middle East, and 6 in Africa) were identified by using a self-programmed Visual Basic script facilitating data sampling.

Some races' report data had to be transformed from PDF to Excel format using pdftoexcel.com. Some of these Excel-converts had to be adjusted to the correct order by hand. Transition times have not been considered. Exclusion criteria were (i) athletes who did not start; (ii) disqualified athletes; (iii) athletes with at least one missing split time; (iv) athletes who did not finish; (v) inappropriate time for an age group (i.e., split time much faster than the appropriate time of the winner of the race, possibly due to technical recording issues); (vi) elite athletes; (vii) athletes aged 75 years old or older. In the end, 206,524 women (24.6%) and 633,576 men (75.4%) were included, totalizing 840,100 athletes' data as the basis for the following results.

2.3. Statistical Analysis

The data were tested for normality and homogeneity with Kolmogorov–Smirnov and Levene's tests, respectively. Two-way ANOVA was applied to compare performance data of swimming, cycling, running, and overall using sex as a fixed factor and age group as a random factor (age groups M20 and 75 and above were excluded because of insufficient sample). Delta percentage ($\Delta\%$) was calculated between adjacent age groups for men and women. Non-linear regression analysis was applied in performance × age in each split and overall race time. Scheffe's post hoc was applied. Statistical significance was set at 1% ($\alpha < 0.01$). All procedures were made using Statistical Software for the Social Sciences (IBM, SPSS v20.0. Chicago, IL, USA) and GraphPad Prism (GraphPad Prism v6.0. San Francisco, CA, USA).

3. Results

An age effect was identified for both men and women for all three disciplines ($p < 0.0001$). The post hoc analysis showed all age groups to be statistically different from their previous one in all three disciplines and for both men and women (Figure 1a–c), i.e., the older the age, the slower the race time. Figure 1 presents performance decline with regard to age and shows age groups from 18–24 until 70–74 years. The three single-split discipline results are plotted in Figure 1a (swimming), Figure 1b (cycling), and Figure 1c (running). Figure 2 adds the respective delta between two adjacent age groups, and Figure 3 shows overall race time age group performance deterioration as well as overall delta change with age. Figure 1a presents swimming speed (km/h), which continuously decreases with age. Its smallest deterioration is between age groups 40–44 and 45–49 years for men (delta 0.76%) and between 45–49 and 50–54 years for women (delta 0.76%). Swimming performance analysis shows an effect for age group (F = 27.0; $p < 0.001$), sex (F = 190.7; $p < 0.001$), and interaction (F = 49.3; $p < 0.001$). Figure 1b shows cycling speed (km/h) for all age groups, which starts to deteriorate at 34 years in men and 35 years in women. Cycling performance analysis shows an effect for age group (F = 38.5; $p < 0.001$), sex (F = 2172.8; $p < 0.001$), and interaction (F = 22.8; $p < 0.001$). Figure 1c is about running speed (km/h). In men, performance decline starts at about 26 years, in women at 28 years. Running performance analysis shows an effect for age group (F = 97.7; $p < 0.001$), sex (F = 853.4; $p < 0.001$), and interaction (F = 20.8; $p < 0.001$).

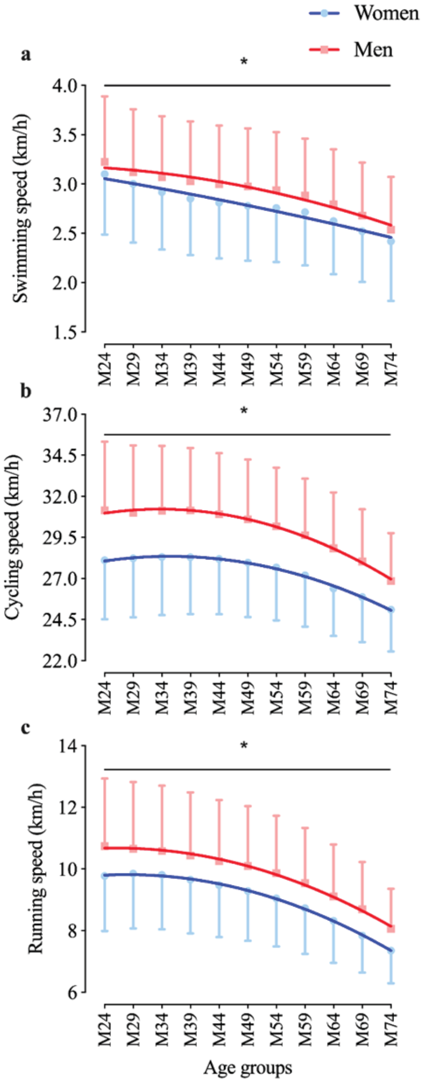

Figure 1. Age group-dependent decline in split discipline performance expressed in km/h; (**a**, top): swimming speed; (**b**, middle): cycling speed; and (**c**, bottom): running speed; (**a**–**c**): men in red, women in blue. * Age group, sex, and interaction effect ($p < 0.05$).

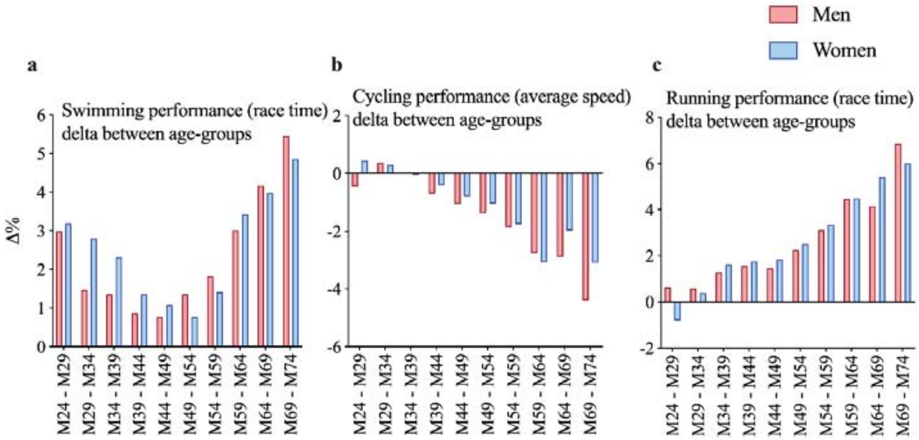

Figure 2. Split performance delta (%) between any two adjacent age groups; (**a**, on the left): swimming speed (km/h); (**b**, in the middle): cycling average speed (km/h); and (**c**, on the right): running speed (km/h); (**a**–**c**): men in red, women in blue.

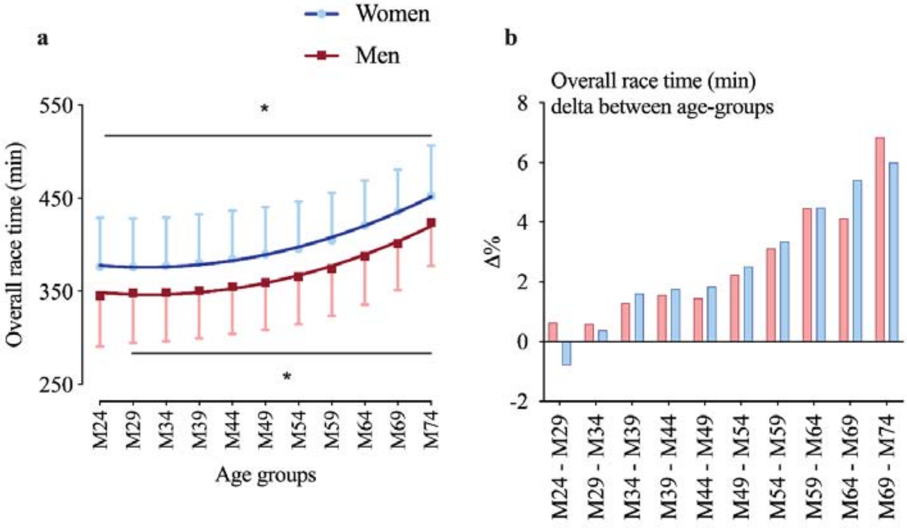

Figure 3. Overall race time; (**a**, on the left): overall race time for both sexes; (**b**, on the right): race time delta (%) between any two adjacent age groups; (**a**,**b**): men red, women blue. * Different from previous age group ($p < 0.05$).

Figure 2 describes how any two adjacent age groups differ in performance (i.e., performance delta). It is interesting to note here that the swimming delta (in Figure 2a) has its absolute minimum between 40–49 and 45–54 years in men and women, respectively. The constant positive delta (%) is higher in the older and the younger age groups (i.e., 55–74- and 18–39-year-old athletes being the highest between 65–69 and 70–74 years at 5.46% in men and 4.86% in women). Figure 2b on the cycling speed delta shows that the decline of all age groups increases except between 55 and 69 years in both sexes (especially in women). The running speed delta is (more or less) constantly deteriorating between age groups, as Figure 2c presents. Between the age groups 45–49 and 50–54 years, the delta rises significantly. From around 1.4% between 30 and 49 years, it increases to about 2.25% between 45 and 54 years and up to 6.85% between 65 and 74 years in men and from 1.74% over 2.52% until 6.01% in women. Both sexes deteriorate in a similar pattern.

Figure 3a on overall performance decline shows a constant deterioration in men and, from 25 to 34 years onward, in women as well (i.e., in women the age group 25–29 years is faster than the age group 18–24 (delta is −0.8%, see Figure 3b)). Overall performance analysis shows an effect for age group (F = 288.8; $p < 0.001$), sex (F = 3206; $p < 0.001$), and interaction (F = 6.7; $p < 0.001$). The age of peak performance for overall race time and each discipline by sex is presented in Table 1.

Table 1. Parameters in second-order polynomial regression $Y = a + bx + cx^2$ using the race time of all athletes in 1-year age intervals by sex.

Sex and Discipline	Parameter			R^2	Age (Years)	Race Time (min) or Speed (km/h)
	a	b	c			
Women						
Swimming	3.265	−0.00776	−0.00004	0.968	-	-
Cycling	25.59	0.1561	−0.00220	0.997	35.48	22.82
Running	8.909	0.06502	−0.00116	0.999	28.03	8.00
Overall	414.8	−2.513	0.04067	0.996	30.90	375.98
Men						
Swimming	3.169	−0.00348	−0.00015	0.964	-	-
Cycling	28.25	0.1765	−0.00262	0.996	33.68	25.28
Running	9.941	0.05702	−0.00109	0.996	26.16	9.20
Overall	386.9	−2.561	0.04063	0.990	31.52	346.54

Y = speed (km/h) in swimming, cycling, and running; Y = race time (min) in overall performance. For both approaches (i.e., 1 and 5 year) a second-degree polynomial analysis does not provide valid data as the youngest age group is the fastest one.

4. Discussion

This study attempted to identify how age groups of recreational master athletes perform in the Half Ironman triathlon. Hence, all athletes worldwide of all officially documented races available were considered. The main results were that (i) the swimming decline started from the youngest age group on, and that (ii) there was a sex difference in swimming and cycling, but not in running. Performance decline in multidiscipline sports depends on locomotion modes [9,11–13,24]. In accordance with Käch et al. [12], who investigated recreational Ironman triathletes, we expected that Half Ironman performance would follow these patterns: swimming decline starting first, followed by running and lastly cycling. Performance decline (at least in professionals) varies with race length and, thus, we expected it to be less prominent in Half Ironman than in Ironman triathletes [9,11].

4.1. Earlier Age-Related Decline in Swim Performance Compared to that in Cycling and Running

The first result was that the age-dependent decline in Half Ironman started earliest in swimming, where every age group was slower than its previous one. The smallest changes take place between the age groups 35–49 and 40–54 in men and women, respectively. Käch et al. also described such a constant decline for recreational Ironman triathletes for women but not for men, where the 25–29 age group was faster than the 18–24 age group (see Figure 1 in [12]). Besides this, there is almost always an inflection point in endurance sports performance decline (see Stones and Hartin [18]), as not only strength but also knowledge and experience matter (especially in ultra-endurance events), enabling older athletes to outperform younger ones. By contrast, in "elitist" groups, i.e., when investigating top 5 or top 10 athletes per groups, the swim decline seems to start at about 40–45 years of age, almost irrespective of race distance, namely in Olympic World Championships [10]; in Half Ironman (20 best athletes per age group in a qualifier race) [9,19]; and in "Ironman Hawaii" [10]. Swim age group decline, however, when studying larger groups, also started earlier than in the abovementioned elitist groups, namely from 30 to 34 years onward in 5549 athletes competing in Half Ironman World Championship races (between 2006 and 2010) [18] and, with an even stronger effect, when analyzing all races worldwide (i.e., constant deterioration from 18 to 24 years onward) in masters Ironmen [12]. The differences between elite and recreational athletes are obvious as their training differs concerning volume, intensity, and motivation.

4.2. Decline in Running Performance with Initial Plateau

In this study on the Half Ironman triathlon, running speed started to decrease at 26 years (men) and 28 years (women) (see Figure 1c and Table 1). Stones and Hartin [18] mention the Half Ironman age group decline to be progressive after the age of 30–34 years. Those differences might be because of the different numbers of athletes and race types investigated (i.e., World Championships and 5549 athletes by Stones and Hartin [18] compared to all races worldwide and 840,100 athletes in this study). A similar relation exists between the study by Lepers et al. [10] and Käch et al. [12]. Lepers stated a progressive running decline for World Champions after the age of 50 years in a study with a rather smaller number of athletes. By contrast, Käch et al. [12], who investigated recreational triathletes, reported a much earlier decline starting at about 30–34 years in women and 35–39 years in men. Elite athletes maintain performance up to the fourth or fifth decade of life, i.e., a curvilinear decline from 50 years onward in Olympic World Championships [10]; from 45 years onward for top athletes in a Half Ironman qualifier [9]; and from 45 years onward in the "Ironman Hawaii" [10]. Elite athletes are capable of maintaining their high performance over a longer period than recreational athletes because of their training being more intensive and high-volume oriented.

4.3. Cycling Performance Decline Is Least Pronounced Compared to Other Split Disciplines

Cycling age-dependent performance decline (at least at World Championship race level) is known to be less pronounced in shorter than in longer distances [10] with air resistance (and therefore cycle time as well) being the primary energy cost factor at high cycling speeds since mechanical power herein depends on the third power of speed [4,13]. Performance of recreational Ironman triathletes, who cycle 180 km per race, is known to start to deteriorate from 30 to 34 years onward in women and 35–39 in men [12]. In this study on races with a 90 km cycling split, the athletes' performance started to decline at 23 years for women and at 25 years for men (Table 1). The discrepancy with the results of Lepers et al. could stem from them investigating professionals, in whom the abovementioned physiologic regularities are effective. However, in recreational age group master athletes, the performance density of all athletes of a given age group could be too wide to see such regularities work.

4.4. Overall Decline Starts Earlier than in Ironman

According to Käch et al., the performance of recreational Ironman athletes started to deteriorate at 30–34 years in women and at 35–39 years in men [12]. In this study on non-elite master athletes, the significant decline started at the age of 31 and 32 years for women and men, respectively (see Figure 3; Table 1). The Ironman decline taking place later than that in the Half Ironman is what literature suggests [9,11].

Stones and Hartin [18] reported that for the oldest age groups in the Half Ironman, cycling and running contributed for the most part to overall performance; approximate relative times spent in each subdiscipline were 12% (swimming), 51% (cycling), and 37% (running). Comparing Käch's results [12] and ours, we have to state that the overall decline in Half Ironman triathlon starts earlier than that in Ironman.

4.5. Swim Performance Decline Does Significantly Differ between the Sexes

Surprisingly, sex difference was highest in swim performance decline. In endurance events, it is usually thought to be 10%–15%, mostly irrespective of exercise duration, widening with age [11], and it is usually thought to be smallest in swimming. Women have 7%–12% higher body fat than men [24] and there is less leg drag due to smaller body size, these factors, with other aspects, give women an advantage compared to men [19,24]. Others found the largest sex differences in short-distance triathlon swimming [11], where the abovementioned benefits should not pay off as well as in longer distances. Lepers stated a 12% sex difference in recreational triathletes [24]. Tanaka and Seals found a greater swim decline in women than in men [22], which goes along with our dataset, and Lepers and Maffiuletti [25] recorded that the sex difference in swimming narrowed with increasing race length—from about 19% in 50 m to approximately 11% in 1500 m events).

4.6. Cycling Performance Decline Does Significantly Differ between the Sexes

Age groups of recreational full-distance ironmen have a cycling sex difference of about 15% [24]. Most interestingly, men's performance decline in this study is more pronounced than women's (see Figure 2b), which we did not expect [12]. A potential explanation could be that a decline which starts at a higher level might deteriorate more pronouncedly compared to the one that starts on lower numbers [26].

4.7. Overall Performance Does Not Exhibit Significant Sex Differences

The age-related decline in overall race time seemed to be greater in women than in men, in the elderly than in the young [19], and usually less pronounced in shorter than in longer distances [15]. In Ironman, the overall sex difference narrowed from 15% (1990s) to 11% (2012) with women's running improvements being most responsible for that [24]. These findings line up with our results (see Figure 3b).

4.8. Potential Physiological Explanations for the Age-Related Decline in Performance

Many factors affect age-dependent declines. Individual medical condition (e.g., disease, trauma sequelae) as well as physiological factors (e.g., maximal oxygen consumption (VO_2max), lactate threshold and exercise economy [16]), which deteriorate while aging, may slow down individuals and therefore have group effects. VO_2max seems to be the most important physiological factor for women as well as men [12]. Peak and mean power output depend on lower-limb muscle mass [19], which in turn explains why women usually have greater decline rates than men since cycling and running account for about 90% of race time [18]. Women's best times may be predicted by many aspects, i.e., relative maximum oxygen uptake while swimming, overall speed in running at ventilatory threshold, and a few more, whereas men's only significant predictor of overall race time was running speed at ventilatory threshold [19].

Another perspective suggested by Lepers [15] differentiates between central physiology (e.g., maximum heart rate, maximum stroke volume, blood volume) and peripheral physiology (e.g., muscle mass, muscle fiber composition, etc.), which might be trained [15], with older women's exercise volume/intensity maybe being smaller than older men's [9]. Sociodemographic factors probably also account for the decline seen in this study [15]. The Half Ironman triathlon, being a young and emerging sport might have more less-trained masters athletes than Ironman events, as participation in an Ironman requires better preparation [4]. Decline deceleration in endurance events mostly depends on experience [12,18], learning, applying, and fine tuning race tactics over the years [4], as well as being aware of and able to deal with psychological factors that enhance or dampen motivation [12]. Better training knowledge (which is easily accessible via the internet) and opportunities (which are easy to partake in for both sexes in a sports-centered century) enable all recreational athletes to perform better than in previous years.

4.9. Characteristics, Strengths, and Weaknesses (Biases) of this Study

We tried to avoid an “elite bias” because only about 1% of participants are elitist in mass endurance events (a number taken from an Ironman study) [12]. The study therefore comprises a large dataset of 840,100 athletes (24.6% female and 75.4% male athletes) with its relative female participation being located within the lower normal range for endurance events [24]. The study’s design is cross-sectional, thus not allowing for longitudinal changes but considering age group changes. External conditions such as weather and racecourse characteristics have not been considered but should have exerted little influence due to the number of race events (which was 690). The graphs exhibit relatively high standard deviations, probably due to the high number of participants in Half Ironman events, widening performance variation.

4.10. Limitations, Practical Applications, and Implications for Future Research

Considering the large number of triathletes participating in various race distances of triathlon—including the Ironman 70.3—and the variation of the age of these athletes, the results of the present study would be of great practical value for practitioners (e.g., coaches and fitness trainers) working in this sport. The large variation of ages implies that practitioners might work with groups of athletes demanding the formulation of individualized age-related training goals. Such age-related training goals would not only concern the overall race time, but also the performance in each discipline. In addition, the majority of triathletes might be considered as non-elite athletes with characteristics differing from their elite counterparts; thus, trends in participation and performance of elite athletes would not be valid for non-elite athletes highlighting the importance of the practical relevance of our findings. In this context, practitioners might use the data of this study as a reference to evaluate the overall race time and performance by discipline and set long-term training goals considering the expected age-related decline in performance.

External conditions (e.g., weather, training background) and motivational aspects have not been included in this analysis. As the Half Ironman has only been under the official label since 2004, it is a young phenomenon. It would therefore be interesting to understand the athletes’ motivation better, especially, what makes them choose this distance and train hard while getting older. A deeper understanding of the training habits of recreational Half Ironman triathletes is needed in order to compare their decline patterns better (comparing age group athletes with same training volume and intensity). A continuation of the study with appropriate physiological indicators (of a subgroup of its over 840,100 athletes) may be of benefit and add to our understanding of performance decline.

With regard to the magnitude of the relationship of race and split times with age, R^2 was close to 1 denoting an almost perfect relationship. This observation might be partially attributed to the application of a second-degree polynomial regression analysis that seemed fitted with age-related decline in performance, i.e., relatively small differences among young age groups, whereas larger differences were shown among old age groups. This notion is in agreement with the theoretical knowledge suggesting a deeper decline in physiological characteristics related to performance after a certain age [4,8–11]. In addition, the R^2 value might also be so high due to the use of a very large sample size that “masked” the influence of any outliers.

5. Conclusions

Performance deterioration in Half Ironman triathlete age groups as shown in this study begins at the first age group with swimming, at 28 (women) and 26 (men) years for running, at 34 (men) and 35 (women) years in cycling (as expected based on Käch et al. [12] for Ironman triathlon), and at 31 (women) and 32 (men) years of age concerning overall race time. Among these numbers, cycling does not seem to play as big a role as one might think because of its short relative duration during a Half Ironman race. Half Ironman athletes spend about 12% of time in swimming, 51% in cycling, and 37% in running [18]. In order to achieve best results, we conclude that every athlete should invest in swim training as early as possible to minimize any possible weaknesses that may inhibit best times. Cycling seems to be least prone to deterioration but still needs lots of training as it constitutes the biggest part of overall time (in order to reduce fatigue during and preserve a high pace for the run split). The strongest training emphasis though should be deployed to running, the second longest discipline, in which the biggest amount of fatigue has to be dealt with. In Ironman, cycling, running, and overall race time started to deteriorate at 30–34 years in women and at 35–39 years in men. The performance in swimming started to deteriorate at 25–29 years in both sexes [12]. Hence, it seems logical that women above 31 and men above 32 years should consider changing to Ironman in order to be able to achieve or preserve best results. In summary, this study's results go along with literature showing that swim performance in recreational Half Ironman athletes seems to be hardest to preserve; that run performance decline is a constant one; and that cycling performance can be preserved up to older age groups.

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Tower Running—Participation, Performance Trends, and Sex Difference

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Abstract: Though there are exhaustive data about participation, performance trends, and sex differences in performance in different running disciplines and races, no study has analyzed these trends in stair climbing and tower running. The aim of the present study was therefore to investigate these trends in tower running. The data, consisting of 28,203 observations from 24,007 climbers between 2014 and 2019, were analyzed. The effects of sex and age, together with the tower characteristics (i.e., stairs and floors), were examined through a multivariable statistical model with random effects on intercept, at climber's level, accounting for repeated measurements. Men were faster than women in each age group ($p < 0.001$ for ages ≤69 years, $p = 0.003$ for ages > 69 years), and the difference in performance stayed around 0.20 km/h, with a minimum of 0.17 at the oldest age. However, women were able to outperform men in specific situations: (i) in smaller buildings (<600 stairs), for ages between 30 and 59 years and >69 years; (ii) in higher buildings (>2200 stairs), for age groups <20 years and 60–69 years; and (iii) in buildings with 1600–2200 stairs, for ages >69 years. In summary, men were faster than women in this specific running discipline; however, women were able to outperform men in very specific situations (i.e., specific age groups and specific numbers of stairs).

Keywords: tower running; sex differences; age; running speed; vertical run

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1. Introduction

Distance running is of high popularity and includes different distances, from 5 to 10 km [1], half-marathon [2,3], marathon [2,4], and up to ultra-marathon of different distances [5,6]. It is well-known that men are faster than women from 5 km to marathon [7], and in ultra-marathon running [8]. However, women were able to reduce the gap with men in ultra-marathon running, with increasing age and at longer race distances [9].

Stair climbing or tower running is a very specific running discipline, in which stair climbing has developed into the organized sport of tower running. Nowadays, tower running is a sport discipline that involves running up tall buildings, such as internal staircases of skyscrapers. However, tower running can cover any running race that involves a course that ascends a building.

To date, we have knowledge about the health benefits of stair climbing [10–13]. However, no data exist about participation and performance trends in tower running, and especially about the sex difference in this specific running discipline. Such information is valuable for athletes and coaches, to better understand and plan a race strategy, and also for race organizers, for insights regarding future events.

Therefore, the aim of the present study was to investigate participation and performance trends in tower running, with the hypothesis that men would also be faster than women in this discipline. Regarding age groups, we expected that women might close the performance gap in the older groups as already shown in long distance races [9].

2. Materials and Methods

2.1. Ethics Approval

This study was approved by the Institutional Review Board of Kanton St. Gallen, Switzerland, with a waiver of the requirement for informed consent of the participants, as the study involved the analysis of publicly available data.

2.2. Methodology

There exists a tower running world association that presents all the results of the known races around the world on their homepage (www.towerrunning.com). In an older version of this homepage, there were only the results of the current year, and sometimes, of the preceding year. We contacted the person in charge at the association to find out whether he could provide us with older data as well. For some races, however, it was not possible to find the older results. For example, for the race at the Willis tower in Chicago, the results before 2018 were not available. For other races, such as the hustle up race in Chicago, the direct link did not work, but the results could be found by searching for the link to the race, which is also provided on the homepage of the tower running world association. Table 1 summarizes all considered events listed by the number of steps of the buildings.

Table 1. Data included in the present study.

Building	City	Steps	Data Available (Years)	Included (Years)
Millennium Tower	Wien	2529	2014–2016	2014–2016
Willis Tower (Sears Tower until 2009)	Chicago	2109	2014–2019	2018
Taipei 101	Taipeh	2046	2014–2019	2017–2018
CN Tower	Toronto	1776	2014–2019	2017–2018
Reunion Tower	Dallas	1674	2018–2019	2018
Eiffelturm	Paris	1665	2015–2020	2015–2018
AON Center	Chicago	1643	none on towerrunning.com	2018
John Hancock Center (875 North Michigan Avenue)	Chicago	1632	2014–2019	2017–2018
Empire State Building	New York	1576	2014–2019	2017–2014
Bank of America Plaza	Dallas	1540	none on towerrunning.com	2018
US Bank Tower	Los Angeles	1500	2014–2019	2018
thyssenkrupp Testturm	Rottweil	1390	2018–2019	2018
Swissôtel The Stamford	Singapur	1336	2014–2018	2017
Rockefeller Center	New York City	1214	2014–2016, 2018, 2019	2019
MesseTurm	Frankfurt am Main	1202	2014–2019	2014–2017
Three Logan Square	Philadelphia	1088	2014–2019	2014, 2018, 2019
Valliance Bank	Oklahoma City	837	2014–2019	2019
Holmenkollbakken	Oslo	800	2015–2018	2015–2017
Run Up Berlin (Park Inn Hotel)	Berlin	770	2015–2019	2015–2018
KölnTurm	Köln	714	none on towerrunning.com	2016–2019
Oakbrook Terrace Tower	Oakbrook	680	2014–2020	2019
Münsterturm	Ulm	560	none on towerrunning.com	2014–2018
Towerrun	Berlin	465	2014–2020	2018
St.George's Tower	Leicester	351	none on towerrunning.com	2018
Matzleinsdorfer Hochhaus	Wien	342	2017	2017
Windradlauf	Lichtenegg	300	2014	2014
Haus des Meeres	Wien	271	2015–2019	2016–2018
Oluempia Hotel	Tallinn	N/A	2015–2019	2017

From the race results, the year of the event, the completed time, the sex, and the name of both the athletes and the building were available. We further looked for the height of the building and the number of stairs and floors. Race time in m:sec was converted to running speed in km/h, using the height of the building. We removed observations from unknown climbers (where the name of the climber was not reported or not known) in order to correctly account for repeated measurements. We also considered multi-climbing.

2.3. Statistical Analysis

The outcome was the tower climbing speed (km/h). Descriptive statistics are presented as means (SD = standard deviations) by sex and age groups. T-tests were performed to assess the outcome difference between sex, overall and for each age groups. Two-way ANOVA tests were also performed to evaluate the multivariable effect of sex and age on the outcome. Then, to control also for repeated measurements and the other covariates, the effects of sex and age, together with the tower characteristics (i.e., stairs and floors) were examined more rigorously through a multivariable mixed effects model, with random effects (intercept) for climbers. The model was specified as follows:

$$\text{Tower climbing speed (Y)} \sim [\text{Fixed effects (X)} = \text{Sex*Age*BS (Stairs, df} = 5) \\ + \text{BS (Floors, df} = 5) + [\text{random effects of intercept} = \text{runners}]$$

where BS (Stairs, df = 5) and BS (Floors, df = 5) are 5 degrees of freedom (df) basis splines changing with the number of stairs and floors, respectively; Sex*Age*BS (Stairs, df = 5) denoted the three-way interaction term Sex–Age–number of stairs. Calendar year was not considered in the above model because it was not significant.

Results of the regression model are presented as estimates and standard errors. Statistical significance was defined as $p < 0.05$. All statistical analyses were carried out with R, R Core Team (2016). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria (www.r-project.org/foundation/). The R packages ggplot2, lme4, and lmerTest were used, respectively, for data visualization and for the mixed model. The R code to reproduce the analysis is provided as supplementary information (Supplement 1 R-code).

3. Results

Between 2014 and 2019, the total number of observations was 28,203 (24,007 climbers). However, the total number of observations, with non-missing sex, was 28,156 (23,960 climbers). The participation and men-to-women ratio is shown in Figure 1. We observed that we had a low number of participants and a high men-to-women ratio before 2017 (i.e., the number of men was three times the number of women in 2015). The highest number of participants was recorded in 2018. In fact, the number of women in 2018 was eight times the number of women in 2014, and the number of men in 2018 was four times the number of men in 2014. In 2019, the number of available observations decreased again. The men-to-women ratio reached a minimum in 2019 with 0.89, meaning that the number of women was higher than the number of men.

In Table 2, the mean performance by sex and age group is reported. Men were faster than women in each age group ($p < 0.001$ for all ages until 69 years, $p = 0.003$ for ages >69 years), and the difference in performance stayed around 0.20 km/h, with a minimum of 0.17 at the oldest age. In Table 3, summary statistics of performance, together with tower characteristics: height, number of floors and stairs are reported by sex. Overall, the sex difference in performance was significant ($p < 0.001$); sex differences were also significant ($p < 0.001$) in average floors and stairs climbed. The results of the multivariable statistical analysis are displayed in Figure 2, to allow an easier interpretation and understanding. Moreover, we had no significant difference between men and women alone, but in the interaction with age groups and stairs climbed (Supplemental 2 Table). The variability, in terms of performance, was greater in very young and very old age groups (<20 years, 60–69 years, and >69 years). This also had an effect on sex differences. Women performed better than men in the following situations: (i) smaller buildings (<600 stairs), for ages between 30 and 59 years and >69 years; (ii) higher buildings

(>2200 stairs), for age <20 years and ages between 60 and 69 years; and (iii) buildings with 1600 to 2200 stairs, for age >69 years. In all other cases, men performed better than women, with the sex difference reducing when the number of stairs increased. In Figure 3, the effect of the number of floors on performance, by sex, is shown. When the number of floors increased, the average speed of tower climbing decreased, but then increased around 90 floors, and decreased again in climbing the highest buildings.

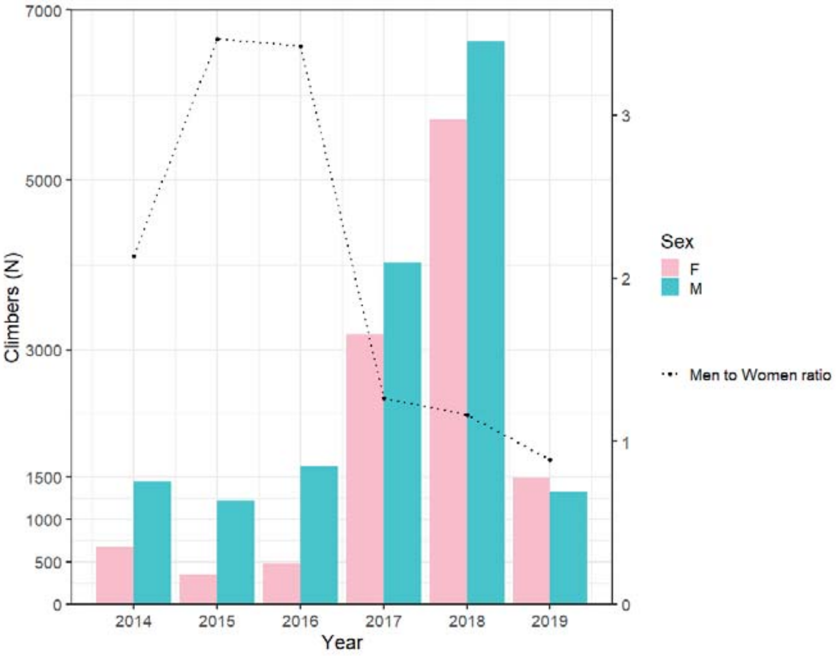

Figure 1. Participation and men-to-women ratio.

Table 2. Summary statistics of tower climbing performance, running speed (km/h), by sex and age groups. *p*-values from t-tests for each subgroup are reported. *p*-values from ANOVA were both $p < 0.001$ for sex and age. Men-to-women ratio, computed with the number of participants, is reported.

Age Group	Sex	N	Mean (SD)	*p*	Men-to-Women Ratio
<20	F	501	0.73 (0.27)	<0.001	1.30
	M	652	0.91 (0.38)		
20–29	F	1887	0.81 (0.24)	<0.001	1.39
	M	2615	0.99 (0.35)		
30–39	F	2552	0.80 (0.30)	<0.001	1.34
	M	3415	1.03 (0.39)		
40–49	F	1941	0.78 (0.32)	<0.001	1.33
	M	2583	1.00 (0.39)		
50–59	F	1220	0.76 (0.30)	<0.001	1.60
	M	1951	0.97 (0.38)		
60–69	F	239	0.72 (0.25)	<0.001	2.62
	M	626	0.90 (0.27)		
>69	F	44	0.66 (0.33)	0.003	4.57
	M	201	0.83 (0.33)		

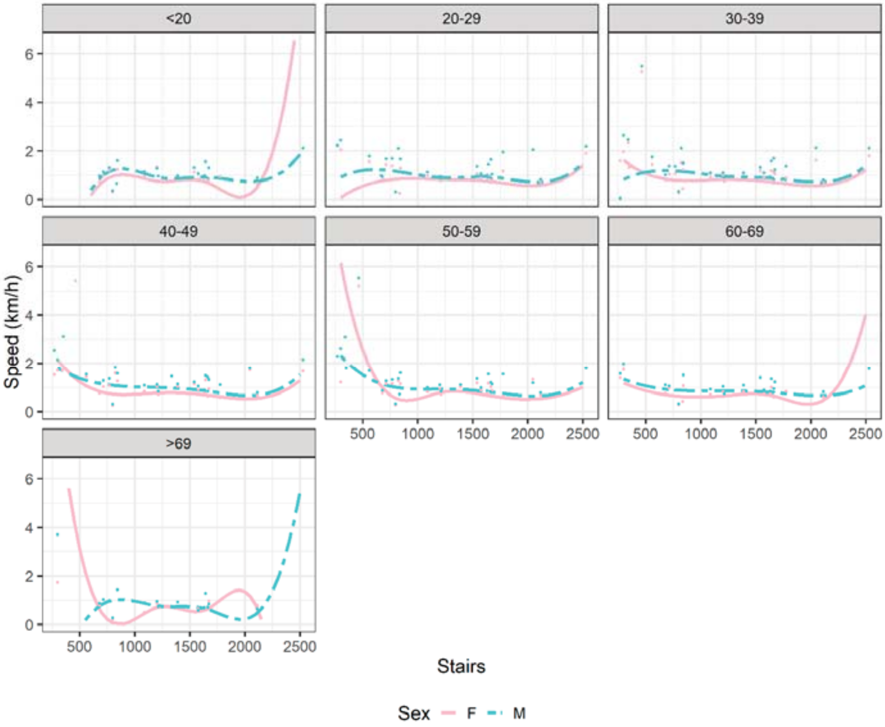

Figure 2. Speed (km/h) by stairs, age, and sex. Lines represent the predicted values from the mixed model and points represent the average of the observed values.

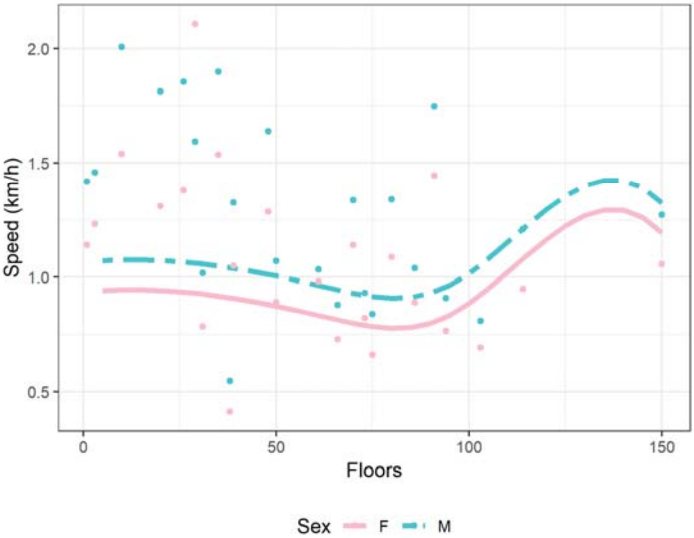

Figure 3. Speed (km/h) by floors and sex. Lines represent the predicted values from the mixed model and points represent the average of the observed values.

Table 3. Summary statistics of running speed (km/h) and race time (min), tower height (m), floors, and stairs by sex. Data expressed as mean (± SD).

	Females (n = 11,886)	Males (n = 16,270)	*p*-Value
Speed km/h	0.85 (0.37)	1.06 (0.46)	<0.001
Time (min)	24.26 (14.16)	18.43 (11.69)	<0.001
Tower height (m)	296.25 (111.37)	276.27 (108.72)	<0.001
Floors	85.44 (36.37)	76.00 (35.97)	<0.001
Stairs	1466.43 (420.36)	1401.18 (429.59)	<0.001

4. Discussion

The aim of the present study was to investigate participation trends, performance trends, and trends in sex difference in tower running, with the hypothesis that men would be faster than women in this discipline. The main findings were: (*1*) more men than women competed before 2017, (*2*) men were faster than women in each age group and the difference in performance stayed around 0.20 km/h, with a minimum of 0.17 km/h at the oldest age, and (*3*) women aged between 30 and 59 years and >69 years performed better than men in smaller buildings (<600 stairs).

4.1. Change in the Men-to-Women Ratio Across Years

Before 2017, we observed a low number of participants and a high men-to-women ratio. The highest number of participants was recorded in 2018. In 2019, the number of participants decreased again and the men-to-women ratio reached the minimum of 0.89, which means that the number of women was higher than the number of men. This could also be due to a selection bias. At the time of the data collection (2017–2019), there were more results available from the earlier races and since the aim of the selection was to represent the sport and include the most important races all over the world, we did not pay attention to compare for every year the exact same number of races. This fact should encourage race directors to join the 'Towerrunning World Association' (www.towerrunning.com), in order to build up a firm data base for future analyses.

Generally, in races of long traditions, the men-to-women ratio is > 1.0, indicating that more men than women competed [14], but the men-to-women ratio can decrease over the years, indicating that the number of women increased over time [15]. Future studies with larger data sets are needed to investigate this trend.

4.2. Sex Difference in Performance

Looking at the anatomical aspect of sex difference, studies have shown that there are differences in the anatomy and physiology of the heart [16], and in the oxygen uptake in repetitive muscle activity [17] between men and women. This fact suggests that there must also be differences in performance between genders in the sport of tower running.

Men were faster than women in each age group and the difference in performance stayed around 0.20 km/h, with a minimum of 0.17 at the oldest age. However, women outperformed men in the following situations: (i) smaller buildings (<600 stairs) and ages between 30 and 59 years and >69 years; (ii) higher buildings (>2200 stairs) and ages <20 years and between 60 and 69 years; and (iii) buildings with 1600–2200 stairs and ages >69 years.

When the number of floors increased, the average running speed of tower climbing decreased, but then increased around 90 floors, and decreased again in climbing of highest buildings. A possible explanation for this fact could be the diversity of the runners. One could think that recreational runners take part in races until a certain height, because of their estimated stamina. Therefore, their running speed decreases until they reach their maximum of the height of the building. More professional runners again might only start in the races in which they have to climb the higher buildings, starting around 90 floors. Again, these professional runners will have to decrease their average running speed, to be able to climb even the highest building. This, on the other hand, is only a hypothesis that we did not investigate, and would need further studies to be verified.

Another explanation could be the men-to-women ratio by age group. When female and male age group ultra-marathoners were investigated, women could close the gap to men in older age groups (>60 years) and longer race distances (i.e., 100 miles compared with 50 miles) [9]. This relative improvement in female performance at higher ages is most likely due to the change in the men-to-women ratio in older age groups. It has been shown for female and male age group freestyle swimmers, from 25–29 to 85–89 years, competing in the FINA World Masters Championships between 1986 and 2014, that women were faster than men for age groups 80–84 and 85–89 years. When the trend for the men-to-women ratio for age groups 25–29 to 75–79 years (i.e., men were faster than women) and age groups 80–84 to 85–89 years (i.e., women were faster than men) was analyzed, the men-to-women ratio remained unchanged in 50 m, 100 m, and 400 m in age groups 25–29 to 75–79 years, but increased in 200 m and 800 m. For age groups 80–84 to 85–89 years, the men-to-women ratio remained unchanged in 50 m and 100 m, but decreased in 200 to 800 m [18]. However, in the present tower runners, the men-to-women ratio increased with increasing age, but was lowest in the youngest age group (Table 2).

Other variables could explain that women outperformed men in some specific situations (e.g., specific age groups and building heights) of this running discipline. Generally, women are lighter than men [19–21], which might help in running upwards. Body mass was, however, not predictive in female mountain ultra-marathoners [21]. Unfortunately, body mass was not available in these runners. Another explanation could be the motivation of female athletes [22]. For example, motivation differs between female and male marathon runners [22]. It has been shown that female marathon finishers exceeded men on the motivational scales for body weight concern, affiliation, psychological coping, life meaning, and self-esteem, and they scored lower on competitive motivation [23]. Future studies might investigate the motivation of female and male tower runners by age group and performance level.

Regarding the health aspect, it has already been investigated that stair climbing brings certain benefits. It could be shown that it helps decrease blood glucose levels [12] and that it brings a cardiac benefit in senior citizens [13]. Therefore, there is a certain interest in investigating this subject regarding public health.

5. Conclusions

Men are generally faster than women in tower running, but women are closing the gap with men, with increasing stairs and increasing age. The reason for the better performance in women with increasing stairs remains unclear and might be a subject for further research.

Supplementary Materials: The following are available online at http://www.mdpi.com/1660-4601/17/6/1902/s1, Table S1: R-Code, Table S2: Regression analysis (mixed model) of speed (km/h) in tower climbing.

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Article

Psychobiological Changes during National Futsal Team Training Camps and Their Relationship with Training Load

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Abstract: The aim of this study was two-fold: (1) to analyze the within-week variations of heart rate, session-rated of perceived exertion (sRPE), total distance, distance in 8.0–11.99 km/h^{-1}, recovery distance in 12.0–17.99 km/h^{-1}, distance in >18.0 km/h^{-1}, maximum speed, number of sprints, heart rate variability, delayed onset muscle soreness (DOMS), and fatigue during training camps of a national futsal team; and (2) to analyze the relationships between load and the well-being. Twenty-eight men from the Chinese Taipei U−20 national futsal team were analyzed. Comparisons of training days revealed that the total distance was significantly smaller on day 1 (d = −1.22) and day 6 (d = −1.95) than on day 3. The sRPE values were significantly lower on day 1 than days 4 (d = −1.53), 5 (d = −2.07), and 6 (d = −2.59). The relationships between training load and recovery parameters revealed moderate correlations between the DOMS and the sRPE recorded one ($r = -0.321$) and two days before training ($r = -0.289$). It is possible conclude that first day imposed a smaller external load and internal load, and that the internal load had a greater dependent relationship with reported DOMS and fatigue during the training camps.

Keywords: youth; performance; workload; physiological phenomena; medical psychology; heart rate

1. Introduction

Congested periods of training (e.g., training camps) are often used by national youth teams to employ new dynamics among teammates and also to find or select new players for the final squad [1,2]. Training camps consist of a period (usually one week or more) in which players may be under evaluation by national coaches executing one or more training session per day. Usually, sessions in training camps are field based and mostly tactical/technical related. Small-sided games, positioning games, and regular games are the most common activities in those training camps. Based on the fact that these periods (training camps) are exclusively dedicated to improving the overall team's dynamics and selecting the players for the final squad, it is expected that a high level of training load will be promoted, mainly considering that in some of the days there may occur two training sessions [3]. This congested calendar may promote changes in the stimulus–recovery dynamics, and, for that reason, it is important to employ a player monitoring cycle that can help coaches and sports scientists properly manage the load and readiness of players [4].

Commonly, players' monitoring cycles are focused on monitoring external and internal loads, the wellness status of players, and readiness [4]. The external load represents the physical and neuromuscular demands imposed on the players by the tasks and drills, while the internal load represents the biological impact of the external load [5]. Typically, the external load is assessed by global positioning systems (GPSs) or inertial measurement units [6]. The internal load can be assessed both by objective (e.g., heart rate (HR) monitors, blood lactate concentrations) and subjective (e.g., rate of perceived exertion (RPE), effort scales) instruments [7]. However, sports scientists should not only consider the dynamics of load. The possible impact of load on the wellness status of players (and recovery, naturally) should also be considered in a well-implemented player monitoring cycle [8,9].

One of the most common measures used to control the wellness and recovery status in players is HR variability (HRV) [10]. This measure characterizes the parasympathetic and sympathetic influences of the autonomic nervous system on the sinus node by recording the beat-to-beat HR intervals [10]. The HRV can be classified as an objective measure; however, other subjective wellness measures have been proposed and applied in the context of team sports monitoring [11,12]. Among other topics, delayed onset muscle soreness (DOMS), stress, fatigue, sleep quality, and mood are normally assessed in players using questionnaires or scales [13]. Both objective and subjective measures of wellness can provide useful information about the daily variations of players, representing a possible interaction with the stimulus imposed by the training [14].

The research on training load and wellness is progressively growing, mainly in football [15]. Research has also been conducted in futsal [16,17]; however, the evidence is scanty, and some important external load measures have been not reported. In a study that described the perceived training load of futsal players over forty-five weeks, Rabelo et al. [18] showed that the players' training loads were typically lower than intended by the coach. Also, using perceptive scales of exertion, Clemente et al. [19] compared two types of training weeks (normal versus congested), revealing that perceptive training load, DOMS, and fatigue were significantly greater in normal weeks and that within-week changes occurred in terms of perceptive load.

In an attempt to identify relationships between daily perceived recovery and circumstantial factors related to training load in futsal, sleep quality, or period of the week, Wilke et al. [20] found that neither recovery classification nor prior training load influenced perceived recovery. Even though the aforementioned study focused on the training load and wellness status of futsal players, his approaches require further study. Among potential research topics is how a training camp can influence the training load and wellness of players and what daily changes can occur during these camps. Another is how training load is related to future wellness variations one day and two days after a training session. Finally, it is important to add objective load measures that consider the physical demands of players instead of only the internal load. Such approaches will provide a better understanding of how both dimensions of load (internal and external) can fluctuate throughout a training camp and how external load may concur to influence the wellness fluctuations.

For these reasons, the purpose of this study was twofold: (i) to analyze the daily variations in training load and psychobiological measures during seven-day training camps attended by national futsal teams and (ii) to test the relationships between training load and psychobiological measures during the training camps.

2. Materials and Methods

2.1. Experimental Approach

This study followed a descriptive research design and a correlational design in which training load measures (both internal and external) and well-being measures were monitored daily in 7 day training camps and tested for its relationships. Players belonging to an under−20 (U−20) national futsal team were monitored daily during six domestic training camps (Table 1). The monitoring process included an analysis of the training load imposed during the sessions (both external and internal) and recovery processes (HRV, DOMS, and fatigue). The weekly protocol consisted in the day 1 of data collection of height, body mass, and skinfolds collected by an expert sports scientist using a calibrated skin folder (Lange Skinfold Caliper, Beta Technology, USA).

Table 1. Description of the number of sessions and time of sessions per day during the included training camps.

Period		*n* of Players	D1		D2		D3		D4		D5		D6		D7	
			Min	Ns	Min	Ns	Min	Ns	Min	Ns	Min	Ns	Min	Ns	Min	Ns
TC_1	July 9th to 15th 2018	21	116	1	116	1	115	1	121	1	225	2	131	1	113	1
TC_2	September 1st to 7th 2018	23	-	1	238	2	260	2	131	1	250	2	277	2	140	1
TC_3	September 21st to 27th 2018	17	-	1	243	2	270	2	131	1	254	2	217	2	98	1
TC_4	October 5th to 11th 2018	19	72	1	167	2	199	2	217	2	258	3	110	1	123	1
TC_5	October 18th to 24th 2018	16	110	1	213	2	197	2	253	2	109	1	228	2	83	1
TC_6	November 5th to 11th 2018	17	136	1	218	2	242	2	98	1	239	2	216	2	121	1

Min: minutes of training in that day; Ns: number of sessions in that day; TC1: first training camp; TC2: second training camp; C3: third training camp; TC4: fourth training camp; TC5: fifth training camp; TC6: sixth training camp.

Additionally, on each day of the training camp, HRV was measured in the morning, and subjective scores of DOMS and fatigue were collected individually. During the training sessions, players used Polar Team Pro sensors to measure external and internal loads. After each session, the RPE was also collected as an internal load indicator. Comparisons of internal and external loads, HRV, DOMS, and fatigue between training days were carried out. Moreover, correlations between training loads and the HRV, DOMS, and fatigue of the next two days were also tested. All training sessions were conducted in a sports complex at Wufeng University, Taiwan. All players stayed in the campus dormitory during the training camps.

2.2. Participants

Twenty-eight male futsal players that were recruited into the Chinese Taipei U−20 national team's training camps participated in this study voluntarily. Twenty-two field players and six goalkeepers were part of the sample (mean ± standard deviation: age = 18.07 ± 0.73 y; height = 169.57 ± 8.40 cm; body mass = 64.51 ± 12.19 kg). Eleven players were called up from university futsal teams, while seventeen players were called up from senior high school futsal teams. The recruited players undertook regular futsal training sessions (around 6–10 sessions per week) in home teams. The criteria for inclusion were as follows: (i) participants had to take part in all training sessions within the training camp; (ii) participants did not report injuries or illness during the training camp.

The participants were informed about the study design and were familiarized with the protocol. After that, all participants voluntarily signed an informed consent form. The study followed the ethical standards of the Declaration of Helsinki regarding the study of humans, and the study design was approved by the committee of the University of Taipei.

2.3. Heart Rate Variability (HRV)

The heart rate variability (HRV) was assessed every morning using an HR monitor (Polar Team Pro©, Polar Electro, Kemple, Finland). The HRV was assessed in a sitting position in the morning, prior to breakfast. Participants sat on chairs in a comfortable position for five minutes which was followed by five minutes of data collection. The data from the HR monitors were exported to the Polar Team Pro web platform, and the raw data was then used for treatment in Kubios HRV analysis software (Premium version 3.2, Kubios, Kuopio, Finland). Medium artefact correction and smoothing priors set at 500 lambda were used for HRV analysis. The natural log of root mean square of successive RR interval differences (LnRMSSD) was calculated in Microsoft Excel 2016 (Microsoft Corporation, Redmond, WA, USA) and was used to evaluate daily cardiac-autonomic functions during the training camps.

2.4. Subjective DOMS and FATIGUE

A 5 point Hooper questionnaire [11] was administered every morning (between 08:00 and 08:30). On the 5 point scale, 1 (lowest score) corresponded to the worst state and 5 (highest score) to the best state. The DOMS and fatigue were used for the present data treatment and study. The scores were provided individually, and a visual analogue scale was used to improve the overall accuracy of the answers.

2.5. Internal Load

Each player used an HR monitor (Polar Team Pro, Polar Electro, Kemple, Finland) during training sessions to determine the maximal heart rate achieved (HRmax) and the average heart rate (HRav) during the full session. The Borg CR−10 scale was applied as a subjective instrument thirty minutes after the end of the session [21]. The players were asked to answer the question, "How intense was the session?" using a visual analogue scale in which 0 represented "not at all" and 10 represented "extremely intense." The scores were provided individually and were then registered and multiplied by the duration of the session (in minutes) to calculate the session-RPE (sRPE) which represents the overall load of the session in terms of arbitrary units [22]. Players were previously familiarized with the scale in order to increase the accuracy of the answers.

2.6. External Load

The external load during the training sessions was monitored by Polar Team Pro sensors (Polar Team Pro, Polar Electro, Kemple, Finland). The sensor is a microsensor system that encompasses a 3 dimension accelerometer, a gyroscope, and a digital compass with a sample rate of 200 Hz. The maximal distance for recording the signal was 200 meters. The activities profiles included total distance, distance in zone 1 (8.0–11.99 km/h^{-1}), recovery distance in zone 2 (12.0–17.99 km/h^{-1}), distance in zone 3 (>18.0 km/h^{-1}), maximum speed (maxSpeed), and number of sprints (>24.0 km/h^{-1}) [3]. The same unit of measurement (GPS) was used by each player to avoid variability.

2.7. Statistical Procedures

Data were preliminary tested for normality ($p > 0.05$) and homogeneity ($p > 0.05$) in the SPSS software (version 23.0, IBM, USA). Thereafter, mean and standard deviation were calculated. Training load measures, HRV, and wellness parameters were compared between days of training (D1, D2, D3, D4, D5, D6, D7) using the standardized effect size of Cohen (d) in the form of pairwise comparisons. The magnitudes of the changes were interpreted based on the following thresholds [23]: 0.0–0.2, trivial; 0.2–0.6, small; 0.6–1.2, moderate, 1.2–2.0, large; >2.0, very large. The calculation of standardized effect size was made on Excel spreadsheets properly designed for the process [24]. Correlations between training load measures, HRV, and wellness parameters were tested using the Pearson's r coefficient for a confidence interval of 95%. The magnitudes of the correlations were defined as in Reference [23]:

0.0–0.1, trivial; 0.1–0.3, small; 0.3–0.5, moderate; 0.5–0.7, large; 0.7–0.9, very large; and 0.9–1.0, nearly perfect. The correlations were executed in the SPSS software (version 23.0, IBM, USA).

3. Results

Descriptive statistics of training load parameters during the seven days of training camps can be found in Table 2. The greater total distance (10111.0 ± 4878.7 m), distance covered between 12.0 and 17.99 km/h^{-1} (2131.7 ± 1720.3 m), distance covered above 18.0 km/h^{-1} (1056.6 ± 743.8 m), and number of sprints (27.4 ± 22.2 *n*) were covered at day 5 of the training camps. Considering the sRPE, the greatest load occurred at day 2 (1105.7 ± 363.3 A.U.) and the greatest HRmax at day 4 (187.2 ± 13.1 bpm^{-1}).

Table 2. Descriptive statistics of training load parameters during the different days of training camps.

	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Total distance (m)	6830.4 ± 2142.7	9257.7 ± 4075	9400.8 ± 3980.1	7964.4 ± 3366.6	10111.0 ± 4878.7	9170.7 ± 4585.3	6191.8 ± 1715.0
D8.0–11.99 km/h^{-1} (m)	1304.6 ± 335.7	1617.4 ± 819.0	1840.0 ± 1197.4	1325.2 ± 619.4	1742.3 ± 1263.4	1566.3 ± 882.7	1015.6 ± 330.1
D12.0–17.99 km/h^{-1} (m)	1250.1 ± 405.8	1851.5 ± 967.5	2080.3 ± 1336.3	1623.0 ± 848.9	2131.7 ± 1720.3	1846.2 ± 1020.6	1351.08 ± 499.3
D > 18.0 km/h^{-1} (m)	582.9 ± 304.2	1000.5 ± 690.3	999.0 ± 709.4	829.7 ± 665.3	1056.6 ± 743.8	899.9 ± 574.1	655.5 ± 394.3
MaxSpeed (km/h^{-1})	28.0 ± 3.3	27.2 ± 2.5	27.2 ± 4.7	27.6 ± 3.3	27.8 ± 3.2	28.1 ± 2.6	28.1 ± 3.3
Sprints (*n*)	14.3 ± 12.2	25.7 ± 21.8	25.1 ± 21.7	21.2 ± 21.9	27.4 ± 22.2	22.8 ± 18.7	14.4 ± 12.4
sRPE (A.U.)	674.5 ± 251.0	1105.7 ± 363.3	1153.3 ± 433.3	772.5 ± 441.4	998.6 ± 491.6	838.4 ± 393.5	609.0 ± 187.8
HRmax (bpm^{-1})	187.3 ± 10.8	185.0 ± 12.2	185.0 ± 14.5	187.2 ± 13.1	183.1 ± 10.6	184.9 ± 11.9	184.9 ± 12.2
HRav (bpm^{-1})	139.8 ± 10.5	134.2 12.9	130.7 ± 13.8	132.5 13.3	127.9 12.4	131.0 11.0	130.0 ± 11.8

D: distance; A.U.: arbitrary units; bpm^{-1}: beats per minute; *n*: number; m: meters; km/h^{-1}: kilometers per hour; HRmax: maximal heart rate; HRav: average heart rate; MaxSpeed: maximal speed; *n*: number; sprints: >24.0 km/h^{-1}.

Descriptive statistics of psychobiological parameters related with recovery and wellness can be found in Table 3. The highest InRMSSD (4.2 ± 0.5 log), DOMS (2.9± 0.9 A.U.), and fatigue (3.1± 0.9 A.U.) were found at the first day of the training camps.

Table 3. Descriptive statistics of psychobiological parameters during the different days of training camps.

	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
InRMSSD (log)	4.2 ± 0.5	3.9 ± 0.5	4.0 ± 0.5	3.8 ± 0.6	3.9 ± 0.6	3.9 ± 0.5	3.9 ± 0.5
DOMS (A.U.)	2.9 ± 0.9	2.8 ± 0.7	2.6 ± 0.8	2.5 ± 0.8	2.6 ± 0.8	2.6 ± 0.7	2.6 ± 0.7
Fatigue (A.U.)	3.1 ± 0.9	2.6 ± 0.6	2.4 ± 0.7	2.3 ± 0.6	2.4 ± 0.6	2.4 ± 0.6	2.5 ± 0.7

InRMSSD: natural log of root mean square of successive RR interval differences; A.U.: arbitrary units; DOMS: delayed onset muscle soreness.

The pairwise comparisons of external and internal training load measures can be found in Tables 4 and 5, respectively. The total distance was largely smaller in the day 1 comparing to day 3 (−35%; d = −1.22) and day 6 (−49.7%; d = −1.95). Distances covered between 8.0 and 11.99 km/h^{-1} were also largely smaller in day 1 comparing to days 3 (−31.9%; d = −1.27), 4 (−35.0%; d = −1.42), 5 (−36.5%; d = −1.5), 6 (−55.8%; d = −2.69), and 7 (−38.2%; d = −1.59). No large differences were found among training days considering the variables of distances covered between 12.0 and 17.99 km/h^{-1} and >18 km/h^{-1}, as well as maxSpeed and number of sprints.

Table 4. Pairwise comparisons (standardized differences of Cohen (95% confident interval)) of external training load measures between training days (D).

Total Distance	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.48 [−1.0; 0.0]	−1.22 [−0.2; −0.6]	−0.97 [−1.8; −0.2]	−1.1 [−2.1; −0.1]	−1.95 [−2.6; −1.3]	−0.34 [−0.7; −0.0]
D2		−0.03 [−0.2; 0.2]	−0.45 [−0.6; −0.3]	−0.09 [−0.4; 0.2]	−0.21 [−0.5; 0.0]	−0.72 [−1.0; −0.5]
D3			−0.40 [−0.6; −0.2]	0.03 [−0.2; 0.3]	−0.10 [−0.3; 0.1]	−0.8 [−1.0; −0.6]
D4				0.37 [0.1; 0.6]	0.12 [−0.2; 0.4]	−0.54 [−0.8; −0.3]
D5					−0.26 [−0.5; −0.0]	−1.02 [−1.2; −0.9]
D6						−0.7 [−0.9; −0.6]
Distances 8.0–11.99 km.h^{-1} (m)	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.70 [−1.3; −0.1]	−1.27 [−1.9; −0.6]	−1.42 [−2.3; −0.5]	−1.5 [−2.5; −0.5]	−2.69 [−3.2; −2.2]	−1.59 [−2.0; −1.2]
D2		0.13 [−0.1; 0.3]	−0.53 [−0.8; −0.3]	−0.27 [−0.6; 0.0]	−0.29 [−0.6; −0.0]	−0.85 [−1.1; −0.6]
D3			−0.45 [−0.6; −0.3]	−0.15 [−0.4; 0.1]	−0.18 [−0.4; 0.1]	−0.69 [−0.9; −0.5]
D4				0.26 [0.0; 0.5]	0.12 [−0.2; 0.4]	−0.48 [−0.8; −0.2]
D5					−0.17 [−0.4; 0.1]	−0.81 [−1.0; −0.6]
D6						−0.63 [−0.8; −0.5]
Distances 12.0–17.99 km.h^{-1} (m)	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.43 [−1.0; 0.2]	−0.87 [−1.6; −0.1]	−0.90 [−1.7; −0.1]	−1.05 [−2.0; −0.1]	−1.75 [−2.3; −1.2]	−1.09 [−1.7; −0.5]
D2		0.03 [−0.2; 0.2]	−0.52 [−0.7; −0.3]	−0.24 [−0.5; 0.0]	−0.25 [−0.5; 0.0]	−0.61 [−0.8; −0.4]
D3			−0.33 [−0.5; −0.2]	−0.06 [−0.3; 0.2]	−0.07 [−0.3; 0.1]	−0.4 [−0.6; −0.2]
D4				0.21 [−0.02; 0.4]	0.11 [−0.1; 0.3]	−0.21 [−0.4; 0.0]
D5					−0.14 [−0.4; 0.1]	−0.62 [−0.8; −0.5]
D6						−0.51 [−0.7; −0.3]
Distances >18.0 km.h^{-1} (m)	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.60 [−1.4; 0.1]	−0.60 [−1.2; −0.0]	−0.48 [−1.1; 0.1]	−1.10 [−1.8; −0.4]	−0.21 [−0.7; 0.3]	−0.48 [−1.0; 0.0]
D2		−0.11 [−0.2; 0.01]	−0.46 [−0.6; −0.3]	−0.18 [−0.4; −0.0]	−0.14 [−0.3; 0.0]	−0.41 [−0.6; −0.3]
D3			−0.37 [−0.6; −0.2]	−0.09 [−0.3; 0.1]	−0.09 [−0.2; 0.1]	−0.49 [−0.6; −0.4]
D4				0.21 [−0.0; 0.4]	0.16 [−0.0; 0.4]	−0.08 [−0.3; 0.1]
D5					−0.04 [−0.2; 0.1]	−0.52 [−0.7; −0.4]
D6						−0.60 [−0.8; −0.5]
Maximum speed (km/h^{-1})	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.44 [−0.8; −0.1]	0.13 [−0.3; 0.6]	0.07 [−0.1; 0.2]	−0.22 [−0.6; 0.1]	0.57 [0.1; 1.1]	−0.81 [−1.7; 0.1]
D2		−0.14 [−0.4; 0.1]	0.03 [−0.2; 0.2]	0.15 [−0.1; 0.4]	0.26 [0.1; 0.4]	0.24 [−0.0; 0.5]
D3			0.10 [−0.0; 0.2]	0.20 [0.0; 0.4]	0.23 [0.1; 0.4]	0.32 [0.1; 0.5]
D4				0.09 [−0.1; 0.3]	0.17 [−0.0; 0.4]	0.17 [−0.1; 0.4]
D5					0.16 [0.0; 0.3]	0.07 [−0.1; 0.3]
D6						0.12 [−0.1; 0.4]
Sprints (n)	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.04 [−0.4; 0.3]	−0.08 [−0.4; 0.2]	−0.18 [−0.6; 0.2]	−0.44 [−1.0; 0.1]	0.27 [−0.2; 0.7]	0.11 [−0.4; 0.6]
D2		−0.05 [−0.2; 0.1]	−0.47 [−0.7; −0.3]	−0.04 [−0.2; 0.2]	−0.14 [−0.3; 0.0]	−0.54 [−0.7; −0.4]
D3			−0.36 [−0.6; −0.2]	0.04 [−0.1; 0.2]	−0.08 [−0.3; 0.1]	−0.62 [−0.8; −0.5]
D4				0.29 [0.1; 0.5]	0.12 [−0.1; 0.3]	−0.26 [−0.5; 0.0]
D5					−0.13 [−0.3; 0.0]	−0.78 [−1.0; −0.6]
D6						−0.65 [−0.8; −0.5]

Legend: D1: day 1; D2: day 2; D3: day 3; D4: day 4; D5: day 5; D6; day 6; D7: day 7; A.U.: arbitrary units; *n*: number; m: meters; km/h^{-1}: kilometers per hour; MaxSpeed: maximal speed; *n*: number; sprints: >24.0 km/h^{-1}.

The sRPE was much lower in day 1 comparing to day 4 (−42.3%; d = −1.53), day 5 (−52.4%; d = −2.07), and day 6 (−60.5%; d = −2.59). The sRPE was much greater in day 2 comparing to day 4 (37.0%; d = 1.27) and day 7 (49.5%; d = 1.88). Finally, sRPE was much lower at day 7 comparing to day 3 (−52.4%; d = −1.76). Considering the average HR, it was found that day 1 was much greater than day 2 (11.3%; d = 1.52) and day 3 (11.4%; d = 1.53).

The pairwise comparisons of HRV and wellness parameters can be found in Table 6. No large differences among training days were found in the pairwise comparisons for the lnRMMSD, DOMS, and fatigue reported.

Table 5. Pairwise comparisons (standardized differences of Cohen (95% confident interval)) of internal training load measures between training days (D).

sRPE (A.U.)	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	0.45 [0.1; 0.8]	−0.37 [−0.7; 0.0]	−1.53 [−2.0; −1.1]	−2.07 [−3.0; −1.1]	−2.59 [−3.3; −1.9]	0.29 [0.2; 0.4]
D2		−0.02 [−0.2; 0.1]	−1.27 [−1.5; −1.01]	−0.78 [−1.1; −0.5]	−1.08 [−1.4; −0.8]	−1.88 [−2.0; −1.7]
D3			−1.07 [−1.3; −0.9]	−0.68 [−0.9; −0.5]	−0.90 [−1.2; −0.7]	−1.76 [−1.9; −1.6]
D4				0.33 [0.1; 0.6]	0.08 [−0.2; 0.3]	−0.56 [−0.8; −0.3]
D5					−0.16 [−0.3; −0.0]	−0.78 [−0.9; −0.7]
D6						−0.76 [−0.9; −0.6]
HRmax (bpm^{-1})	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.91 [−1.5; −0.3]	−0.22 [−1.3; 0.8]	0.03 [−0.3; 0.4]	−0.38 [−0.9; 0.1]	−0.23 [−0.6; 0.2]	−0.69 [−2.2; 0.8]
D2		−0.15 [−0.4; 0.1]	−0.05 [−0.2; 0.1]	−0.32 [−0.5; −0.1]	−0.09 [−0.3; 0.1]	0.01 [−0.2; 0.3]
D3			0.03 [−0.2; 0.2]	−0.17 [−0.4; 0.0]	−0.01 [−0.3; 0.2]	0.11 [−0.2; 0.4]
D4				−0.35 [−0.6; −0.2]	−0.17 [−0.4; 0.0]	−0.05 [−0.3; 0.2]
D5					0.09 [−0.1; 0.3]	0.02 [−0.2; 0.2]
D6						0.07 [−0.1; 0.3]
HRav (bpm^{-1})	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−1.52 [−1.9; −1.1]	−1.53 [−2.4; −0.7]	−0.93 [−1.8; −0.1]	−0.60 [−0.9; −0.3]	−0.45 [−0.7; −0.2]	−0.97 [−1.7; −0.2]
D2		−0.47 [−0.6; −0.3]	−0.34 [−0.5; −0.2]	−0.71 [−0.9; −0.5]	−0.29 [−0.5; −0.1]	−0.36 [−0.6; −0.1]
D3			0.02 [−0.1; 0.2]	−0.32 [−0.5; −0.1]	0.01 [−0.2; 0.2]	0.02 [−0.2; 0.3]
D4				−0.45 [−0.6; −0.3]	−0.10 [−0.3; 0.1]	−0.60 [−3.3; 2.3]
D5					0.19 [0.0; 0.4]	0.16 [−0.1; 0.4]
D6						−0.70 [−2.8; 1.3]

Legend: D1: day 1; D2: day 2; D3: day 3; D4: day 4; D5: day 5; D6; day 6; D7: day 7; sRPE: session-RPE; HRmax: maximal heart rate; HRav: average heart rate; A.U.: arbitrary units; bpm^{-1}: beats per minute.

Table 6. Pairwise comparisons (standardized differences of Cohen (95% confident interval)) of HR variability and wellness parameters between training days (D).

lnRMMSD	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.21 [−0.5; 0.0]	−0.03 [−0.2; 0.1]	0.80 [0.5; 1.1]	0.02 [−0.1; 0.1]	−0.31 [−0.8; 0.2]	−0.12 [−0.5; 0.2]
D2		0.09 [−0.1; 0.2]	−0.12 [−0.4; 0.1]	0.08 [−0.1; −0.2]	−0.04 [−0.3; 0.2]	−0.01 [−0.3; 0.2]
D3			−0.23 [−0.5; 0.0]	−0.10 [−0.3; 0.1]	−0.13 [−0.3; 0.1]	−0.05 [−0.3; 0.1]
D4				0.08 [−0.1; 0.3]	0.10 [−0.1; 0.5]	0.18 [−0.1; 0.5]
D5					0.07 [−0.1; 0.2]	0.09 [−0.1; 0.3]
D6						0.09 [−0.1; 0.3]
DOMS	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.07 [−0.5; 0.4]	−0.30 [−0.9; 0.3]	0.50 [0.3; 0.7]	−0.09 [−0.8; 0.6]	0.08 [−0.4; 0.6]	−0.14 [−0.6; 0.3]
D2		−0.28 [−0.5; −0.1]	−0.38 [−0.6; −0.2]	−0.37 [−0.6; −0.2]	−0.32 [−0.5; −0.1]	−0.28 [−0.5; −0.1]
D3			−0.14 [−0.3; 0.0]	−0.12 [−0.3; 0.1]	−0.06 [−0.2; 0.1]	−0.01 [−0.2; 0.2]
D4				0.08 [−0.1; 0.3]	0.10 [−0.1; 0.3]	0.18 [−0.1; 0.5]
D5					0.07 [−0.1; 0.2]	0.09 [−0.1; 0.3]
D6						0.09 [−0.1; 0.3]
Fatigue	**D2**	**D3**	**D4**	**D5**	**D6**	**D7**
D1	−0.27 [−0.8; 0.2]	−0.52 [−1.1; 0.0]	0.99 [0.3; 1.7]	−0.38 [−0.9; 0.2]	−0.12 [−0.6; 0.4]	0.07 [−0.4; 0.5]
D2		−0.41 [−0.6; −0.2]	−0.60 [−0.8; −0.4]	−0.41 [−0.6; −0.2]	−0.26 [−0.5; −0.1]	−0.16 [−0.4; 0.1]
D3			−0.20 [−0.4; −0.0]	−0.09 [−0.3; 0.1]	0.05 [−0.1; 0.2]	0.15 [−0.0; 0.3]
D4				0.13 [−0.0; 0.3]	0.18 [−0.0; 0.4]	0.27 [0.1; 0.5]
D5					0.11 [−0.0; 0.3]	0.21 [0.1; 0.4]
D6						0.09 [−0.1; 0.2]

Legend: D1: day 1; D2: day 2; D3: day 3; D4: day 4; D5: day 5; D6; day 6; D7: day 7; lnRMSSD: natural log of root mean square of successive RR interval differences; A.U.: arbitrary units; DOMS: delayed onset muscle soreness.

Correlations between training load measures (both internal and external) and HRV and wellness parameters were executed. All the valid moments of assessment (sessions per trainin camps) per players were used for the analysis (n = 457). The correlations were made between the collected HR and wellness parameters of the day and the training load measures imposed one and two days before. Correlations between lnRMSSD and training load can be found in Figure 1. Small but significant correlations were found between the lnRMSSD and HRmax of one day before ($r = -0.186$; $p = 0.001$) and two days before ($r = -0.188$; $p = 0.004$) and HRav of one ($r = -0.250$; $p = 0.000$) and two days before ($r = -0.271$; $p = 0.000$).

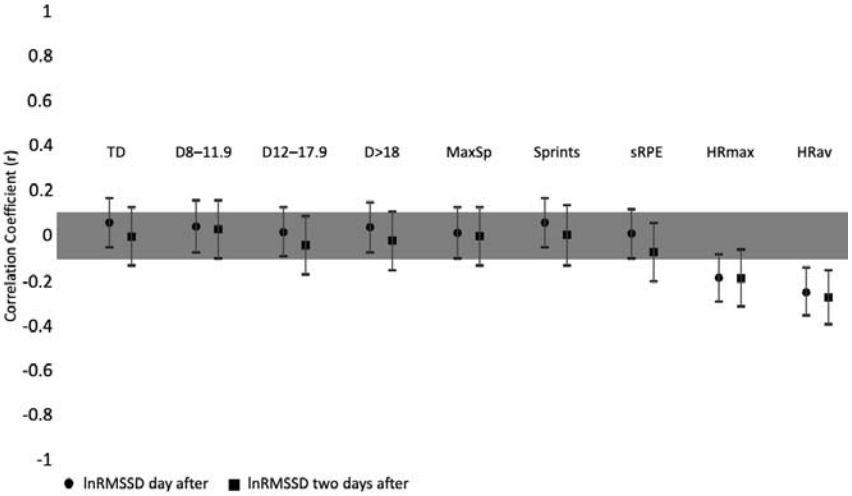

Figure 1. Correlation coefficients between lnRMSSD and training load parameters (one day before and two days before). TD: total distance; D: distance at km/h^{-1} intervals; MaxSp: maximum speed; sRPE: session-RPE; HRmax: maximal heart rate; HRav: average heart rate.

Correlations between DOMS and training load can be found in Figure 2. Moderate and significant correlations were found between DOMS and the sRPE of one ($r = -0.321$; $p < 0.000$) and two days before ($r = -0.289$; $p < 0.000$). The DOMS was small but significantly correlated with total distance of one day before (r = −0.170; $p = 0.000$). Significant but small correlations between DOMS and total distance 8.0–11.9 km/h^{-1} of one day (r = −0.210; $p < 0.000$) and two days before the session ($r = -0.150$; $p = 0.006$) were also found. The DOMS was also small but significantly correlated with maximum speed of one ($r = 0.105$; $p = 0.029$) and two days before ($r = 0.145$; $p = 0.008$). Finally, DOMS was small but significantly correlated with average HR of one ($r = -0.139$; $p = 0.004$) and two days before ($r = -0.145$; $p = 0.008$).

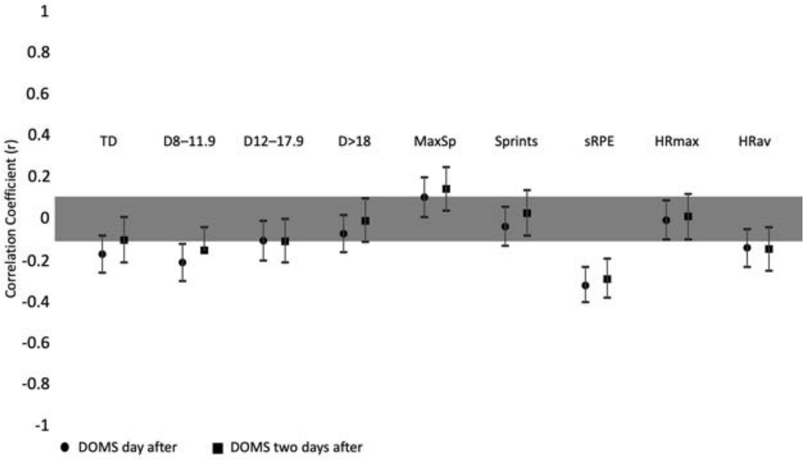

Figure 2. Correlation coefficients between DOMS (delayed onset muscle soreness) and training load parameters (of one day before and two days before). TD: total distance; D: distance at km/h^{-1} intervals; MaxSp: maximum speed; sRPE: session-RPE; HRmax: maximal heart rate; HRav: average heart rate.

Correlations between subjective fatigue and training load measures can be found in Figure 3. Moderate correlations were found between fatigue and the sRPE of one (r = −0.401; $p < 0.000$) and two days before (r = −0.344; $p < 0.000$). Fatigue was small but significantly correlated with total distance of one ($r = -0.214$; $p < 0.000$) and two days before ($r = -0.221$; $p < 0.000$).

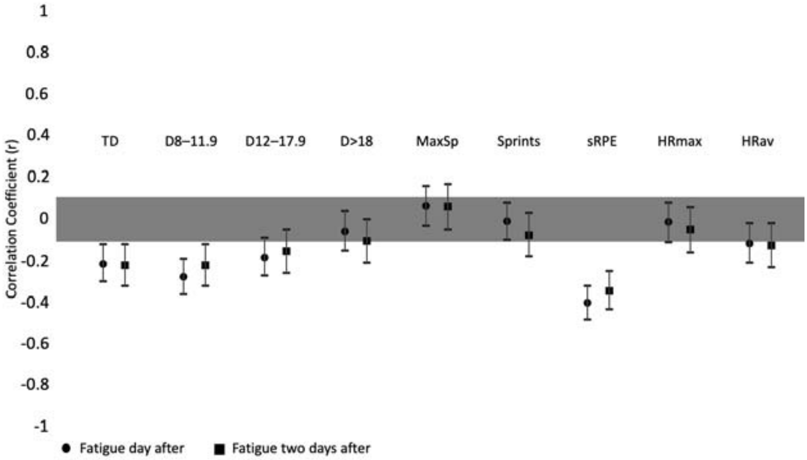

Figure 3. Correlation coefficients between fatigue and training load parameters (of one day before and two days before). TD: total distance; D: distance at km/h^{-1} intervals; MaxSp: maximum speed; sRPE: session-RPE; HRmax: maximal heart rate; HRav: average heart rate.

4. Discussion

The present study analyzed the within-week variations of training load measures and wellness parameters and tested possible relationships between the load imposed and the fluctuations in wellness across the week. The main findings revealed that the load on day 1 was significantly smaller than on day 7 in terms of total distance, distances covered between 8.0 and 11.99 km/h^{-1}, sRPE, and average HR. On the other hand, the highest HRV and better fatigue perception were found on the first day of the week. Relationships between training load variables and wellness indicators revealed moderate correlations between the load imposed and the DOMS and fatigue reported in the first and second days after the session.

The external load during training sessions has been extensively described in soccer [15]; however, in the context of futsal, such evidence is very limited. Most of the studies that report external load in futsal are centered on match demands [25]. During matches, distances of 100–120 m/min^{-1}, total distance in high-intensity running (>15 km/h^{-1}) of between 13% and 25%, and total distance in sprinting (>20 km/h^{-1}) of between 3% and 9% [26,27] are expected to be observed. Overall, during a futsal match, there can be found values between 319 and 3757 meters covered, and from such distances, 47% is performed at low intensity, 33% at medium intensity, 17% at high intensity, and 4% at maximum intensity [28].

In our study, we monitored the external load during training sessions, and the evidence pointed to distances that varied between 6000 and 10,000 meters, depending on the day. Pairwise comparisons revealed that days 1 and 7 of the training camps contributed to significantly smaller distances compared to the remaining days and that the greatest distances occurred on days 2, 3, 5, and 6 of the seven-day period. Additionally, it was found that distances between 12 and 18 km/h^{-1} varied between 1250 and 2132 meters per session, with the smallest distances occurring on days 1 and 7 and the highest occurring on days 3 and 5. Similarly, distances above 18 km/h^{-1} were the greatest on days 2 (1001 meters) and 5 (1057 meters) and smallest on days 1 (583 meters) and 7 (656 meters). No significant differences among training days were found considering maximal speed. However, the data for the number of sprints per session revealed that the fewest sprints occurred on days 1 and 7 (n = 14) and that the greatest number of sprints were observed on days 2 (n = 26), 3 (n = 25), and 5 (n = 27).

Internal load in futsal was researched more than external load. Session-RPE, as an easy-to-use and valid approach to quantifying internal load, is one of the most common measures described in the literature of futsal and training [16,19]. In the study by Moreira et al. [16], values between 6000 and 2000 A.U. per week were found in a four-week analysis of training sessions. In a longitudinal study of a full season, Clemente et al. [19] revealed values of between 338 and 693 A.U. per session. In our study, conducted during training camps, sRPE values of between 609 and 1153 A.U. per day were observed, thus showing the great load imposed during these kinds of periods. The greatest loads occurred on days 2 and 3 of the week (1106 and 1153 A.U., respectively), and the lowest loads occurred on days 1 and 7 (675 and 609 A.U., respectively). This accumulation of load may have some impact on the mechanisms of recovery. We have tested this hypothesis using a correlational approach.

Interestingly, among all the training load measures, sRPE had the strongest correlation with subjective fatigue (reported on the first and second day after the load imposed) and DOMS (also reported on the first and second day after the load imposed). The moderate and significant correlations were inverse, thus suggesting that a greater load was related to a worse perceptive status of fatigue and DOMS. Such evidence is in line with the studies conducted in soccer that revealed moderate correlations between sRPE and fatigue, DOMS, and Hooper's scores [29]. Similarly, an acute load seems to be moderately to largely correlated with DOMS and Hooper's score in elite volleyball players [30]. In our study, DOMS was also inversely correlated with total distance, distances covered between 8 and 11.9 km/h^{-1}, max speed, and average HR, albeit with a small magnitude. This suggests that sRPE can provide useful information for coaches regarding the relationships among the overall impact of a training session in the perception of recovery. In fact, a meta-analysis dedicated to examining the relationships between internal and external load measures revealed that sRPE has a consistent positive association with external load measures [31].

The present study had some limitations. One of the limitations was that the data came from a single national team, and for that reason, our findings can be used only for descriptive purposes. The training load and the associated mechanism were dependent on this team's training plans, and for that reason, studies using more than one team are highly recommended. A second limitation was that the values were means of the team; however, variability between subjects should be considered to improve the generalizability of the data. Despite this study's limitations, this study is one of the few (as far as we know) that describe the external load demands of futsal players during training camps. Moreover, it is also one of the few that analyze the load and recovery mechanisms and attempt to draw connections between them. This provides valuable information to coaches and a new insight on futsal, mainly considering the inverse relationship found between overall load imposed and the perception of DOMS and fatigue of the players.

5. Conclusions

Considering the objective of analyzing the daily variations of load and well-being, the main findings of this study revealed that during seven-day training camps, the smallest external and internal loads were imposed on days 1 and 7, while the greatest loads occurred in the middle of the week. In short, it is possible to conclude that the first and last days of training camps imposed a significantly smaller external load on players (in different intensities) and that the middle of the week was more dedicated to increasing the overall volume of physical demands. No within-week changes were found in terms of HRV, DOMS, or fatigue. Considering the second objective of the study (analyze the relationships between load and well-being measures, the main findings revealed that the DOMS and fatigue were moderately and inversely correlated with sRPE as reported one and two days before the camp.

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Supplementation with a Bioactive Melon Concentrate in Humans and Animals: Prevention of Oxidative Damages and Fatigue in the Context of a Moderate or Eccentric Physical Activity

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Abstract: Exercise is recognized to provide both physical and psychological health benefits. However, oxidative stress can occur and induce muscular damages. SOD B® M is a melon concentrate, well known to counteract oxidative stress and prevent its side effects. The present study aimed to evaluate the potential of the melon concentrate in the context of both a strong and isolated effort associated with deleterious effects, and a moderate and regular physical activity considered as beneficial. First, a preclinical study was set up on rats to evaluate its potential on the prevention of damages induced by an eccentric exercise. Secondly, the combined effect of the melon concentrate and a regular standardized physical training was studied on the overall physical condition of healthy subjects in a randomized, double-blind, placebo-controlled trial. Repeated measures Analysis of Variance (ANOVA), student's t test and Mann–Whitney test were used for statistical analyses. Melon concentrate helped to prevent gastrocnemius damages induced by the eccentric exercise. It allowed a reduction of fibrosis by approximately 38% and a reduction of Tumor Necrosis Factor- α (TNF-α) plasma level by 28%. This supplementation also induced a rearrangement of myosin fibers and an increase in PGC-1α plasma level. In the clinical study, melon concentrate was able to decrease oxidative stress and C-Reactive protein (CRP) plasma level. Besides, magnesium (Mg) plasma level was higher in the context of a regular training performed by healthy subjects supplemented with the melon concentrate. Therefore, the melon concentrate allowed a better adaptation to effort linked to PGC-1α activation: a regulator of energy metabolism. The antioxidant properties of the melon concentrate and its ability to mobilize magnesium also suggest that the supplementation could induce a better resistance to fatigue and recovery during regular physical activity.

Keywords: physical activity; oxidative stress; superoxide dismutase; eccentric exercise; melon concentrate; fatigue; PGC-1α; magnesium

1. Introduction

A regular physical activity has long been recognized as a positive health behavior, known to provide lifelong benefits such as a reduced risk of cardiovascular disease, cancer, and diabetes, but also numerous psychological benefits [1,2]. It is all the more important that modern lifestyle behaviors, which include the highest ever level of sedentarism, are recognized risk factors for chronic disorders, even responsible for a shift in disease patterns [3]. However, whereas physical activity is strongly recommended, it is also well recognized that prolonged and intense exercise is responsible for Reactive Oxygen Species (ROS) overproduction [4–6].

ROS are highly reactive molecules but natural by-products of normal cellular metabolism. Whereas a tightly regulated production of ROS is beneficial for normal physiological functions, an excessive production may have several detrimental effects. When an imbalance between their production and detoxification sets in, oxidative stress occurs and with it cells and tissues damages, especially in the muscles and joints [7–9]. To assure its oxidative balance, the body constantly acts against the formation of pro-oxidants through different defense mechanisms either enzymatic (superoxide dismutase or SOD, catalase and glutathione peroxidase), or non-enzymatic (vitamins E and C, β-carotene, uric acid and flavonoids, etc.) [6,10].

Although regular physical training is normally beneficial for fatigue and recovery, oxidative stress is likely to result in poor adaptation to effort and reduced training effectiveness. Therefore, antioxidant supplementation for athletes seems to be an interesting way to counteract oxidative stress and its side effects.

SOD B® M, a melon concentrate supplement rich in Superoxide Dismutase (SOD), has proven antioxidant and anti-inflammatory effects through a specific mechanism of action [11]. In particular, beneficial effects have been observed on markers of muscular integrity [12], cartilage integrity and inflammation of race horses supplemented with the melon concentrate [13], as well as on running performance and muscle mass of extensor digitorum longus in aged mice [14]. Additionally, melon concentrate has been shown to allow a cardiac protection of the spontaneously hypertensive rats [15,16], as well as several other benefits on diverse oxidative stress associated disorders [17–19].

Knowing the involvement of oxidative stress in physical exercise and these previous results, a two-part study was set up in order to further investigate the potential of SOD B® M supplementation on the problematic of oxidative stress in physical activity. In particular, the idea was not only to confirm the relevance of a melon concentrate supplementation in the context of an intense physical activity, but also to determine the degree of health benefits that could be observed on an ordinary population taking a moderate exercise. As a first step, the effect of the melon concentrate was assessed in rats submitted to an eccentric exercise: an unaccustomed and intense exercise known to cause muscle damages, Delayed-Onset Muscular Soreness (DOMS), as well as a high oxidative stress [20–23]. For that, we followed the protocol described by Armstrong et al. [24] with minor changes. In order to screen the global potential of the supplementation, we then studied the effect of the melon concentrate and a regular standardized physical training, considered beneficial, on the overall physical condition of healthy subjects resuming sport.

2. Materials and Methods

2.1. Preparation and Characterization of the Melon Concentrate

SOD B® M (Bionov, Avignon, France) is a dried melon juice concentrate particularly rich in SOD, which results from a patented process. Briefly, the pulp of a specific proprietary and non-GMO melon variety (Cucumis melo L.) is separated from skin and seeds and crushed before centrifugation. Then the melon juice undergoes filtration and concentration steps. Lastly, the obtained melon juice concentrate is freeze-dried. For oral administration, this dried melon juice concentrate is coated with palm oil in order to preserve its activity from the digestive enzymes secreted above the small intestine. Detailed information about the antioxidant content of this melon concentrate has been previously published [25].

2.2. Preclinical Study Design

2.2.1. Animals and Experimental Design

The present animal experiment complied with the European and French laws (permit numbers D34-172-25 and 34,179) and conform to the Guide for the Care and Use of Laboratory Animals published by the National Institute of health (National Academies Press US, Eighth edition, 2011).

A rational approach was used in order to reduce the number of animals involved in the study. Considering the statistical analyses planned (analysis of variance preceded by a normality test), n = 9 animals per group was considered appropriate in order to attain sufficient statistical power (alpha risk at 0.05).

Thirty-six Sprague-Dawley (Janvier, le Genest-St-Isle, France) of 9 weeks-old were used in the present experiments. They were housed at 22 ± 1 °C, subjected to a 12 h light/12 h dark cycle with free access to both food (A04, SAFE, Augy, France) and tap water. After one week of an adaptation period, rats were subjected to 3 days of familiarization with treadmill. During this familiarization phase, the animals ran on the treadmill for 5 min at 9 m/min the first day then 10 min at a speed of 12 m/min the two following days (non-inclined treadmill, which corresponds to an isometric work). On the fourth day, the maximum aerobic running speed (VMA) was determined. The exercise was performed on a non-inclined treadmill and consisted of an initial period called "warm up" of 5 min at 15 m/min, followed by an increment phase during which the speed was increased by 2 m/min every minute. The VMA was determined when the animal was not able to maintain the running speed. Then, rats were randomly divided into four groups (n = 9 in each) and subjected or not to an eccentric exercise with or without melon supplementation. In previous studies, melon concentrate supplement demonstrated antioxidant and anti-inflammatory effects on diverse pathologic models for daily doses between 4 U SOD [15,16] and 16 U SOD (rat equivalent dose) [17,18]. Besides, a study conducted on race horses submitted to intense training sessions [13] demonstrated the efficacy of the ingredient at a daily intake of 16 U SOD (rat equivalent dose). Therefore, the melon concentrate was given at the dose of 16 U SOD, once a day during 5 days as a pellet mixed with food.

The first group received no treatment and was not exercised (Control group: C). The second group received no treatment and was subjected to an eccentric exercise (Eccentric Exercise group: EE). The third group received the melon concentrate for 5 days with no exercise (Control treated group: CT) and the fourth group received the melon concentrate for 5 days and was subjected to an eccentric exercise (Eccentric Exercise + treatment group: EET). The eccentric exercise was performed on a rodent treadmill and the four groups completed a 3 days familiarization period with the treadmill and one session of VMA test before the start of the supplementation. Then, C and CT groups were not subjected to exercise anymore. After the last supplementation, only exercised rats without or with treatment (EE and EET group) were immediately subjected to an eccentric exercise using the protocol described by Armstrong et al. [24,26] with minor changes: rats were subjected to intermittent downhill using treadmill with a —16° inclination at the speed of 16 m/min for a total of 90 min. Therefore, the protocol included eighteen 5-min bouts separated by 2-min rest period.

At the end of the experimental period, rats were weighed and anesthetized (ketamine and xylazine, 75 and 25 mg/kg). Then, 4 mL of blood was sampled in all rats by cardiac puncture, centrifuged (2000× *g*, 10 min, 4 °C), and plasma was stored at −80 °C until analysis. Gastrocnemius muscle was excised, sectioned and stored at −80 °C or fixed in formalin 10% until histological analysis.

2.2.2. Histology of the Gastrocnemius Muscle

Paraffin-embedded gastrocnemius muscle was cut in 5 µm slices sections and mounted on Superfrost Plus glass slides (Menzel, Braunschweig, Germany).

For morphological analysis, slices were stained with hematoxylin-eosin. The number of muscle fibers was evaluated and the myocyte size was determined by measuring the shortest transverse diameter. All the analyses were performed in a blind fashion by three different observers on at least 5 transverse sections per muscle using image analysis software (ImageJ, Bethesda, MD, USA).

For fibrosis determination, gastrocnemius sections were stained with 0.1% picrosirius red and mounted in Eukitt medium. Fibrosis was quantified in five to ten given fields per animal, and expressed as the percentage of fibrous tissue area stained with picrosirius red.

2.2.3. Gastrocnemius Western Blot Analysis

The muscle protein extraction was carried out on ice in 20 mM of Tris buffer (pH 6.8) containing 150 mM sodium chloride, 1 mM Ethylenediaminetetraacetic acid (EDTA), 1% Triton 20%, 0.1% sodium dodecyl sulfate (SDS), 1% protease inhibitor cocktail (Sigma-Aldrich Darmstadt, Germany). After centrifugation (5500× *g* 15 min, 4 °C), the supernatant was collected and the extracted tissue proteins were then separated by SDS polyacrylamide gel electrophoresis. Equal amounts of proteins were loaded onto a 5% or 15% acrylamide gel with a 4% stacking acrylamide gel. Migration was conducted in a Tris-glycine-SDS buffer. After separation, proteins were transferred onto nitrocellulose membranes (Whatman, Germany).

Myosin proteins were detected by Western blot analysis. The following primary antibodies against rat skeletal slow myosin heavy chain (Sigma-Aldrich, Darmstadt, Germany), skeletal fast myosin heavy chain (Sigma-Aldrich, Darmstadt, Germany), and the control protein tubulin (R&D Systems, Minneapolis, MN, USA) were used. Expression of tubulin was used for checking the equal protein load across gel tracks. Secondary antibodies (Sigma-Aldrich, Darmstadt, Germany), coupled with alkaline phosphatase, were used for revealing the primary antibodies. Western blotting was performed according to Amersham ECL select protocol (GE Healthcare, Velizy-Villacoublay, France) and was acquired with a chemiluminescence detection system (Chemi-smart 5000, Vilbert Lourmat, Marne-la-Vallée, France). Image analysis (ImageQuant TL, GE Healthcare, France) was used for quantification after standardization within membranes by expressing the density of each band of interest relative to that of tubulin in the same lane. Results are then expressed as percent of values obtained in untreated animals.

2.2.4. Plasma Immunoassay Measurements

Plasma PGC-1α and TNF-α levels were assessed using enzyme immunoassay kits from Mybiosource (San Diego, CA, USA) and R&D Systems (Minneapolis, MN, USA). The PGC-1α immunoassay kit used gastrocnemius nuclear extract and absorbance was measured at 450 nm using a microplate reader. TNF-α immunoassay kit used gastrocnemius extract and absorbance was measured using the absorbance difference 450 nm–540 nm using a microplate reader. Results are expressed as picograms of PGC-1α or TNF-α per milligrams of total proteins.

2.3. Clinical Study

2.3.1. Methods: Tools for the Physical Evaluation

The investigation focused on the effects of the melon concentrate on physical condition improvement of healthy subjects undergoing a physical training program. The main criterion of this study concerned the improvement of the physical condition whereas secondary criteria assessed changes in physical performance, quality of life and tiredness, inflammation, ionic modifications and changes in blood oxidative status.

The overall physical condition improvement was evaluated with the Ruffier test [27,28]. In this validated test, subjects completed 30 flexions in 45 s. Three measurements of Heart Rate (HR) were taken: pre-test resting HR, HR immediately after performing the flexions, and recovery HR 60 s post-test. Those three HRs were then used to calculate the Ruffier index.

The modification of physical performance was assessed using the half Cooper test [29], an exercise consisting of running the biggest distance possible (D) in 6 min. The half Cooper allows us to evaluate the Maximal Aerobic Speed (MAS), described as the smallest running speed from which a person uses the maximum of O2 or reaches the VO_{2max} (maximum volume of oxygen that the body can use during an effort). MAS is calculated with the formula MAS = D/100 and VO_{2max} as follows: $VO_{2max} = MAS \times 3.5\ mL{\cdot}min^{-1}{\cdot}kg^{-1}$ [30].

Besides, the Resting Heart Rate and the Maximum Heart Rate (MHR) were measured at rest and during the half Cooper test respectively.

The influence of a physical activity on the quality of life and fatigue was evaluated with 2 auto evaluation surveys: SF 36® and Prevost subjective fatigue scale. SF 36® (The Health Institute, Boston, MA, USA) allows the measurement of eight aspects of the quality of life: general physical and mental health state, physical and social functioning, physical and emotional health, pain, and vitality [31]. The SF 36® questionnaire is composed of 36 questions with a total score from 36 to 149 and allow us to obtain 8 scores in every aspect.

Prevost subjective fatigue scale is a questionnaire intended to measure the impact of stress on physical fatigue of the subjects [32]. The subject has to score from 1 to 7 the following impact points on their fatigue: global fatigue perceived level, muscle pain, sleep troubles and stress.

2.3.2. Study Design

The clinical trial was an intervention study based on the individual evaluation scales described above and below. It was a monocentric study performed from November, 2016 to March, 2018. The protocol followed was a controlled clinical study vs. placebo, randomized, and double blind during 56 days ± 8 days. It was approved by the Comité de Protection des Personnes (CPP) Sud-Ouest et Outre-Mer 1 on the 29th of August 2016 and the Agence National de Sécurité du Medicament (ANSM) on the 26th of April 2016 (clinical trial registration: NCT02880657). It was also declared to the Commission Nationale de l'Informatique et des Libertés (CNIL). The calculus of the sample size needed for the clinical study was done considering that the combination of training and SOD B® M would decrease the Ruffier by 1 point with a variability of ± 1 compared to training alone [33]. In this way it was assessed that 21 subjects per group would be needed to have an adequate statistical power for 90% potency and an alpha risk equal to 0.05. Taking into account dropouts estimated at 17%, it was planned to randomize a total of 50 subjects. A call for volunteers was made in the clinical investigation centers and the volunteers for the study were pre-screened by the investigators.

The inclusion criteria were to be a man between 30 and 55 years old, to have a Body Mass Index between 18.5 and 29.5 kg/m^2, to have an insufficient/average adaptation to effort corresponding to a Ruffier index between 8 and 12 (limits included), to have a stable weight (variation < 5% over the last 3 months), a stable diet over the past 3 months, normal biological exams, a blood pressure lower than 140/90 mm Hg and no contraindication to the practice of running.

Subjects with a familial dyslipidemia or treated with statins, and subjects presenting pathologies likely to distort the results of the study or to interfere with the specific assays (arterial hypertension, type II diabetes, chronic respiratory pathology(s), rheumatic or orthopedic diseases) were excluded. Treatment with drugs that may have a doping effect (glucocorticoids, narcotics, stimulants, etc.) or consumption of psychoactive substances (cannabis, heroin, cocaine, ectasia, amphetamines) were also part of exclusion criteria.

After a pre-selection phone interview, a pre-inclusion visit (V1) took place. This visit included, as a first step, the signing of the consent and the verification of the clinical inclusion/exclusion criteria (including the Ruffier), at the Clinical Investigation Center (CIC).

Directly after V1, subjects conducted a baseline test session including the Prevost test, the SF 36® survey and the half Cooper test.

The second visit (V2) corresponded to the inclusion visit. Various blood parameters were determined after a test training session and used as baseline values. Forty-one volunteers were definitively included and participated in the clinical trial. The volunteers were assigned by randomization into two groups of 21 subjects for the treated group and 20 subjects for the placebo group

(P) and given a food supplement or a placebo for 56 days ± 8 days. The capsules were indistinguishable and were administered in a double-blind approach. Volunteers were given two small hard capsule per day corresponding to 40mg or 560 U SOD and excipients for the active supplement, and excipients only for the placebo. This dose was chosen in correlation with the preclinical study.

The training phase then started: this phase was composed of 16 standardized training sessions, performed over a period of 8 weeks, twice a week, with 2–3 resting days between each session. Those training sessions aimed to improve the physical condition of the subjects. Training session took place on a treadmill. Endurance and aerobic sessions of variable duration were alternated during this physical training. All trainings were supervised by the same referent trainer in order to harmonize training follow-up. Before each session, the coach checked the ability of the subject to perform the training (injury, tiredness, etc.) and collected the potential adverse events.

The blood samples were used for complete blood count. Biochemical analyses were also performed to evaluate the ionic changes. C-Reactive Protein (CRP) level was assessed as a marker of inflammation and the overall antioxidant defense potential was estimated via the KRL (Kit Radicaux Libres) test (“SPIRAL” laboratories, Couternon, France) which is a biological test of blood resistance based on free radical induced haemolysis. The latter test measures the global antioxidant capacity (enzymatic and non-enzymatic) of erythrocytes against a standardized assault with free radicals. Therefore, it allows the dynamic evaluation of the overall antioxidant defense potential of an individual. All the blood analyses and tests were realized at the Institut Fédératif de Biologie - CHU Toulouse Purpan.

A follow-up visit (V3) was conducted midway, after the 8th training session at the CIC. V3 aimed to check treatment adherence, potential adverse effects, and physical condition improvement at half training with the Ruffier test.

The 16th and last training session was directly followed by a final test session identical to the baseline one, in order to evaluate the evolution of the Prevost and SF $36^{®}$ scores as well as parameters of the half Cooper test. Results were attached to the final visit (V4) which took place 2 days later. At V4, subjects conducted the Ruffier test, blood was sampled and a clinical exam was performed. The clinical study design is schematized on Figure 1.

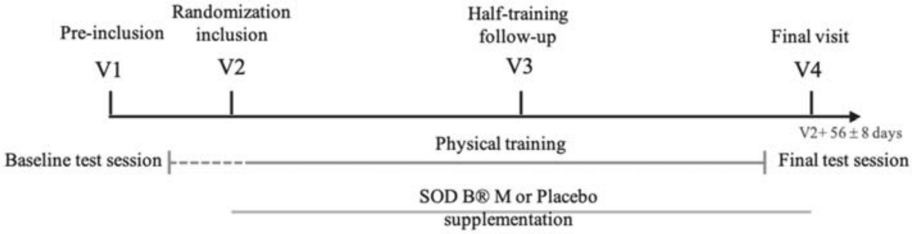


Figure 1. Schematic representation of the clinical study design. V1 includes the Ruffier test, Baseline test session includes the Prevost test, SF $36^{®}$ survey and Half Cooper test. V2 comprises a blood sampling, V3 includes the Ruffier test, Final test session includes the Prevost test, SF $36^{®}$ survey and Half Cooper test. V4 includes the Ruffier test and blood sampling.

2.4. Statistical Analyses

Values from preclinical study are presented as means ± SEM. Statistical analysis of the data was carried out using GraphPad Prism software (La Jolla, CA, USA) by one-way ANOVA followed by Mann–Whitney’s test. P-values less than 0.05 were considered to be significant.

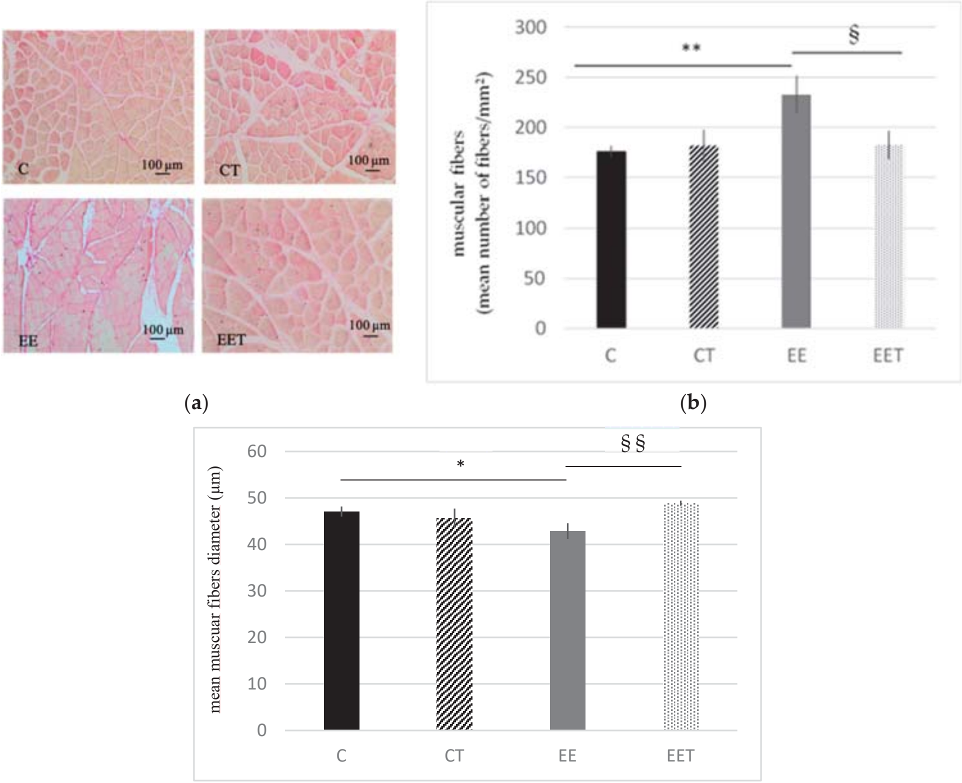
The clinical data were expressed as means ± SEM. The comparison between supplement and placebo was carried out on the differences V3–V1, V4–V1, V4–V3 and V4–V2 using either student’s t test for intragroup analysis and an Analysis of Covariance (ANCOVA), student’s t test or Mann–Whitney test for intergroup analysis. The software used for those analyses was $SAS^{®}$ version 9.4 (SAS, Cary, NC, USA). *p*-values less than 0.05 were considered to be significant.

3. Results

3.1. Preclinical Study

3.1.1. Melon Concentrate Supplementation Reduced Myocytes Damages Induced by Eccentric Exercise

As shown in Figure 2, eccentric exercise induced a 32% approximate increase in the number of muscular fibers (C 176 ± 5.5 vs. EE 233 ± 18 fibers/mm^2, $p < 0.01$) and a 9% decrease of their diameter (C 47 ± 1 vs. EE 43 ± 2 μm, $p < 0.05$). There is no significative difference between C and CT groups. On the other side, with no significant difference between C and EET group, melon concentrate supplementation was able to fully preserve muscular fibers from damages induced by eccentric effort.



(**a**) (**b**)

(**c**)

Figure 2. Influence of melon concentrate supplementation on the number and diameter of muscular fibers (gastrocnemius muscle). (**a**) Hematoxylin-Eosin Staining (HES) histochemical staining of muscular fibers. (**b**) muscular fibers were quantified on five transverse sections per muscle. Results are expressed in number of fibers/mm2 of muscle ± SEM. ** $p < 0.01$ effect of the eccentric exercise compared with control group, § < 0.05 effect of melon concentrate supplement compared with the EE group. (**c**) muscular fibers diameter was measured on five transverse sections per muscle ± SEM. Results are expressed in μm. * $p < 0.05$ effect of the eccentric exercise compared with control group, §§ < 0.01 effect of melon concentrate supplement compared with the EE group.

3.1.2. Melon Concentrate Supplementation Reduced Muscular Fibrosis Induced by Eccentric Exercise

The evaluation of muscular collagens is shown in Figure 3. The staining tends to increase in EE, compared to C group (EE 1.17 ± 0.28 vs. C 0.56 ± 0.13% of stained area, $p < 0.1$). Melon concentrate supplementation allowed us to significantly reduce this alteration by 38% ($p < 0.05$) and to preserve muscular fibers from fibrosis, in the context of an eccentric exercise. No difference was observed between C and CT groups.

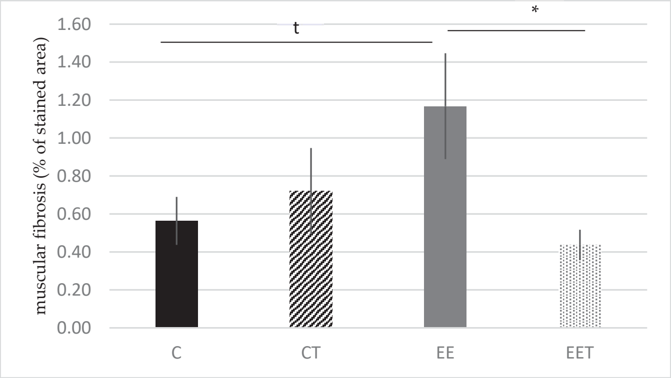


Figure 3. Influence of melon concentrate supplementation on the muscular fibrosis (gastrocnemius muscle). Muscular fibrosis measured on five transverse sections per muscle. Results are expressed as percentage of fibrous tissue area stained with picrosirius red ± SEM. t $p < 0.1$ effect of the eccentric exercise compared with control group, * < 0.05 effect of melon concentrate supplement compared with the EE group.

3.1.3. Melon Concentrate Supplementation Induced a Rearrangement of Myosin Fibers

Figure 4 represents the rearrangement of myosin fibers following melon concentrate supplementation. There was a trend that fast-twitch myosin fibers were reduced by around 38% and slow-twitch myosin fibers were 2.5 times increased in EET group in comparison with EE group ($p < 0.1$). Melon concentrate supplementation thus induced a rearrangement of myosin fibers in the context of an eccentric exercise. No difference was observed without eccentric exercise.

(**a**) (**b**)

Figure 4. Influence of melon concentrate supplementation on the gastrocnemius protein expression of myosin fibers and the PGC1α plasma level. (**a**) Quantification was made after standardization within membranes by expressing the density of the band of slow-twitch or fast-twitch myosin relative to that of tubulin in the same lane. Results are then expressed as relative change from untreated EE band intensity ± SEM. t $p < 0.1$ effect of melon concentrate supplement compared with the EE group. (**b**) Results are expressed as mean [PGC1α]/[total protein] (pg/mg) ± SEM. * $p < 0.05$ effect of the melon concentrate supplementation in the context of an eccentric exercise compared with EE group.

3.1.4. Melon Concentrate Supplementation Induced an Increase in PGC-1α Plasma Level

There was no difference in PGC-1α plasma level between group C and group EE (69.07 ± 6.29 vs. 66.50 ± 9.07 pg/mg of proteins). On the other side, as displayed in Figure 4, we observed a significant increase by approximately 76% in PGC-1α plasma level following melon concentrate supplementation, in the context of eccentric exercise (EE 66.50 ± 9.07 vs. EET 117.14 ± 8.80 pg/mg of proteins, $p < 0.05$).

3.1.5. Melon Concentrate Supplementation Induced a Decrease of TNF-α Plasma Level

Figure 5 presents the evolution of TNF-α plasma level. Eccentric activity tends to increase TNF-α level by 26% in the plasma (EE 104.32 ± 9.77 vs. C 83.03 ± 4.97 pg/mg of proteins). In addition, melon concentrate supplementation was able to significantly lower by 28% this biomarker (EET 75.04 ± 6.51 vs. EE 104.32 ± 9.77 pg/mg of proteins, $p < 0.05$). The melon concentrate supplementation allowed to maintain a normal TNF-α plasma level in the context of an eccentric effort (C vs. EET).

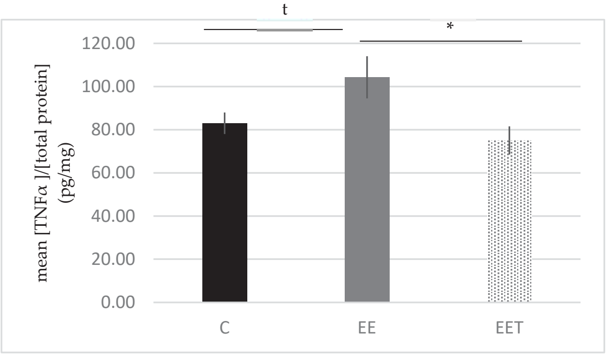


Figure 5. Influence of melon concentrate supplementation on the TNFα plasma level. Results are expressed as mean [TNFα]/[total protein] (pg/mg) ± SEM. t $p < 0.1$ effect of the Eccentric exercise compared with Control group. * $p < 0.05$ effect of the melon concentrate supplementation compared with EE group.

3.2. Clinical Study

3.2.1. Study Population

Forty-one volunteers aged between 30 and 53 years old (mean 38.8 years old) were recruited and randomized into two test groups (n = 21 for melon concentrate supplemented group (verum) and n = 20 for placebo group). Three subjects were excluded during the course of the study and therefore 38 subjects were analyzed. There were no statistical differences between the two groups at baseline.

3.2.2. Physical Condition Improvement

As shown in Table 1, there was a highly significant decrease of the Ruffier index between V1 and V3 ($p < 0.001$), observed both in the treated and in the placebo group, following intragroup analysis. The score kept decreasing during the second part of the study (V3 vs. V4) but in a non-significant manner. Overall (V1 vs. V4), both groups experienced a significant intragroup decrease of the index but with no intergroup difference.

3.2.3. Physical Performance Improvement

As presented in Table 1, HR and MHR decreased in a non-significant manner between V1 and V4 whatever the group. Besides, MAS and VO_{2max} were increased between V1 and V4. Even if there is no significant difference in MAS and $VO2_{max}$ evolution with intergroup analysis, those parameters were significantly increased in the placebo group and the supplemented group following intragroup comparison ($p < 0.001$).

Table 1. Influence of the melon concentrate supplementation on the evolution of physical performance parameters, physical condition and SF $36^{®}$ and Prevost scores. Intragroup comparison of Verum and Placebo groups. *** $p < 0.001$, t $p < 0.1$ effect of melon concentrate supplement between V1 and V4 or V1 and V3.

		Placebo	**Verum**
Evolution V1–V4	HR (b/min)	−3.3 ± 8.1	−2.9 ± 12.2
	MHR (b/min)	−4.33 ± 10.09	−2.35 ± 20.40
	MAS (km/h)	1.75 ± 1.14 ***	1.66 ± 1.24 ***
	VO_{2max}	6.14 ± 3.98 ***	5.80 ± 4.34 ***
	Prevost score	−0.8 ± 3.8	−1.4 ± 3.2 ^{t}
	SF $36^{®}$-Physical score	1.97 ± 3.69	4.8 ± 1.68 ***
	SF $36^{®}$-Mental Score	13.04 ± 3.16 ***	8.4 ± 3.14 **
Evolution V1–V3	Ruffier index	−3.00 ± 2.42 ***	−2.78 ± 2.64 ***
Evolution V3–V4	Ruffier index	−0.87 ± 2.42	−1.1 ± 3.05
Evolution V1–V4	Ruffier index	−3.87 ± 2.32 ***	−3.93 ± 2.63 ***

3.2.4. Quality of Life: Prevost Subjective Fatigue Scale and SF $36^{®}$ Health Survey

Regarding the Prevost scale, the mean of total index at baseline was 11. As reported on Table 1, Prevost score did not change in any groups between V1 and V4 using intergroup statistical analysis. However, intragroup analysis revealed a downward trend of the score in the treated group ($p = 0.068$). There was no significant difference between the two groups using intergroup analysis. However, intragroup analysis shown that physical and mental scores (SF $36^{®}$) were both increased in the supplemented group (respectively $p = 0.001$ and $p = 0.006$) whereas only the mental score was improved in the placebo group ($p = 0.001$).

3.2.5. Melon Concentrate Supplementation Decreased Oxidative Stress

Figure 6 shows the evolution of globular KRL between V2 and V4 for both groups. As presented, the erythrocyte resistance to hemolysis increased in the treated group (1.75 ± 4.04 eq mM) whereas it decreased in the placebo group (−2.44 ± 8.74 eq mM), with no intragroup variation but a significant intergroup difference ($p < 0.05$).

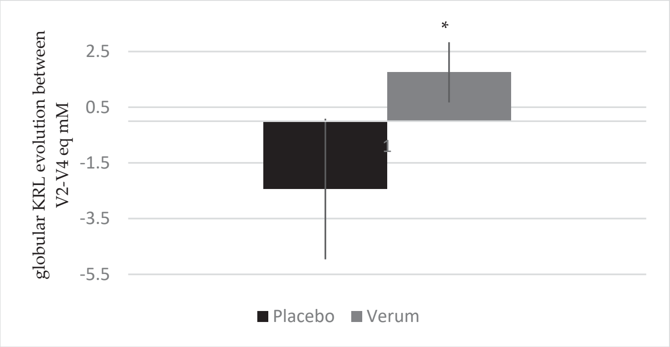


Figure 6. Influence of melon concentrate supplementation on the oxidative defenses. * $p < 0.05$ effect of the melon concentrate supplementation compared with placebo group.

3.2.6. Melon Concentrate Supplementation Tended to Decrease CRP Plasma Level

Figure 7 presents the evolution of CRP plasma level between V2 and V4. Results show that CRP slightly increased in the placebo group (1.38 ± 3.35 mg/L) and slightly decreased in the treated group (−0.68 ± 2.34 mg/L) (non-significant intragroup analysis). These opposed progressions of the two groups allow us to observe a trend in intergroup analysis ($p < 0.1$).

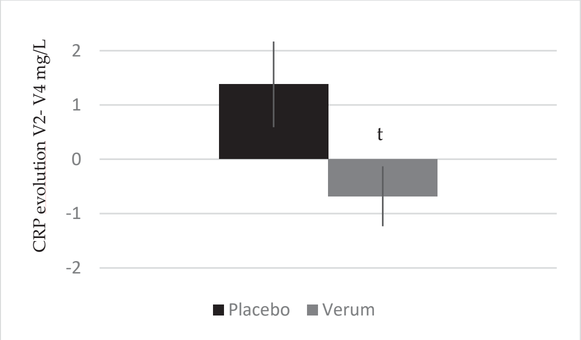


Figure 7. Influence of melon concentrate supplementation on the CRP plasma level. Results are expressed as [CRP] (mg/L) ± SEM. t $p < 0.1$ effect of the melon concentrate supplementation compared with placebo group.

3.2.7. Melon Concentrate Supplementation Modified the Platelet Level and Induced an Increase in Magnesium Plasma Level

There was a significant decrease of the platelet level between V2 and V4 in the placebo group (−5.8 ± 29.3 G/L) in comparison to the treated group where the platelet level remained stable (1.2 ± 20.2 G/L) ($p < 0.05$). There were no changes regarding the evolution of the other blood cells. Regarding blood ionic parameters, only magnesium (Mg) plasma level was impacted by the supplementation, with a significant intragroup and intergroup increase in the treated group between V2 and V4 compared to placebo (verum 0.029 ± 0.038 vs. placebo −0.010 ± 0.047 mmol/L; $p < 0.01$ in both cases) (Figure 8).

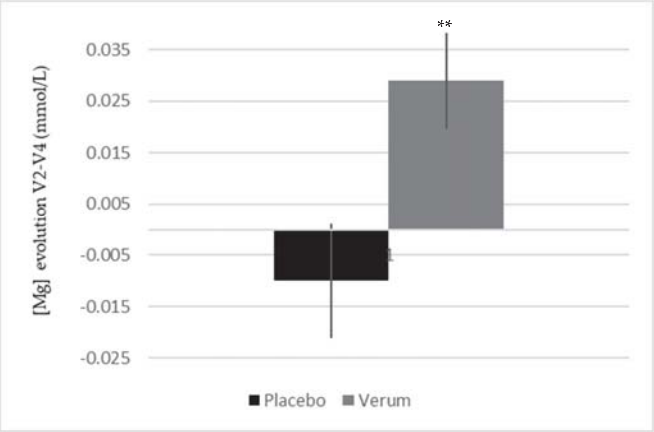


Figure 8. Influence of melon concentrate supplementation on the magnesium plasma concentration. Results are expressed as [Mg] (mmol/L) ± SEM. ** $p < 0.01$ effect of the melon concentrate supplementation compared with placebo group.

4. Discussion

The present preclinical study aimed to evaluate the potential of the melon concentrate on the prevention of damages induced by an eccentric exercise in rats. It is demonstrated that strong and unaccustomed eccentric exercise is associated with muscle damage and DOMS [23,24,34–39]. Eccentric contractions are characterized by the lengthening of the muscle-tendon complex; since fibers do not change in volume when they deform, they reduce in diameter as they increase in length [20–23]. In this preclinical study, the eccentric exercise was, as it could be expected, associated with enhanced muscular fibrosis, an increase in the density of muscular fibers suggesting an increase in their number and a reduction in their diameters in the gastrocnemius, as well as an increase in TNF-α plasma level. Conversely, the data demonstrated that the supplementation enabled all four parameters to be fully normalized. These results confirmed and extended previous reports on the beneficial effects of melon concentrate supplementation in models subjected to stressful conditions, including humans with perceived stress, horses subjected to an intense physical exercise, obese hamsters or spontaneously hypertensive rats [13,15,17,19,40].

It was evidenced in a previous unpublished transcriptomic study that the expression of PGC-1α gene was significantly increased following a melon concentrate supplementation. Therefore, special attention was paid to this key regulator of energy metabolism.

The present study confirmed that melon concentrate was able to induce an increase in PGC-1α plasma level. PGC-1α is well known to stimulate mitochondrial biogenesis, but also to promote the remodeling of muscle tissue by modulating fiber-type composition [41–43]. Interestingly, the study evidenced that the supplementation tended to induce a decrease in fast-twitch myosin fibers and an increase in slow-twitch myosin fibers following the eccentric exercise. Physiologically, fast-twitch fibers use glycolytic metabolism allowing fast and powerful contractions during a short period of time. On the other hand, slow-twitch myosin fibers are rich in mitochondrial network and with good vascularization, they allow slower and less powerful contraction, but more energetically efficient [44–46]. It is therefore possible to hypothesize that melon concentrate, through PGC-1α transcriptional coactivator, promoted the transition from fast-twitch to slow-twitch fibers in order to encourage a more energetically efficient metabolism. This would allow an improvement of endurance, performance, adaptation to effort as well as resistance to fatigue.

Besides, PGC-1α has also been involved in the antioxidant defense system and several studies indicate that it is required for the induction of many ROS-detoxifying enzymes [47–51]. At this point it is interesting to note that according to previous studies, the melon concentrate provides its antioxidative capacity through a stimulation of endogenous antioxidant defenses [16,25,52]. PGC-1α could be a link in the cascade of reaction responsible for this induction, and add new perspectives in the understanding of the melon concentrate mechanism of action.

Overall, the preclinical data demonstrated the efficacy of the melon concentrate in the context of an eccentric exercise, responsible for muscular damages. Simultaneously, the findings on PGC-1α and myosin fibers brought us a different perspective. In particular, it suggested that melon concentrate not only prevents damages induced by an eccentric effort, but also benefits the adaptation to effort in the context of a moderate and regular physical activity. The aim of the clinical study was here to evaluate the effect of the melon concentrate on the adaptation to effort and physical performance and to assess the overall potential of the supplementation in two types of exercises. For this, a healthy volunteer population, with low or medium capacity of recovery and starting a moderate physical activity, was the most relevant. Fifty healthy subjects were planned for this clinical trial but only 41 subjects reaching inclusion criteria were recruited and 38 subjects were analyzed. Even if initial population was not reached, statistical analyses were still compliant.

First of all, the training followed during this study resulted in a clear improvement of physical parameters (MAS and Ruffier index), similarly observed in the two groups. More specifically, a fast and significant reduction of the Ruffier index was observed between V1 and V3, whereas only a slight further improvement occurred during the second half of the trial (between V3 and V4). These results indicated that training had a great and rapid beneficial impact on subjects' physical condition but also suggest that intermediary assessments after a shorter training period (between V1 and V3) would have increased the probability to observe an effect of the supplement.

Among the biomarkers assessed, the antioxidant capacity of the ingredient was analyzed through the KRL, a validated and widely used test assessing blood resistance to a free radical attack [53]. Whereas no effect of the training was observed, the supplementation significantly increased the anti-free radical protection of erythrocytes compared to placebo. This indicated a better resistance to oxidative stress in the context of a moderate-intensity physical training. Similar results were previously obtained in Standardbred trotters subjected to intense and repeated training sessions after 60 days of supplementation [12]. These results supported the antioxidant effect of the melon concentrate previously demonstrated [16–18] and suggest that this prevention of oxidative damages should be applicable in both cases of an intense and a moderate activity.

The strong relationship between physical activity and inflammation has often been reported in the literature. It is well established that physical activity can increase inflammatory markers or conversely decrease them, depending on the type, the intensity and the duration of the exercise [54]. Here, training was regular and of moderate intensity, which explains that no significant increase in CRP level was observed in placebo group. On the other side, a downward trend was observed in the supplemented group, thus indicating a reduction of systemic inflammation. This finding is consistent with that of Barbé et al. [13], who observed a reduction of plasma prostaglandin E2 level in athletic horses after melon concentrate supplementation. In this particular case, the supplementation enabled to lessen the initial inflammatory state and to maintain this inhibition during and after the training session, whereas the control group experienced a significant increase in inflammation. These data confirmed the anti-inflammatory potential of the ingredient, no matter the type of exercise and the level of inflammation induced [13,18].

Another important finding was the effect of supplementation on the plasmatic Mg level. Mg is an essential mineral involved in many enzymatic systems, energetic metabolism, protein biosynthesis, as well as neuro-muscular excitability [55–58]. Several reports have shown that prolonged strenuous exercise is accompanied by hypomagnesemia. Indeed, the body responds to exercise by redistributing Mg to locations with increased metabolic need, for processes such as energy production or counteracting oxidative stress [55,56,59]. A decrease in Mg level is then characterized by a general fatigue and a tense state, both muscularly and nervously [55–58].

In this study, the concentration of plasmatic Mg was significantly increased in supplemented group, whereas it remained stable in the placebo one. This result may be explained by the antioxidant capacity of the melon concentrate: by decreasing oxidative stress, the melon juice concentrate limits the enhanced need for Mg of the most metabolically active cells, as well as its redistribution. This protective effect on Mg level could be correlated with an increased resistance to fatigue and a better recovery, with the implication for example of a delayed lactate accumulation in the muscle [60]. Moreover, these data could explain the positive effects observed on well-being and fatigue.

To that extent, Prevost and SF 36® scales were used, in order to assess a general evaluation of physical and mental condition in subjects. First, the results showed that supplementation tended to reduce Prevost score, thus traducing improved fatigue, muscular pain, sleep troubles and stress levels. Besides, melon concentrate also enabled to increase SF 36® score, with a significant rise observed for both physical and mental sections whereas placebo only had a positive effect on mental score. Those results indicated that the regular training by itself induced a positive effect on mental well-being whereas the supplementation was able to act on the physical sphere of wellness. These beneficial effects of the melon juice concentrate are in line with those reported by Milesi et al. [19] and also by Carillon et al. [61]. In those studies, conducted on subjects with perceived stress and tiredness, even more favorable outcomes were observed, not only on physical well-being but also on stress and mental health. This could suggest that, as already observed in the literature [15,62], a lower baseline is associated with an increased efficacy. Therefore, the supplementation could also be relevant in a population practicing intensive activities or in athletes submitted to both physical and mental tension.

Overall, the combined study of those experiments performing two specific types of exercises, allowing us to collect a large range of complementary data. This original approach enabled to highlight the positive effect of the melon concentrate on several biomarkers, as well as its potential on different types of physical activity. With the benefit of hindsight, the main limitation of this study was that combining the supplementation with a protocol aiming to have beneficial effects made it even more difficult to observe a positive effect of the ingredient. Adding intermediary assessments after a shorter training period could be a means of enhancing this point, and should be considered in a future trial.

5. Conclusions

Preclinical study confirmed the positive effect of melon concentrate in presence of stress factors, here an eccentric exercise. In particular, the supplementation helped to prevent gastrocnemius damages induced by the exercise, most probably through a regulation of oxidative stress. The identification of the rearrangement of myosin fibers mediated by PGC-1α oriented the study on the field of adaptation to effort, endurance, performance. The clinical trial that followed demonstrated that melon concentrate also had direct benefits on oxidative stress, inflammation reduction and Mg level protection correlated with fatigue reduction. Altogether, the present results confirm the positive effect of the melon concentrate in two types of exercise: a strong and isolated eccentric effort considered as an unfavorable exercise condition and a moderate and regular physical activity considered as favorable. It also suggests a beneficial effect on the recovery stage following an effort, which deserves further investigation.

Author Contributions: M.S. conceived, designed, and analyzed the experimental data. S.B. performed the experiments. M.S. was the coordinator of the clinical study. A.G., M.S., L.E. and B.J. interpreted the data and designed the paper. A.G. wrote the paper. All authors have read and agreed to the published version of the manuscript.

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

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Seven Weeks of Jump Training with Superimposed Whole-Body Electromyostimulation Does Not Affect the Physiological and Cellular Parameters of Endurance Performance in Amateur Soccer Players

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Abstract: Intramuscular density of monocarboxylate-transporter (MCT) could affect the ability to perform high amounts of fast and explosive actions during a soccer game. MCTs have been proven to be essential for lactate shuttling and pH regulation during exercise and can undergo notable adaptational changes depending on training. The aim of this study was to evaluate the occurrence and direction of potential effects of a 7-weeks training period of jumps with superimposed whole-body electromyostimulation on soccer relevant performance surrogates and MCT density in soccer players. For this purpose, 30 amateur soccer players were randomly assigned to three groups. One group performed dynamic whole-body strength training including 3 x 10 squat jumps with WB-EMS (EG, $n = 10$) twice a week in addition to their daily soccer training routine. A jump training group (TG, $n = 10$) performed the same training routine without EMS, whereas a control group (CG, $n = 8$) merely performed their daily soccer routine. 2 (Time: pre vs. post) x 3 (group: EG, TG, CG) repeated measures analyses of variance (rANOVA) revealed neither a significant time, group nor interaction effect for VO_2peak, Total Time to Exhaustion and La_{max} as well as MCT-1 density. Due to a lack of task-specificity of the underlying training stimuli, we conclude that seven weeks of WB-EMS superimposed to jump exercise twice a week does not relevantly influence aerobic performance or MCT density.

Keywords: electrostimulation; soccer; lactate; VO_2peak; monocarboxylate transporter

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1. Introduction

The physical demands of soccer players have increased notably within the last 10 to 20 years due to modern game tactics and their variability. For example, the ability of a team to successfully play high pressing mainly depends on the physical characteristics of the players. The distances covered in the higher intensities and the number of quick and explosive actions such as accelerations, turns, and jumps have increased within the recent years [1–3]. A player's capacity to perform numerous of those actions with a highly intense load is considered crucial in modern soccer. This ability relies on (1) adequate intra- and intermuscular coordination of soccer-specific movements and (2) metabolism that ensures proper energy delivery [4]. Both has been shown to be affected by electromyostimulation (EMS) training [5,6].

Jumps with superimposed Whole-Body EMS (WB-EMS) in addition to soccer training sessions can be effective for improving accelerations, turns, jumps, and kicking velocity [5]. WB-EMS potentially supports the athlete achieving higher power outputs and faster sport-specific movement velocities using resistance training [7] by increased firing rates and synchronization of motor units, resulting in a more pronounced activation of fast-twitch fibers at relatively low force levels [8]. Previous studies showed that local EMS is beneficially affecting muscle metabolism and can elevate energy expenditure and carbohydrate oxidation to a higher degree than voluntary contraction only [9–11]. Moreover, WB-EMS seem to stimulate anaerobic glycolysis for energy production with higher lactate accumulation [12,13]. The beneficial effects of EMS on transportation of lactate have to be taken into account as lactate shuttling via monocarboxylate transporters (MCTs) has been shown to improve high-intensity intermittent exercise performance [14,15].

MCTs are considered essential for lactate shuttling and pH regulation during exercise and can undergo notable adaptational changes depending on physical activity levels [16,17]. Due to a 1:1 ratio of lactate and H+ being transported by MCTs, an increase in the two isoforms MCT-1 and MCT-4 in skeletal muscle reduce the intracellular pH perturbations [18]. In line with this, studies revealed that the density of MCT-1 and MCT-4 proteins in muscle is elevated after a macrocycle of endurance training [19–21]. However, some training studies did not find relevant increases in MCT-4 density [22–24]. It has been assumed that MCT-1 production is more sensitive to physical stress than MCT-4. Since the biochemical characteristics of MCT-1 favors lactate uptake, it has been suggested that erythrocytes provide a lactate storage compartment in situations of physical exercise, thereby reducing the exercise-induced increase in plasma lactate concentration [25].

Interestingly, Fransson et al. [26] showed remarkable changes in MCT-4 protein expression after 4 weeks of soccer specific training regimes like speed endurance (+30%) and small sided games (+61%) in well-trained soccer players. An increase in MCT-1 and MCT-4 density in skeletal muscle after 6 weeks of strength training was however merely reported by Juel et al. [27]. No available study investigated the effects of WB-EMS on relevant endurance capacities like VO_{2max} and MCT-1 and MCT-4 in soccer players. Against this background, the aim of our 3-armed randomized controlled trial was to elucidate whether WB-EBS supplemented to a traditional soccer training routine can improve endurance capacities indices and MCT density of soccer players. Our primary hypothesis was that a training program of jumps with superimposed EMS may pronouncedly stimulate MCT-1 and MCT-4 density. Our secondary hypothesis was that endurance performance surrogates will not be affected by the training program, because of the relatively low additional training volume and the subject´s high overall training status.

2. Material and Methods

2.1. Participants

Only healthy field soccer players were included which means no cardiovascular or metabolic diseases and no preinjury in the tested muscle groups. Participants needed to compete on a regional level for the last 3 years and train 2–4 session per week with strength and conditioning training contents and play one soccer match per week. In a randomized control trial twenty-eight soccer players from 10 different teams were assigned to three different groups. Control group was assigned based on preferences and availability, whereas both intervention arms have been assigned based on coin toss. The EMS group (EG, $n = 10$) performed jumps with superimposed WB-EMS twice a week accompanied by 3 × 10 squat jumps in addition to the daily soccer routine over a period of 7 weeks that is a sufficient intervention period with WB-EMS to improve strength abilities [5,28,29]. To differentiate between the effects caused by EMS and by the squat jumps and soccer training respectively, two control groups were included. A jump training group (TG, *n* = 10) performed the same number of squat jumps without EMS on the same days as the EG and a control group (CG, *n* = 8) that only performed the daily soccer routine. All subjects were non-smokers. Basal anthropometric parameters of the subjects were presented in Table 1.

This study was carried out in accordance with the recommendations of the "Ethics Committee of the German Sports University Cologne". All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the "Ethics Committee of the German Sports University Cologne" (06–02–2014).

Table 1. Anthropometric data (mean ± SD) and Total Training Load (arbitrary units) during the 7-weeks intervention period calculated by Polar Team-2 Software according to training time spent in defined heart rates zones.

Group	Age [Year]	Height [m]	Weight [kg]	Bodyfat [%]	relVO$_2$peak [ml/kg*min^{-1}]	Sessions/ Week	Total Training Load [a.u.]
EG (*n* = 10)	24.4 ± 4.2	1.82 ± 0.03	81.4 ± 5.3	12.9 ± 2.1	52.1 ± 3.4	3.4 ± 1.2	3431 ± 911
TG (*n* = 10)	21.1 ± 1.9	1.83 ± 0.06	79.7 ± 5.5	10.8 ± 2.8	56.3 ± 5.7	3.4 ± 1.3	3479 ± 1723
CG (*n* = 8)	23.6 ± 3.9	1.82 ± 0.05	79.7 ± 7.5	14.1 ± 3.6	54.3 ± 7.2	2.6 ± 0.7	2644 ± 1437

2.2. Daily Soccer Routine

The participants performed 3.2 ± 1.0 soccer training sessions per week and competed once a week in the championships. The standard training sessions lasted approximately 90 min including technical skill activities, offensive and defensive tactics, athletic components with various intensities, small-sided game plays and continuous play. In a normal training week during season with a match on Sunday training was scheduled on Tuesdays, Wednesdays (optional), Thursdays and Fridays. Number of training sessions and the training days varied according to the game schedule playing Sunday-Sunday or Sunday-Saturday. The number of training sessions and the total training minutes were documented. The training load was measured according to the training time spent in defined heart rate zones during soccer training or match via Polar Team-2 Software (Polar Electro, Büttelborn, Germany) (see Table 1). The training load [arbitrary units] provided by the Polar-Software aims to determine internal training load based on background variables (sex, training history, metabolic thresholds, and maximal oxygen consumption [VO$_2$max]) and parameters measured during training sessions (exercise mode, and energy expenditure) (c.f. [30]). The heart rate zones (100–90%, 89–80%, 79–70%, 69–60%, 59–50%) were defined according to the individual maximum heart rate measured in the maximal ramp test (see endurance test).

The players were asked to maintain their usual food intake und hydration according to the recommendations for soccer players [31] and no nutrition supplementation was used. Additional strength training was not allowed during the study.

All players had a constant training volume during the first half of the season (July till December) and were in a well-trained condition with a relative VO$_2$peak of 54.2 ± 5.9 mL/kg·min^{-1}. All players regularly conducted strength training during first half of the season and had overall experience in strength training of 5.4 ± 3.9 years. The intervention period started after the three weeks mid-season break from end of December till mid of January. During these three weeks the training load was relatively low (moderate endurance training twice per week) in order to maintain fitness level and not negatively affect Baseline testing.

2.3. WB-EMS Application and Protocol

In order to obtain a rest interval of 48 h between the two sessions and the championship game on Sunday WB-EMS training was conducted on Tuesdays and Friday. All subjects abstained from alcohol consumption for 24 h prior to and during the training intervention. The EMS Training was conducted with a WB-EMS-system by Miha Bodytec (Augsburg, Germany). WB-EMS was applied with an electrode vest to the upper body with integrated bilaterally two paired surface electrodes for the chest (15 × 5 cm), upper and lower back (14 × 11 cm), latissimus (14 × 9 cm), and the abdominals (23 × 10 cm) and with a belt system to the lower body including the muscles of the glutes (13 × 10 cm), thighs (44 × 4 cm) and calves (27 × 4 cm). Biphasic rectangular wave pulsed currents (80 Hz) were used with an impulse width of 350 µs [5]. The stimulation intensity (mA) was determined and set separately for each muscle group (0–120 mA) by using a Borg Rating of Perceived Exertion [32]. The training intensity was defined for each player in a familiarization session two weeks before and set at a sub-maximal level that still assures a clean dynamic jump movement (RPE 16–19 "hard to very hard") and was saved on a personalized chip card. The EG performed 3 × 10 maximal squat jumps with a set pause of 60 s (no currents) per session. Every impulse for a single jump lasted for 4 s (range of motion: 2 s eccentric from standing position to an knee angle of 90°–1 s isometric–0.1 s explosive concentric–1 s landing and stabilisation) followed by a rest period of 10 s (duty cycle approx. 28%). This results in an overall time of 8.5 min per session an effective stimulation time of 2 min per session. The players started with a 2–3 min standardized warm-up with movement preparations including squats, skipping and jumps in different variations (squat jumps, jumps out of skipping or double jumps) at a light to moderate stimulation intensity. The players were told to slowly increase the intensity every few impulses. The training started when the players reached the defined training intensity that was saved on the chip card from the last session according to the RPE 16–19 ("hard to very hard"). The stimulation intensity was constantly increased individually every week (Tuesdays) controlled by the coaches in order to maintain a high stimulation intensity. The intensity was increased after the warm-up during the first and the second set of 10 squat jumps starting from calves up to the chest electrodes. The TG conducted the same standardized warm-up and performed the same amount of jumps with identical interval and conduction twice per week without EMS. The CG only performed the 2–4 soccer training session plus one match per week.

2.4. Experimental Protocol

2.4.1. Endurance Test and Assessment of Anthropometrics

For determination of the endurance parameters spirometry was performed on a WOODWAY treadmill (Woodway GmbH, Weil am Rhein, Germany) one week before (Baseline) and after the 7-weeks intervention period (Post-test) (Figure 1). Furthermore, bodymass and body composition were determined via bioelectrical impendence analysis (TANITA corp., Tokyo, Japan). Endurance tests were conducted three days after the soccer match to assure adequate recovery and not negatively influence performance. Respiratory gases were analyzed via the ZAN600-System and ZAN-Software GPI 3.xx (ZAN Austria e.U., Steyr-Dietach, Austria), using standard algorithms with dynamic account for the time delay between the gas consumption and volume signal. To calibrate the device according to the manufacturer´s guidelines, a gas mixture consisting of 5% CO_2, 16% O_2, and rest nitrogen was used (Praxair Deutschland GmbH, Düsseldorf, Germany). To measure the maximum oxygen uptake (VO_2peak), the subjects performed an incremental ramp test [33]. Thereby, the players performed a warmup at moderate speed ($3\ m\cdot s^{-1}$) with 1% incline for 3 min. In the last 30 s the incline was increased to 2.5%. Subsequently, running speed was then increased every 30 sec by 0.3 m/s until subjective exhaustion was reported. Heart rate was documented in the last 10s of a ramp stage. The VO_2peak was determined as average maximum oxygen uptake of the first 20 s after ending the test. Additionally, maximum heart rate, time to exertion (TTE) and maximum lactate concentration (La_{max}) was recorded. 27 players completed the two endurance diagnostics. One player of the TG was removed from the study due to an ankle joint injury prior to post testing.

Figure 1. Timeline of endurance testing and muscle biopsy withdrawal during the study in the 2nd half of the season.

2.4.2. Muscle Biopsies and Tissue Treatment.

Muscle biopsies via Bergström method [34] were taken from each player two weeks before (Baseline) and in the week after the last training intervention (Post-test). All biopsies were obtained under local anaesthetic from the middle portion of the vastus lateralis between the lateral part of the patella and spina iliaca anterior superior 2.5 cm below the fascia. The muscle samples were freed from blood and non-muscular material and embedded in tissue freezing medium (TISSUE TEK, Sakura, Zoeterwoude, The Netherlands). Samples were frozen in liquid nitrogen-cooled isopentane and stored at −80 °C for further analysis. The distance between the Baseline and Post-test incision was approx. 2.5 cm.

2.5. Immunohistochemistry

Muscle samples from 26 subjects were used for histology. 7 µm cross-sectional slices were obtained from the frozen muscle tissue using a cryo-microtome Leica CM 3050 C (Leica Microsystems, Nußbach, Germany) and placed on Polysine™ microscope slides (VWR International, Leuven, Belgium) [35]. Sections were fixed for 5 min in −20 °C pre-cooled acetone and air dried for 10 min at room temperature (RT), before blocking for one hour at RT with TBS (tris buffered saline, 150 mM NaCl, 10 mM Tris-HCl, pH 7.6) containing 5% BSA (bovine serum albumin). After blocking, sections were incubated overnight (4 °C) with primary antibody for MCT-1 (ab3538P; 1:500; Merck Millipore, Burlington, MA, USA) and MCT-4 (sc-376140; 1:400; Santa Cruz Biotechnology, Dallas, TX, USA), diluted in 0.8% BSA. To confirm antibody specificity, control sections were incubated in TBS containing 0.8% BSA but without primary antibody. After incubation, sections were washed 5 times short and twice for 10 min with TBS and incubated for one hour with biotinylated goat anti-rabbit secondary antibody for MCT-1 (VECTOR Laboratories, Burlingame, CA, USA), diluted 1:500 in TBS and goat anti-mouse for MCT-4 (VECTOR Laboratories), diluted 1:400 in TBS, at RT.

After that, sections were washed again 5 times for 30 s and twice for 10 min before incubation with fluorescent Alexa 488 secondary antibodies (Life Technologies, Carlsbad, CA, USA); diluted 1:500 in TBS for an hour. Afterwards sections were blocked with 5% BSA (TBS-Tween) for 30 min. Slides were then incubated overnight at 4 °C with A4.951primary antibodies (A4951; type-I myosin heavy chain; Developmental Studies Hybridoma Bank, Iowa City, IA, USA) diluted 1:200 in 0.8% BSA.

On the third day, sections were washed 5 times short and twice for 10 min with TBS before incubated again with secondary antibodies and fluorescent Alexa 555 (red) diluted 1:500 in TBS for an hour at RT. After washing again the samples, fixed on microscope glass slides, were embedded with aqualpolymount and stored at RT.

2.6. Data Analysis

The analysis of immunofluorescence stained myofibers were conducted with a confocal laser scanning microscope (LSM 510, Zeiss, Jena, Germany) at 63X fold magnification. For the analysis of MCT density in sarcoplasm and myofiber membranes, two laser channels 543 nm (for Alexa 555) and 488 nm (for Alexa 488) were used.

Two separate line-scans were conducted per measurement of membrane and sarcoplasmic areas of single myofibers to determine the staining intensity for MCT 1 and MCT 4 and the means was used for analysis (Figure 2). 1000 pixels were standardized analyzed per line scan along the membrane and the sarcoplasm. MCT density was then calculated as the mean staining intensity of all pixels along each line scan. For the analysis of type I fibers, only the green channel (Alexa 488) was used for analysis and the red channel (Alex 555) was used for fiber type determination. Laser intensity was standardized for each subject without changing throughout the analysis.

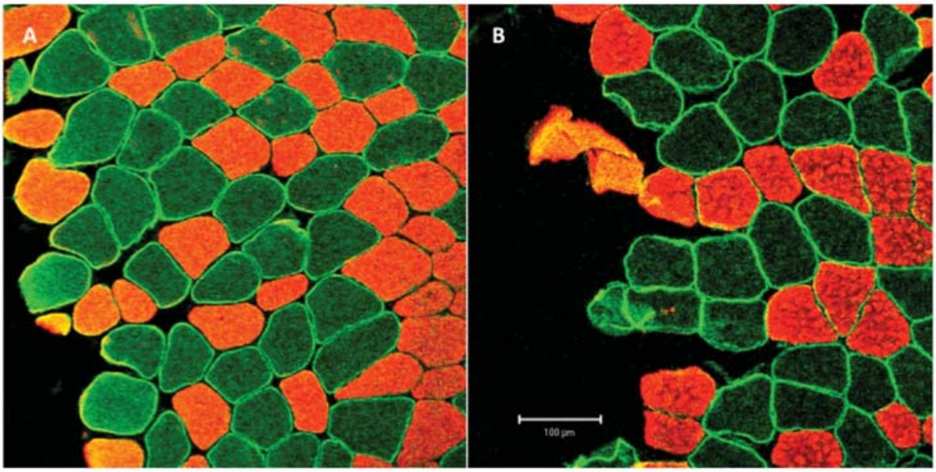

Figure 2. Representative pictures of immunofluorescence stained myofiber cross-sections showing specific MCT-1 and MCT-4 staining (green) and type 1 myofiber staining (red) within membrane and sarcoplasmic areas of myofibers (10× fold magnification). (**A**) MCT-4 Posttest, (**B**) MCT-1 Posttest.

2.7. Statistical Analysis

To determine the effect of the training interventions on endurance parameters, MCT-1 and MCT-4, separate 2 (time: pre vs. post) × 3 (group: EG, TG, CG) mixed ANOVA with repeated measures were conducted. ANOVA assumption of homogenous variances was tested using Maulchy-test of Sphericity. A Greenhouse-Geisser correction was used when a violation of Mauchly´s test was observed. To estimate overall time and interaction effect sizes, partial eta squared (η^2_p) was computed with $\eta^2_p \geq 0.01$ indicating small, ≥ 0.059 medium and ≥ 0.138 large effects [36]. If 2 × 3 mixed ANOVA revealed a time*group interaction effect on any variable, this effect was further investigated using Bonferroni post hoc tests for pairwise comparison. For all inferential statistical analyses, significance was defined as a p-value less than 0.05. All descriptive and inferential statistical analyses were conducted using SPSS 25® (IBM®, Armonk, NY, USA). Results were presented as means and standard deviations (SDs). Figures were created with Prism 6 (GraphPad Software Inc., La Jolla, CA, USA).

3. Results

3.1. Training Load

No significant differences were observed between the groups in the total number of training sessions (EG 23.9 ± 7.8; TG 25.9 ± 6.6; CG 18.1 ± 5.6 sessions), training minutes (EG 2103 ± 630; TG 1812 ± 919; CG 1437 ± 381Min), and the total recorded training load via Polar Team-2 software (Table 1). All subjects of TG and EG had a compliance of 100% (14 training sessions) for jump training and WB-EMS sessions, respectively.

3.2. Endurance Parameters

2 × 3 (time × group) ANOVA of repeated measures revealed no significant time, group or interaction effect for relative VO_2peak, TTE and La_{max}. No group differences were observed at Baseline or Posttest in none of the analyzed parameters (Figure 3).

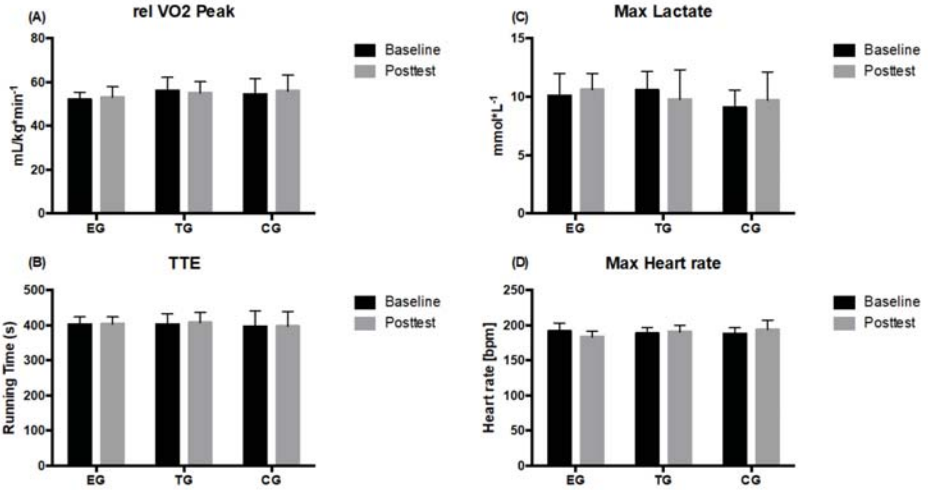

Figure 3. (**A**) Relative maximum oxygen uptake (relVO_2peak), (**B**) maximal lactate concentration, (**C**) maximal running time till exertion (TTE), and (**D**) maximal heart rate) determined at the endurance ramp-test on the treadmill in EMS-Group (EG), Training-Group (TG) and Control-Group (CG) measured before (Baseline) and after the 7 weeks intervention period (Posttest). Values are presented in means ± SD.

3.3. MCT-4

3.3.1. Type-I Fibers

The 2 × 3 (time × group) repeated measures ANOVA revealed no significant time (p = 0.119, η^2_p = 0.102), group or intervention effect (p = 0.165, η^2_p = 0.145) for the MCT-4 density in the membrane of type-I muscle fibers. Regarding cytoplasm density of the MCT-4, a large significant effect over time (p = 0.009, η^2_p = 0.26) was shown. No group*time effect (p = 0.318, η^2_p = 0.095) was however observed. Subsequent post-hoc analysis showed a significant decrease in MCT-4 density after 7 weeks for TG only (p = 0.005). No group differences were detected at Baseline and Posttest for MCT-4 density in membrane and cytoplasm in type-I fibers (Figure 4).

3.3.2. Type-II Fibers

With respect to the membrane density of the MCT-4, no time effect ($p = 0.172$, $\eta^2_p = 0.079$) or group*time interaction ($p = 0.315$, $\eta^2_p = 0.096$) of type-II fibers was shown. For the cytoplasm density of the MCT-4 a large significant main effect for the factor time ($p = 0.001$, $\eta^2_p = 0.382$) was observed in type-II fibers. No group*time interaction effect ($p = 0.333$, $\eta^2_p = 0.091$) was observed. Subsequent post-hoc analysis showed a significant decrease in MCT-4 distribution only for TG ($p = 0.004$). No differences were shown between the groups at Baseline or Posttest for MCT-4 density in the membrane and cytoplasm of the type-II fibers (Figure 4).

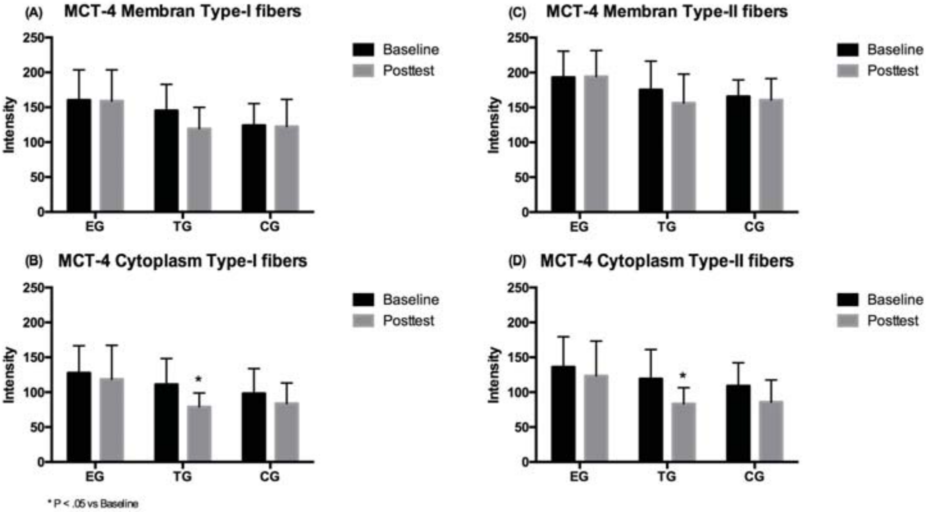

Figure 4. MCT-4 density in type-I fiber (**A**) membrane and (**B**) cytoplasm, and in type-II fiber (**C**) membrane and (**D**) cytoplasm for EMS-Group (EG), Training-Group (TG) and Control-Group (CG) measured before (Baseline) and after the 7 weeks intervention period (Posttest). Values are presented in means ± SD.

3.4. MCT-1

3.4.1. Type-I Fibers

The 2 × 3 (time × group) repeated measure ANOVA showed no significant effect over time ($p = 0.230$, $\eta^2_p = 0.065$) as well as no significant group*time interaction effect ($p = 0.045$, $\eta^2_p = 0.246$) for MCT-1 density in the membrane. Post-hoc analysis showed a significant decrease in density for the TG ($p = 0.032$). For the cytoplasm density of the MCT-1 no main effects over time ($p = 0.114$, $\eta^2_p = 0.110$) or group*time ($p = 0.416$, $\eta^2_p = 0.077$) were found. No group differences were detected at Baseline and Posttest for MCT-1 density in the membrane and cytoplasm of the type-I fibers (Figure 5).

3.4.2. Type-II Fibers

The 2 × 3 ANOVA revealed a large significant time effect for cytoplasm MCT-1 ($p = 0.009$, $\eta^2_p = 0.269$) but no group × time interaction effect ($p = 0.933$, $\eta^2_p = 0.006$). However, no significant alternations in the cytoplasm were found in the three different groups over time. For membrane density of the MCT-1 neither a time ($p = 0.104$, $\eta^2_p = 0.115$) nor an interaction effect ($p = 0.480$, $\eta^2_p = 0.065$) was observed in type-II fibers. Group comparison revealed no differences between the three groups at baseline and post-testing for MCT-1 density in the membrane and cytoplasm of the type-II fibers (Figure 5).

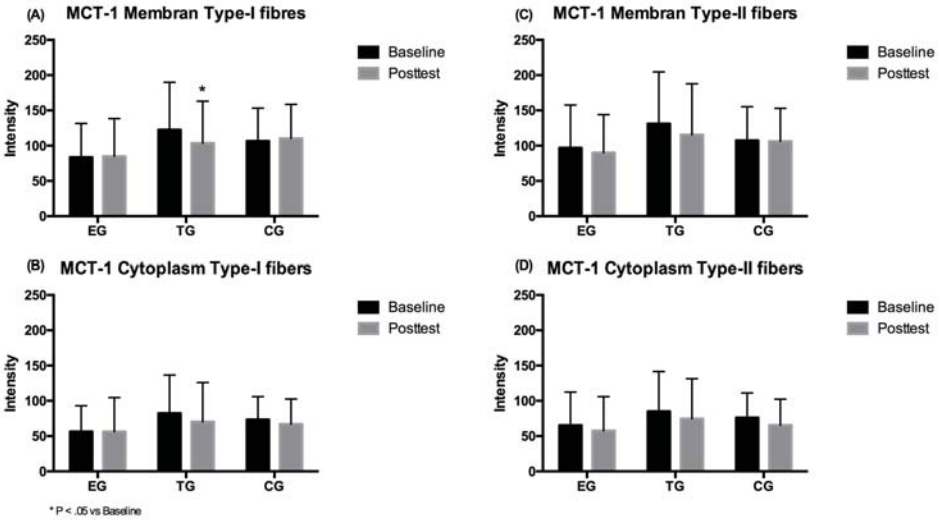

Figure 5. MCT-1 density in type-I fiber (A) membrane and (B) cytoplasm, and in type-II fiber (C) membrane and (D) cytoplasm for EMS-Group (EG), Training-Group (TG) and Control-Group (CG) measured before (Baseline) and after the 7 weeks intervention period (Posttest). Values are presented in means ± SD.

4. Discussion

The main finding of this intervention is that 7 weeks of a dynamic WB-EMS program (2 sessions per week) in addition to the regular soccer training does not relevantly influence endurance performance indices as well as MCT-1 or MCT-4 density in the muscle. We surprisingly observed that MCT-4 density in the cytoplasm and MCT-1 density in the membrane of type I muscle fibers notably decreased in TG, the group that completed jumps without EMS. Participants of all three groups (EG, TG, CG) did their weekly soccer sessions since years and training volume and training intensity was not changed during the intervention period. Thus, the results could be explained by the high overall training status of the subjects. Due to the documented effects of strength training on runner´s performance [37] and effects of EMS application on runner´s VO_2max [6], we analysed effects on some endurance parameter for EG. Indeed, WB-EMS intervention with dynamic exercises and training status of the subjects (VO_2max: 53 mL/min/kg) were similar to the study of Amaro-Gahete et al. [6]. However, the differential results might be attributed to differences in current frequency (12–90 vs. 85 Hz), higher time under tension (6 vs. 2 min) and higher intensities of exercises with superimposed EMS (strength and interval exercise vs. jumps) in the cited study. With regard to the training status of the subjects and the general high metabolic demand in soccer games and -training, the EMS stimulus could have been too low for further adaptations in endurance capacities. Consequently, no changes were observed in any parameter obtained during incremental treadmill running test. With respect to the results of Amaro-Gahete and coworkers and in order to improve endurance parameter, it might be promising to adjust exercise to higher training intensities, e.g., by shortening rest intervals of jumps or include other exercises within high intensity intervals. The authors provided recommendations for an undulating modulation of current adjustments [38], but without physiological explanation or reasoning. There are no studies available that support these results and we found only one study that applied EMS during endurance training. In this regard, Mathes and coworkers showed that, although metabolic stimuli and markers of muscle damage were higher in cycling with superimposed EMS compared to cycling without EMS, improvements of endurance performance and capacity were not significantly different between both training methods [39].

The disposed EMS-protocol concurrently to soccer training enhanced strength and myofiber adaptations [40]. Furthermore it revealed to be effective for accelerations, direction changes, vertical jumping ability and kicking velocity in elite soccer players [5]. Training design was identical within the present study. Improving such surrogate parameters of aerobic or anaerobic endurance capacities seems also promising to improve indices of soccer performance. The ability to perform sprints with high intensity bouts is influenced by anaerobic capacity. The ability to do that repeatedly critically depends on the aerobic metabolism. Both metabolic pathways are inter-linked with each other. However, for the recommendation of WB-EMS, it would be also important that no degradation occurs since soccer players need concurrent abilities of strength, speed, and endurance in the sense of repeated high-intensity actions.

MCT-1 and MCT-4 content in the muscle was not influenced by the intervention of EG. Interventions that showed increases of MCT-1 and MCT-4 conducted higher intensities and metabolic demanding exercises. It is known that high-intensity endurance training increases MCT-1 in trained subjects [41,42] and strength training increases MCT-1 and MCT-4 in untrained subjects [27]. The training program of 3 × 10 maximum jumps and 10 s between each jump was seemingly less intense. Indeed, jumps are metabolically demanding, but 10 s of rest enable adequate delivery of oxygen. Unfortunately, lactate accumulation was not measured during training in the present study. However, hundreds of repeated jumps with 8 s rest between the jumps can result in moderate steady state lactate concentrations of 3–4 mmol·L^{-1} [43]. It might be a question of the stimulus´ intensity or accumulation that need to be analyzed in exercise constellations that increase metabolic stimulus like high intensity interval training. The superimposed WB-EMS on/off-time ratio should be increased accordingly.

Our results show a significant decrease in MCT-4 and MCT-1 content after jump training without EMS (TG), which can be hardly explained, as EG and CG did not show significant differences in MCT-4 and MCT-1 content. Although the EG and TG showed equal training load (see Table 1) generally, high-intensity anaerobic effort in soccer greatly varies according to the playtime and different playing position requirements within a squad [44,45]. Furthermore, there can be differences of intensity in daily soccer training routine that could lead to fluctuations of MCT´s. This speculation is indicated by large standard deviations in total training load of TG (Table 1). Although subjects were assigned to play and train as usual, it was not possible to adjust for this influence in the study. Replication studies with accelerometer-based monitoring of the total loads are required to verify this issue. Additionally, findings warrant further studies about strength training effects to MCT. In this regard, authors have demonstrated the importance of detailed characterization of the training stimulus and the subjects [46,47]. A specification of muscular time under tension and movement dynamics like reactivity are missing in recent studies that dealt with EMS or MCT´s. The reduction effect of MCT could also be attributed to a shift of MCTs to the membrane. A previous study has shown that MCT-1 localisation after training in diabetic patients increased in the sarcolemma of muscle fibers while the sarcoplasmic content was reduced [48].

Some more limitations of the present study have to be mentioned for further research on the effects of WB-EMS to endurance capacities. Due to the small sample size the study has pilot character. Since we did not measure lactate production during training, we are not able to characterize the metabolic stimulus of the training. Moreover, the effects of running performance mainly include improved running economy, time trial performance and sprint performance all of which were not tested in the current work. A more sports-specific testing set such as the Yo-Yo Intermittent Recovery test in combination to sprint tests would have been useful in terms of ecological validity. A Further aspect of limitation is that including players from different teams can result in differences in training sessions for the CG (Table 1). A more detailed documentation of players match and training loads would be helpful to avoid bias. Future research may consider changing study design to evoke higher metabolic stimuli by increasing time under tension, reducing rest intervals or increasing intervention duration. However, the present training stimulus was designed to improve soccer specific high-intensity actions and could be integrated into daily training on a professional level [5].

5. Conclusions

We conclude despite findings that the disposed WB-EMS protocol can enhance strength [5] and myofiber adaptations [40] it is not a potent stimulation to improve VO_2max and lactate transport proteins.

Author Contributions: Conceptualization: A.F. and N.W.; Methodology: A.F., T.S. and M.d.M.; Software: A.F.; Validation: S.G., N.W. and A.F.; Formal Analysis: A.F.; Investigation: A.F., T.S. and M.d.M.; Resources: W.B.; Data Curation: A.F.; Writing—Original Draft Preparation: N.W.; Writing—Review and Editing: L.D., S.G. and N.W.; Visualization: A.F.; Supervision: L.D.; Project Administration: A.F.; Funding Acquisition: A.F. All authors have read and agreed to the published version of the manuscript.

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Article

Mental Recovery and Running-Related Injuries in Recreational Runners: The Moderating Role of Passion for Running

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Abstract: This pilot study investigates the moderating role of passion for running in the relation between mental recovery from running and running-related injuries (RRIs). We predict that the relation between recovery and injuries is dependent on the level of passion. A cross-sectional survey study was conducted among 246 Dutch recreational runners. Multivariate logistic regression analyses revealed that the negative association between mental recovery after running and RRIs is moderated (i.e., strengthened) by harmonious passion. Put differently, runners who are able to mentally recover well after running were less likely to report RRIs in the case of harmonious passion. Additionally, findings demonstrated that obsessively passionate runners were more likely to report RRIs. Passionate runners may benefit from education programs to help them integrate running more harmoniously with other aspects of life, and to prevent injuries. In addition, they should be educated about the crucial role of appropriate mental recovery from running. Considering mental aspects in running such as mental recovery from running and passion for running seems to be worthwhile to gain a better understanding of the incidence and/or prevalence of RRIs. Future (quasi-experimental) studies should investigate the issues raised here more profoundly.

Keywords: mental recovery; mental detachment; harmonious passion; obsessive passion; running-related injury; recreational running

1. Introduction

Running is becoming an increasingly popular activity among participants of recreational sports activities [1]. Although recreational running is in general considered a health-promoting activity with associated benefits such as social participation (e.g., through an increase in running groups and running events [1,2]) and stress reduction [3], running-related injuries (RRIs) occur quite often (e.g., [4]). Incidence and prevalence rates of RRIs reported in the literature are rather high (i.e., up to 80%; e.g., [5,6]). In The Netherlands, injury incidence is 6.1 injuries per 1000 sporting hours, which is about three times higher than the national sports average (i.e., 2.1 injuries per 1000 sporting hours). Specifically, Dutch runners suffer from 710,000 RRIs yearly, of which 220,000 are medically treated [7]. Next to soccer, running is the Dutch sport with the highest number of injuries. Forty percent of RRIs are overtraining/overuse injuries, and approximately one-third concerns a recurrence. Male runners are more often injured but the injury risk is higher among female runners. Runners between

20 and 34 years old are more prone to RRIs, especially female runners. Main injury locations are knees (29%), lower legs (25%), and ankles (17%) [7]. From a societal point of view, RRIs cost Dutch society approximately 10 million euros a year expressed in medical costs and costs due to work-related sickness absence and reduced work productivity [8]. Jungmalm and associates [9] concluded that RRIs can be viewed as recreational runners' primary enemy, and that the public health gains of keeping runners active and healthy should not be underestimated.

Most researchers agree that the majority of RRIs are sustained as a consequence of structural overtraining/overuse (e.g., [10]), or as a consequence of underrecovery (e.g., [11]). Yet, most existing empirical research on injury prediction and prevention focuses heavily upon the physical aspects of overtraining/overuse and underrecovery (e.g., [12]), and focuses less on their mental aspects, despite the potential role of mental aspects in injury prediction and prevention mentioned in the literature (e.g., [3,10,13,14]). As a result, evidence-based knowledge on the role of mental aspects in RRIs is still in its infancy. For that reason, the aim of the present pilot study is to investigate the role of two particular mental aspects in RRIs, namely mental recovery from running and passion for running. Specifically, using the Dualistic Model of Passion [15,16] as a heuristic framework, this study explores and tests the moderating role of passion in the mental recovery-injury relation.

1.1. Mental Recovery and Injuries

Runners are exposed to all kinds of running-related efforts. Next to the plausible and logical physical effort, they have to face mental effort as well [3]. For instance, runners often have to run focused and concentrated during races. Research has shown that it is important to compensate running-related efforts with adequate recovery to prevent RRIs (e.g., [11]). Recovery can generally be defined as a dynamic process of restoration and unwinding in which a person's functioning and efforts return to their initial levels before the efforts took place [17]. From a physical and physiological perspective, recovery reduces and prevents the accumulation of physical fatigue that leads to poor health. From a psychological perspective, it allows the individual to prepare for current or new efforts [18]. A large body of research has investigated the role of a variety of strategies aimed at promoting physical and physiological recovery from training and race efforts (e.g., [19]). In contrast, studies investigating the role of mental recovery in preventing RRIs are scarce [3,20].

In general, there are different perspectives on recovery. It can be considered as an outcome and a process [21]. Recovery as an outcome refers to a person's physical, physiological and mental state after a recovery or relaxation period. Recovery as a process refers to the activities and experiences that may lead to a change in functioning and health status. As far as the latter is concerned, several authors have argued that it is not the actual recovery activity which helps recovery (such as going for a walk, watching TV, or taking a nap), but rather the psychological processes and mechanisms behind it (e.g., [17,21,22]). In other words, persons may differ with regard to preferred recovery activities while the underlying psychological processes crucial for recovery may be uniform across persons [22]. These psychological processes and mechanisms are called 'recovery experiences' (e.g., [22]). One experience that seems to be very important for recovery to occur is mental detachment from running [3,20]. Mental detachment refers to the personal experience of leaving running behind, to mentally switch off completely, and to forget about running immediately after the run training or race [3,17,20,22]. Mental detachment goes beyond the pure physical absence from running and abstaining from running-related efforts. It implies leaving running behind oneself in psychological terms. We are aware of one recent study among 161 recreational athletes showing that mental detachment was related to reporting less sport injuries [20]. To conclude, a completely recovered runner is not only physically recovered, but is also able to mentally detach from and mentally recover after running. If recovery through effective energy management is successful, health and performance will improve, and runners may report less RRIs accordingly [3].

1.2. Passion and Injuries

A mental aspect that has gained more and more attention in sport research is passion [15,16,23]. The Dualistic Model of Passion (DMP; [15,16,24]) defines passion as a strong inclination toward a self-defining activity that people like, value, and consider important, and in which they invest considerable time and energy. The DMP suggests that different individuals can be highly committed to the same extent toward an activity such as running, and yet pursue it in qualitatively different ways. Accordingly, the DMP posits the existence of two specific types of passion: harmonious and obsessive. Harmonious passion (HP) results from an autonomous internalization of an activity into one's identity, and is characterized by a strong desire to freely engage in the passionate activity [25]. With HP, the passionate activity occupies a meaningful—but not overwhelming—place in one's life and remains in harmony with other aspects of a person's life [26]. HP is assumed to lead to flexible persistence: one is in full control of the passionate activity, so that when conditions become harmful, involvement in the activity should decline or even stop [27]. The second type of passion, obsessive passion (OP), also refers to a strong desire to engage in the passionate activity [25]. However, OP overwhelms one's attention, and is postulated to result from an overcontrolled internalization of an activity into one's identity. OP is also assumed to lead to rigid persistence: one comes to be fully controlled by the passionate activity at the expense of other activities [25,27]. OP leads the person to value the passionate activity over and above all other important activities. This often leads to conflicts either between the passionate activity and other activities, or with one's partner and relatives [26]. Empirical findings have been consistent with this conceptualization of passion (e.g., [16,24]). Where both types of passion predict similar commitment to an activity and are part of someone's identity, they have been found to be differentially associated with various outcomes (e.g., [15,16,24]). There is considerable evidence that HP is positively related to psychological outcomes (e.g., positive affect, flow, self-esteem), whereas OP is either unrelated or negatively related to these (e.g., [16,28,29]). In addition, HP has been shown to be positively associated, whereas OP is negatively associated, with experiences of conflict between one's passion and other life domains [15,30,31]. With regard to performance as an outcome, both types of passion seem to be important. However, OP may at times lead to higher performance levels than HP [16,25]. Furthermore, research on the DMP lends support for the model in sports and sport-related injuries as well. For instance, a study among 80 student dancers showed that harmoniously passionate dancers reported less acute injuries [27]. In addition, OP was associated with prolonged suffering from chronic injuries as well as more rigid involvement in dance activities when injured, whereas HP was unrelated to chronic injuries. Another study of Vallerand and colleagues [15] found that cyclists with OP were still cycling in winter on icy roads, and thus engaged in risky (i.e., injury-promoting) activities while they may be better abstain from such activities. Similar findings regarding the OP-risky behavior relation were also found in a sample of swing dancers [16] and in a study with professional dancers [32]. Finally, in their study of 170 competitive runners, Stephan and his team [33] found that OP was positively associated with perceived susceptibility to sport-related injury.

1.3. Mental Recovery, Passion, and Injuries

The current pilot study investigates the moderating role of passion for running in the relation between mental recovery and RRIs in a sample of recreational runners. First, in the case of HP, runners feel engaged with running but remain in harmony with other important activities of life. They are in full control of the passionate activity and are able to stop it whenever necessary. This implies that runners with HP are able to cease running activities at any time, are able to engage in recovery from running, mentally detach from running after a run, and feeling mentally recovered at the end. So, we expect that mental detachment from running and mental recovery after running will be negatively related to RRIs and that these relations are moderated (i.e., strengthened) by HP (Hypothesis 1). Put differently, harmoniously passionate runners who are able to mentally detach from running and/or recover well after running are less likely to report RRIs. Second, runners with OP have an uncontrollable urge to engage in running, and they highly value it over all other important activities of life. They are fully controlled by the passionate activity (rather than that they are in full control of this activity, as in HP) and will persist in running despite the body and mind signals that recovery is necessary. Thus, obsessively passionate runners will disregard their need for recovery and, hence, will be less able to mentally detach from running as well as to mentally recover after running. Consequently, they may negate minor RRIs and overtrain/overuse, or underrecover, themselves, leading to more serious RRIs in the long run (cf. [10,11]). We expect that mental detachment from running and mental recovery after running will be negatively related to RRIs, and that these relations are moderated (i.e., buffered) by OP in such a way that the associations will be less negative (Hypothesis 2). In other words, the association between (1) mental detachment from running and mental recovery after running and (2) RRIs will be weaker in the case of obsessively passionate runners.

2. Materials and Methods

2.1. Study Design, Data, and Procedure

A cross-sectional survey study was conducted in the Summer of 2017. Recreational runners were recruited via all Dutch running associations (N = 371) that were mentioned on the website of the Dutch Athletics Foundation (AU). The AU is the national umbrella organization of all Dutch athletics and running clubs, and closely linked to the International Association of Athletics Federations (IAAF) and European Athletics (EA). Both novice and more experienced runners received a unique, secured link to an online survey, where they had to fill out their email address. All participants gave their informed consent for inclusion before they participated in the study. They received information about the aim of the study and voluntary participation, and were told that their data would be handled confidentially. This pilot study was conducted in accordance with the Declaration of Helsinki and the American Psychological Association, and received institutional approval. Moreover, the Medical Ethics Committee of the University Medical Center Utrecht in the Netherlands has exempted our series of survey research studies in runners from further ethical approval (reference number: NL64342.041.17). Initially 254 recreational runners who ran at least once a week returned the questionnaire. The ultimate sample consisted of 246 runners due to some missing data. More than half of the participants were male (53.7%) and 46.3% was female. Mean age was 47.2 years (SD = 13.4; range 19–77). Average running experience was 14.4 years (SD = 12.0). On average, participants engaged in running activities 2.8 times a week (SD = 1.0). The average running distance was 26.5 kilometers per week (SD = 16.6), whereas the average running time was 3.2 hours per week (SD = 1.8). Overall, the average running speed was 10.1 km/h (SD = 18.8). Forty-two participants (17.1%) ran at least four times a week with an average running distance of 47.6 kilometers per week and an average running speed of 9.3 km/h. Ten people (4.1%) ran at least five times a week with an average running distance of 62.2 kilometers per week and an average running speed of 9.5 km/h. Two-thirds of the runners ran in groups (68.0%), and approximately half of the runners (45.5%) used an individualized training schedule for their training activities. Of all participants, 51.2% self-reported RRIs over the past 12 months, such as knee, Achilles tendon and foot injuries. These training and injury figures were comparable to other Dutch studies among recreational runners (e.g., [3,5,34]).

2.2. Variables and Instruments

2.2.1. Mental Recovery

We used two scales for mental recovery reflecting the two different perspectives mentioned earlier; that is, mental detachment from running (‘recovery process’) and mental recovery after running (‘recovery outcome’; cf. [21,22]). Scales are available from the first author upon request.

Mental detachment from running was measured with a slightly adapted scale developed by De Jonge and colleagues [35]. This scale had been used and well-validated in sports before (e.g., [3,36]). Participants were asked if they could mentally switch off from running immediately after a run training or race. The scale was measured with three items, e.g., "I could mentally distance myself from running directly after a run". Items were scored on a 5-point Likert scale, ranging from 1 (never) to 5 (always). Internal consistency of the scale expressed in Cronbach's alpha was 0.90.

Mental recovery after running was assessed with an adaptation for running of the well-validated recovery measure developed by Sonnentag and Kruel [37] to running. Participants were asked if they feel mentally recovered a couple of hours after a run training or race. The scale consisted of three items, scored on a 7-point Likert scale, ranging from 1 (totally disagree) to 7 (totally agree). An example item is: "A couple of hours after my running activities, I usually feel recovered mentally". Cronbach's alpha was 0.90. The factor structure of mental detachment and mental recovery was investigated with a factor analysis (PAF) with oblimin rotation. This factor analysis resulted in an obvious two-factor solution with all detachment items loading on one factor and all recovery items loading on the other. Eigenvalues were 3.50 and 2.15 respectively, explaining 80.7% of the variance. Pearson zero-order correlation for the two scales was $r = 0.22$ ($p = 0.001$), showing that mental detachment after running was positively but moderately related to mental recovery from running.

2.2.2. Harmonious and Obsessive Passion

Harmonious and obsessive passion were measured by the respective scales developed by Vallerand and colleagues [15,16]. The scales were slightly adapted as the passionate activity used here is 'running'. Harmonious passion emphasized a strong inclination where the runner feels engaged and has full control over running, and the activity is in harmony with the person's other activities. An example item is: "Running is well integrated in my life". As one item of the original scale did not pass psychometric scrutiny, our scale consisted of five items, with an internal consistency (Cronbach's alpha) of 0.79. Obsessive passion reflected a strong inclination where the runner feels compelled to engage in running, running takes a lot of space, the runner loses control over running, and experiences conflict with other life activities. This scale consisted of six items, for instance: "I have almost an obsessive feeling for running" (Cronbach's alpha = 0.90). Both scales were scored on a 7-point Likert scale, ranging from 1 (do not agree at all) to 7 (completely agree). We tested the factor structure of both passion scales with a factor analysis (PAF) with oblimin rotation. Results revealed a clear two-factor solution with eigenvalues of 4.92 and 1.41, explaining 63.3% of the variance. All OP items loaded on the first factor and all HP items loaded on the second factor. The two scales were not significantly related to each other ($r = 0.08$, $p = 0.222$).

2.2.3. Running-Related Injuries

Running-related injuries were self-reported by runners, and consisted of a time frame of the past 12 months. Based on a consensus definition [38,39], RRIs were defined as: "injuries, impairments or wounds, whether or not associated with pain, caused by or developed during a running training, that causes a restriction on running (in terms of duration, speed, frequency, distance, or intensity) or stoppage of running for at least seven days or three consecutive scheduled training sessions". In line with other large-scale research studies in RRIs (e.g., [6,34]), we assessed RRIs by means of a well-validated single question with a dichotomous response scale (0 = no; 1 = yes): "In the past 12 months, have you suffered one (or more) sport injuries following the above definition as a result of your running?". Injuries were overall injuries; no difference was made in acute injuries or overtraining/overuse injuries.

2.2.4. Control Variables

We controlled for gender (0 = female; 1 = male), age (years), use of an individualized training schedule (0 = no; 1 = yes), running distance per week (kilometers), and running time per week (hours). Past studies have shown that these characteristics could have a significant influence on runners' injuries (e.g., [3,5,34]). In addition, a recent meta-analysis showed that the remaining running-related characteristics are less relevant as control variables [40].

2.3. Statistical Analysis

Firstly, means, standard deviations, and Pearson zero-order correlations were calculated using IBM SPSS Statistics 25 (SPSS Inc., Chicago, IL, USA). Secondly, multivariate logistic regression analyses were used to determine the associations between mental detachment, mental recovery, passion, and RRIs. Multivariate odds ratios (ORs) and 95% confidence intervals (CIs) were derived from the logistic regression models. In all analyses, gender, age, training schedule use, running distance and running time were controlled for. Postulated moderating effects of passion (i.e., HP and OP) with recovery (i.e., mental detachment and mental recovery) were tested by adding multiplicative interaction terms (recovery × passion) of standardized recovery and passion scales into the regression model. Since we expected differential effects for the two passion scales, we performed two regression analyses accordingly: one for HP and one for OP. Nagelkerke R^2 was used as an approximation of the explained variance of the logistic regression model.

3. Results

Means (M), standard deviations (SD), and Pearson zero-order correlations for the different variables are displayed in Table 1. A first inspection of the Pearson zero-order correlations shows that our control variables were moderately but significantly related to several predictor variables and outcome variables. For instance, age was significantly related to mental detachment from running ($r = 0.24$, $p = 0.000$), mental recovery after running ($r = 0.21$, $p = 0.001$), HP ($r = 0.21$, $p = 0.001$), and RRIs ($r = -0.13$, $p = 0.046$). Next, gender was significantly associated with both running distance ($r = 0.23$, $p = 0.000$) and running time ($r = 0.20$, $p = 0.002$), while age was significantly related to running time ($r = 0.19$, $p = 0.003$). Interestingly, mental detachment from running was significantly and negatively linked to running distance ($r = -0.24$, $p = 0.000$) and running time ($r = -0.21$, $p = 0.001$) as well. Finally, both HP ($r = -0.15$, $p = 0.022$) and OP ($r = 0.14$, $p = 0.026$) as well as the interaction between mental recovery after running and HP ($r = -0.13$, $p = 0.048$) were moderately associated with RRIs.

Table 2 depicts the logistic regression results for RRIs, which showed support for an interaction model in the case of HP and, hence, a moderating effect of mental recovery after running and HP. Specifically, the negative association between mental recovery after running and RRIs is moderated (i.e., strengthened) by HP. Put differently, harmoniously passionate runners who are able to mentally recover well after running were 0.72 times (or 28%) less likely to report RRIs (OR = 0.72; 95% CI = 0.54–0.96; $p = 0.031$). However, findings did not show an interaction effect of mental detachment from running and HP, and did not show direct negative associations between mental recovery after running, mental detachment from running and RRIs as well. Overall, the predictor variables were able to explain 10.4% of the variance in RRIs. At the end, the classification accuracy shows that this prediction was correct 62.2% of the time. With respect to OP, logistic regression results showed a main effect-only model rather than an interaction model. Findings demonstrated that obsessively passionate runners were 1.36 times (or 36%) more likely to report RRIs (OR = 1.36; 95% CI = 1.03–1.85; $p = 0.047$) than others. Again, findings did not show direct negative associations between mental recovery after running, mental detachment from running and RRIs. Nagelkerke R^2 shows that the predictor variables together were able to explain 7.9% of the variance in RRIs. Finally, the classification accuracy shows that this prediction was correct 59.2% of the time.

Table 1. Descriptive statistics and Pearson zero-order correlations among study variables ($n = 246$).

Variables	M	SD	1	2	3	4	5	6	7	8	9	10	11	12	13
1. Gender (0 = female; 1 = male)	0.54 #	0.50													
2. Age (years)	47.15	13.41	0.26 **												
3. Training schedule (0 = no; 1 = yes)	0.46 #	0.50	−0.07	−0.01											
4. Running distance (km)	26.53	16.56	0.23 **	0.08	0.14 *										
5. Running time (hours)	3.20	1.82	0.20 **	0.19 **	0.08	0.69 **									
6. Mental detachment from running	2.84	1.19	0.01	0.24 **	−0.11	−0.24 **	−0.21 **								
7. Mental recovery after running	5.67	1.16	0.11	0.21 **	0.04	0.02	0.05	0.22 **							
8. Harmonious passion (HP)	2.60	1.35	−0.01	0.21 **	−0.10	−0.34 **	−0.26 **	0.32 **	0.17 **						
9. Obsessive passion (OP)	3.41	1.50	−0.05	−0.10	0.13 *	0.37 **	0.29 **	−0.43 **	−0.17 **	0.08					
10. Mental detachment × HP	0.32	1.10	0.06	−0.14 *	0.12	0.23 **	0.09	−0.06	−0.04	−0.25 **	0.05				
11. Mental recovery × HP	0.17	1.00	0.05	−0.01	0.07	0.11	0.09	−0.05	−0.17 **	−0.09	0.05	0.13 *			
12. Mental detachment × OP	−0.43	0.99	0.05	0.11	−0.06	−0.17 **	−0.09	−0.07	0.02	0.05	−0.04	−0.43 **	−0.11		
13. Mental recovery × OP	−0.17	0.96	0.07	0.06	−0.04	0.04	0.02	0.02	0.29 **	0.05	0.05	−0.12	−0.55 **	0.20 **	
14. RRIs (0 = no; 1 = yes)	0.51 #	0.50	0.07	−0.13*	−0.07	0.05	0.10	−0.11	−0.04	−0.15 *	0.14 *	0.05	−0.13 *	−0.06	0.04

* Significant at $p < 0.05$; ** significant at $p < 0.01$ (two-tailed); # these are dichotomous variables, their means can thus be interpreted as a percentage.

Table 2. Logistic regression models of running-related injuries with detachment, recovery and passion as predictor variables ($n = 246$).

	Running-Related Injuries					
	Harmonious Passion			**Obsessive Passion**		
	B	**SE**	**OR (95% CI)**	**B**	**SE**	**OR (95% CI)**
Control variables						
Gender	0.46	0.29	1.59 (0.90, 2.81)	0.49	0.28	1.63 (0.92, 2.88)
Age	−0.02	0.01	0.98 * (0.96, 0.99)	−0.03	0.01	0.96 * (0.94, 0.98)
Training schedule use	−0.40	0.28	0.67 (0.39, 1.15)	−0.37	0.27	0.69 (0.41, 1.17)
Running distance	−0.01	0.01	0.99 (0.96, 1.02)	−0.01	0.01	0.99 (0.96, 1.02)
Running time	0.17	0.13	1.18 (0.92, 1.51)	0.16	0.12	1.17 (0.92, 1.48)
Predictor variables						
Mental detachment from running	−0.03	0.15	0.97 (0.72, 1.32)	0.02	0.16	1.02 (0.75, 1.39)
Mental recovery after running	−0.02	0.14	0.98 (0.74, 1.30)	0.03	0.14	1.02 (0.78, 1.35)
Harmonious passion (HP)	−0.31	0.17	0.73 (0.53, 1.02)			
Obsessive passion (OP)				0.32	0.15	1.36 * (1.03, 1.85)
Moderating variables						
Mental detachment × Passion (HP)	0.02	0.16	1.02 (0.81, 1.36)			
Mental recovery × Passion (HP)	−0.32	0.14	0.72 * (0.54, 0.96)			
Model test	$\chi^2 = 19.37$, $df = 10$, $p = 0.036$			$\chi^2 = 15.89$, $df = 8$, $p = 0.044$		
Nagelkerke R^2	10.4%			7.9%		
Classification accuracy	62.2%			59.2%		

* Significant at $p < 0.05$ (two-tailed).

4. Discussion

The general purpose of this pilot study was to investigate the moderating role of passion for running in the relation between mental detachment from running, mental recovery after running and running-related injuries (RRIs). Based upon scientific literature and preliminary evidence, we formulated and tested two hypotheses accordingly. First, we hypothesized that both mental recovery components (i.e., mental detachment from running and mental recovery after running) will be negatively related to RRIs, and that this relation is moderated (i.e., strengthened) by HP. Findings do show the expected interaction effect between mental recovery after running and HP in the prediction of RRIs. In other words, there is only a negative association between mental recovery after running and RRIs in the case of being a harmoniously passionate runner. This supports Hypothesis 1. However, findings did not show the expected interaction effect of mental detachment from running and HP in the prediction of RRIs, which is not in support of Hypothesis 1. Second, we hypothesized that both mental recovery components will be negatively related to RRIs, and that this relation is moderated (i.e., buffered) by OP in such a way that the associations will be weaker. Results do not show the proposed interaction effect between mental recovery and OP in the prediction of RRIs. Instead, they only demonstrate a main effect of OP in the prediction of RRIs. In other words, obsessively passionate runners are more likely to report RRIs. These findings do not support Hypothesis 2. Finally, the predictor variables were able to explain about 8% to 10% of the variance in RRIs, and the predictions were correct in approximately 60% of the time. Although these effects are not very strong, they are interesting and promising.

4.1. Theoretical Implications

Findings with regard to the two mental recovery components (i.e., mental detachment from running and mental recovery after running) and RRIs are interesting. Although mental recovery after running is in general considered as being beneficial for injury prevention (e.g., [3,20]), this study shows that this may particularly be the case with harmoniously passionate runners. HP is characterized by a more flexible psychological state that should lead the runner to focus and to concentrate better, to experience less pressure, and to relax better accordingly (cf. [15,16]). In addition, mental recovery research showed that mental recovery potential is highest in cases where the need for recovery is intrinsically motivated [41]. HP could be such an intrinsic motivator.

The present study extends the work of Balk and associates [20] who showed that mental detachment from running was related to athletes' report of less injuries. In our study, however, it is not mental detachment from running but mental recovery after running in harmoniously passionate runners which seems to be negatively associated with RRIs. As both recovery measures are self-report instruments, an explanation for this finding is that recovery *outcome* measures seem to be more sensitive and concrete mental recovery measures than recovery *process* measures [21]. While the recovery outcome is related to the recovery process, it is the concrete mental recovery state directly after running which matters most for harmoniously passionate runners in the prediction of their RRIs. Moreover, if one moves beyond self-report ratings in the direction of more objective data (e.g., psycho-physiological data), disentangling the recovery process from the recovery outcome will be more difficult, or even not possible at all [21]. To conclude, this study shows that, for harmoniously passionate runners, mental recovery after running as an outcome of a successful recovery process is more important than mental detachment from running as part of the process itself to predict less RRIs.

The findings for passion for running are consistent with previous research on the concept of passion and the Dualistic Model of Passion [15,16]. The two types of passion demonstrate the way running has been internalized into a runner's identity: HP in which the person controls the activity, and OP where the activity controls the person [15]. In line with earlier passion research [16,27,33], it might be that harmoniously passionate runners are being able to detect early warning signals related to injuries and to adopt precautionary behavior such as taking a mental break in time. Conversely, obsessively passionate runners cannot stop running even when positive returns are no longer forthcoming and running has become harmful to them [15]. The non-existence of interaction effects between mental detachment, mental recovery and OP could be explained by the fact that obsessively passionate runners are not capable of detecting early warning signals related to injuries as well as adopting precautionary behavior such as taking a mental break in time. In other words, they will disregard their need for recovery and, hence, will not be able to mentally detach from running as well as to mentally recover after running. Thus, while such rigid persistence to running may initially lead to benefits such as improved performance, it may also come at personal costs such as RRIs in the end.

Finally, since obsessive passion is considered to be one of the key predictors for exercise addiction [42], our results are also consistent with research on exercise and running addiction (e.g., [42–45]). Exercise addiction can be defined based on the same criteria used to define other addictive behaviors including tolerance, withdrawal, lack of control, time, reduction in other rewarding activities, and continuance despite negative outcomes [46]. Of all the types of sports studied, endurance sports such as long-distance running are those showing the greatest risk of addiction [42,43]. For example, runners who find they need to run more to experience the same positive neuropsychological effects (e.g., runners high), experience irritability or even depression when unable to run, find that running time interferes with responsibilities in other domains (e.g., work or family), or exercise despite RRIs may have an addictive-like relation with exercise [47]. In a literature review of 25 empirical studies, Nogueira and associates [42] concluded that excessive practice may indeed cause the appearance of addictive behaviors and serious health problems. A recent study of Martin and her team [47] has highlighted the fact that endurance runners with high levels of exercise addiction pressed on in spite of the negative consequences brought about by not running, because the compensation they derive is greater than any rewards from not doing so.

4.2. Limitations and Future Research Directions

Besides its valuable insights, this study has several limitations. A first limitation concerns its cross-sectional design which does not permit any causal conclusions for the variables under study. However, this was due to the pilot character of this study. A two-wave cross-lagged panel study by Carbonneau and associates [48] in a non-sports sample showed that passion leads to changes in outcomes, but not the other way around. Further research using longitudinal study designs is needed to replicate and corroborate current findings (cf. [3]). Such studies would also contribute to the understanding of sports-related, social and psychological factors that promote or hinder the development of one type of passion over the other [27]. A second limitation is that common method variance due to using self-report data may have played a role, although recent research studies have shown that this influence is not as strong as sometimes believed (e.g., [49]). This risk was minimized by measuring our self-report scales as objective as possible ('facts') with clear instructions to fill out, accompanied with concrete and different response rates as well as profound tests on validity and reliability. The risk was further reduced by assessing the outcome measure with a different response format and anchors compared to the predictor variables, as suggested by Podsakoff, MacKenzie, and Podsakoff [50]. A third limitation is that self-reported RRIs were used. This implies that the runners had to judge the injuries themselves, without a formal diagnosis from a medical practitioner. This is partly solved by providing the runners with a clear consensus definition of RRIs as well as using the same survey question as used in other, large-scale, empirical research (e.g., [6,34]). Furthermore, the quality of RRIs was not taken into account. For instance, RRIs due to overuse or overtraining might be qualitatively different in their genesis than RRIs due to trauma. Similarly, the seriousness of RRIs might vary greatly and could have an impact on recovery schemes. It is also recommended to add more formal and comprehensive diagnostic information of RRIs by practitioners, which could enhance a study's validity in future research. Fourth, although we found direct associations between passion and RRIs, we do not know if some runners in our sample were physically predisposed to RRIs. A physical screening program at forehand would be recommended in this respect. Fifth, our logistic regression models have been adjusted for various control variables. Nevertheless, the question remains which other control variables such as participation in competition or extent performance level are associated with HP and OP. Future research might therefore consider assessing similar questions in different groups of runners. Sixth, current findings are likely to be valid for all types of recreational runners. However, it is plausible that the associations are underestimated due to the absence of elite runners (i.e., restriction of range effect). Finally, given the current sample of recreational runners, its sample size and pilot character, future research is needed whether or not the current results will hold in other samples of recreational and elite athletes as well. An example of such research is a randomized controlled trial with a 12-month follow-up [3]. After completing a web-based baseline survey, 425 half and full marathon runners were randomly assigned to either an intervention group or a control group. Participants of the intervention group obtained access to an online injury prevention programme, consisting of a running-related smartphone application and activity trackers. The smartphone application provided the participants of the intervention group with information on how to prevent overtraining/overuse and RRIs with special attention to mental aspects such as mental recovery, passion and mental fatigue. Due to a wait list control group design, participants in the control group got access to the application and related preventive information after the first follow-up measurement as well. Data collection and analysis is in progress, and will be published elsewhere (cf. [3]).

4.3. Practical Implications

The present study demonstrates the important role of passion in the relation between mental recovery and RRIs. Because many runners are devoted to and passionate about their sports, it is important to help them understand that there are two different types of passion: harmonious passion and obsessive passion. HP entails control over running and a harmonious co-existence of running with other activities in life, such as adequate mental and physical recovery. In contrast, OP entails little or even lack of control over running, rigid persistence, and conflict with other activities in a runner's life. So, HP seems to be a more desirable type of passion than OP in the case of RRIs, and runners should be encouraged to develop a more harmonious passion in this respect (cf. [26,27]). However, this does not mean that OP is negative. It may not lead to outcomes as adaptive as those derived from HP, but OP is still more adaptive than being amotivated [15]. For instance, benefits from OP are reflected by the immediate positive consequences associated with increased performance (e.g., [16,25]). Further, OP may lead to long-term commitment and persistence in running, despite its potential countereffects on RRIs. Passionate runners may benefit from education programs in order to help them integrate running more harmoniously with other aspects of life. In addition, they should be educated about the crucial role of appropriate recovery from and after running. Moreover, run coaches and trainers should be aware of the two types of passion as well, and how they characterize different ways running has been internalized into a runner's identity. Periodized training schemes and smartphone applications could then be adapted to the individual runner (cf. [3]), and ideally should take into account how to take mental breaks next to regular physical breaks (cf. [20]). Our study shows that this is particularly relevant for obsessively passionate runners.

5. Conclusions

This pilot study in recreational runners suggests that particularly the combination of harmonious passion for running and mental recovery after running is important to predict and prevent RRIs. Moreover, it suggests that obsessive passion for running is a mental risk factor for RRIs itself. So, considering mental aspects in running seems to be valuable to gain a better understanding of the incidence and/or prevalence of RRIs. Preventing and/or reducing RRIs will facilitate runners to remain active, which in turn may contribute to their health, vitality and sustainable performance—not only in sports but also in work and private life activities [51]. This can reduce medical costs and costs due to absence from work as well. Further research on the issues raised here would be rather promising.

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Article

Assessing the Perceived Exertion in Elite Soccer Players during Official Matches According to Situational Factors

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Abstract: This study aimed to assess the match perceived exertion (PE) declared by starter and non-starter junior elite soccer players, according to the level of the opponents, and by playing at home or away. Nineteen young soccer players who competed in the Spanish U19 League participated in this study. PE was registered during the entire regular season (30 official matches). Players were grouped by match playing time: starters (players who started the game and played at least 45 min) and non-starters (substitute players who participated for less than 45 min). Moreover, the matches were classified according to the opponent level (i.e., high, medium, or low) and the match location (i.e., home or away). Starters who competed against high-level opponents (8.7 ± 0.6) declared higher PE ratings than against medium (8.1 ± 0.7, $p < 0.01$) and low (8.4 ± 0.7, $p < 0.01$) level opponents. In addition, starters competing at home declared lower PE ratings than when playing away (8.2 ± 0.8 vs. 8.5 ± 0.6, $p < 0.01$). However, no significant differences ($p > 0.05$) were observed for non-starters. Coaches should consider not only tactical–strategic needs, but also these contextual factors when managing the match playing time of the starter players.

Keywords: football; match intensity; quantification; playing time

1. Introduction

The training process in soccer is considered to be an effective means for the soccer player to achieve a state of physical performance suitable for the development of the game model proposed by the coach [1]. Therefore, the challenge for physical trainers is to obtain an optimal performance level from players in official matches. Competition has high physical requirements; for example, during an official match, professional soccer players cover approximately 1000 m at high intensity (>18 km/h) and 250 m at a sprint (>21 km/h) [2]. In addition, during a soccer match, the percentage of maximum heart rate (HR_{max}) reached is close to the anaerobic threshold, usually 80–90% of HR_{max} [3], but with peaks that can reach up to 98% [4]. Several investigations have analyzed the influence of contextual factors (e.g., match location, opponent level, and match status) on these external and internal load indicators in elite soccer players [5–7]. However, in youth soccer players, the acquisition of this type of technology is complicated, which makes it difficult to quantify physical and physiological demands during official matches.

Perceived exertion (PE) has been shown to be a useful alternative to solve this problem, mainly due to it being inexpensive, easy to use, and taking little time to process the data [8,9]. In addition, it has been demonstrated in several studies on soccer players of different categories that PE is a valid and reliable tool [10–12]. In relation to playing time, only one study has shown that playing more minutes affects the PE declared by the players [13]. In this sense, more information is needed on how playing time affects the PE declared by starters and non-starters. Moreover, using the PE in the academy of elite soccer clubs would help to assess the PE match load (ML) declared by youth soccer players. Consequently, coaches could design the weekly microcycle, including adequate day-to-day training sessions and rest protocols.

In soccer, the influence of various contextual factors, such as match location, playing style, opponent ranking, time of possession, and/or match outcome, among others, could vary the demands encountered by soccer players during the match [6]. This fact could affect the training context because coaches should adapt the weekly training load of subsequent training weeks [14,15]. In this respect, only one study has analyzed the impact of these variables, specifically the match outcome on the PE reported by soccer players [16]. Fessi and Moalla [16] showed that the match PE was higher after losing in comparison to drawing or winning and higher after drawing than after winning. However, to our knowledge, no study has assessed the influence of situational factors of opponent level and match location on the PE declared by soccer players in official matches. Therefore, it would be interesting for soccer coaches to assess the consequences of these situational factors in elite junior soccer players.

Therefore, the aim of this study was to assess the match PE declared by starter and non-starter junior elite soccer players according to the level of the opponent (i.e., high, medium, or low level) and match location (i.e., home or away).

2. Materials and Methods

2.1. Participants

Nineteen outfield professional soccer players (age: 18.0 ± 0.6 years, height: 177 ± 5 cm, weight: 70.1 ± 6.8 kg, and body mass index: 22.3 ± 1.5 kg m^{-2}) participated in the study. Participants belonged to a soccer academy of the Spanish La Liga Club and they competed in the Spanish First Division Under-19 Championship. The inclusion criterion was that players had to be taking part in any match across the season. The team finished the regular season in 7th position of 16 teams. All of the participants were informed of the objectives of the research, participated voluntarily, and had the possibility to withdraw at any time from the investigation without any penalty. All of the participants, or their parents or tutors, provided written informed consent. The study was conducted according to the Declaration of Helsinki, and the protocol was fully approved by the local research ethics committee before recruitment.

2.2. Procedures

Match PE data was collected over a 30-week in-season period during the 2016–2017 season. The competition period started (i.e., first competitive match) on 4 September 2016 and ended (i.e., the last competitive match) on 7 May 2017 (i.e., a full competitive season). In order to classify the teams according to the level of the opponent, the qualifying position obtained by the team after the last day of the official league was considered [17]: (a) high-level, the top 5 classified (n = 10 matches), (b) medium-level, teams ranked between 6 and 11 (n = 10 matches), and (c) low-level, the last 5 classified (n = 10 matches). Players were also classified into two groups: (a) starters, players who started the game and played at least 45 min (n = 300 occurrences), and (b) non-starters, substitute players who participated in less than 45 min (n = 109 occurrences).

2.3. Perceived Exertion (PE)

In order to quantify the soccer match PE [13,18], the Foster's 0–10 scale was used [19]. Players responded to the question "How hard was the match?" 10 min after every match [20]. Players were allowed to mark a plus sign (interpreted as 0.5 points) alongside the integer value [13,21]. The physical trainer was responsible for asking the question of the players. Each player completed the 0–10 scale randomly without the presence of other players and could not see the values of other participants. All players were familiarized with this method during preseason training and friendly matches (six weeks). The match duration excluded the warm-up and halftime rest [13,20].

2.4. Statistical Procedures

Standard statistical methods were used for the calculation of the means and standard deviations (SDs). Match-to-match PE variability was calculated by means of the coefficient of variation ((SD/mean) × 100)). All the variables were normally distributed according to the Shapiro–Wilk test, so we used a parametric analysis. Independent paired *t*-tests were used to determine if any significant differences existed between the PE declared between starters and non-starters. The two-way ANOVA with the Bonferroni post-hoc test was used to assess the impact of the opponent level, the match location, and the interaction of both factors on the PE declared by starter and non-starter players. Practical differences were calculated using Cohen's *d* effect size (ES, large: >0.8; moderate: between 0.8 and 0.5; small: between 0.5 and 0.2; and trivial <0.2) [22]. The data analysis was carried out using the Statistical Package for Social Sciences (SPSS 21.0, IBM Corp., Armonk, NY, USA). Statistical significance was set at $p < 0.05$.

3. Results

Starters (82 ± 13 min) declared a higher PE (8.6 ± 0.7) than non-starters (23 ± 13 min; PE = 6.7 ± 2.0) ($p < 0.01$, ES = 0.95, large) during official matches. Figure 1 shows the pattern of PE declared by starter (A) and non-starter (B) players in each match during all competitive season matches (30 matches). Match-to-match PE variability was 7.6 ± 2.4% for starters and 26.0 ± 17.6% for non-starters.

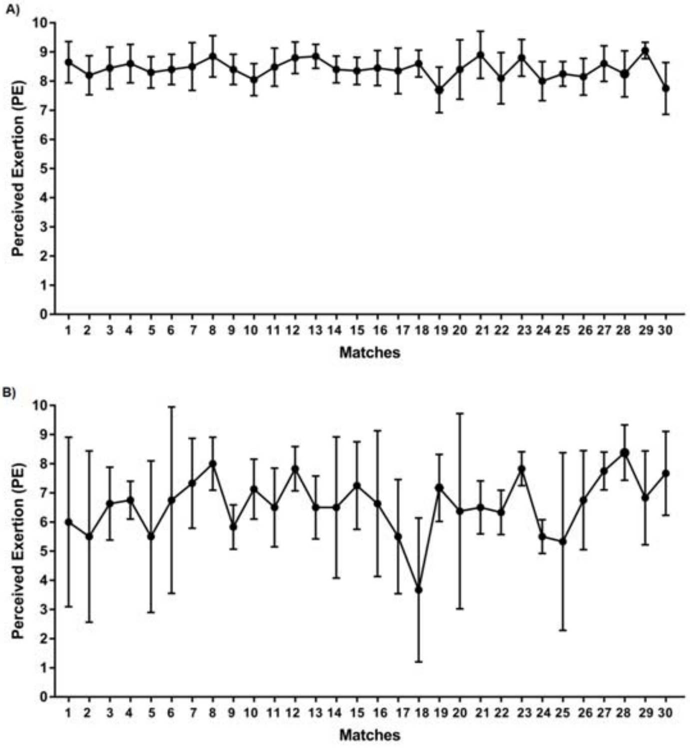

Figure 1. Perceived exertion (PE) declared by starters (**A**) and non-starters (**B**) during official matches along the season.

Starters declared a higher PE (8.7 ± 0.6) after competing against high-level opponents than after competing against medium (8.1 ± 0.7, $p < 0.01$, ES = 0.86, large) or low (8.4 ± 0.7, $p < 0.01$, ES = 0.43, small) level opponents (Figure 2). In addition, the PE declared by players was higher ($p < 0.01$, ES = 0.43, small) after competing against low-level opponents in comparison to medium-level opponents. However, no significant differences ($p > 0.05$) were observed in the PE declared by non-starters when playing against opponents of different levels.

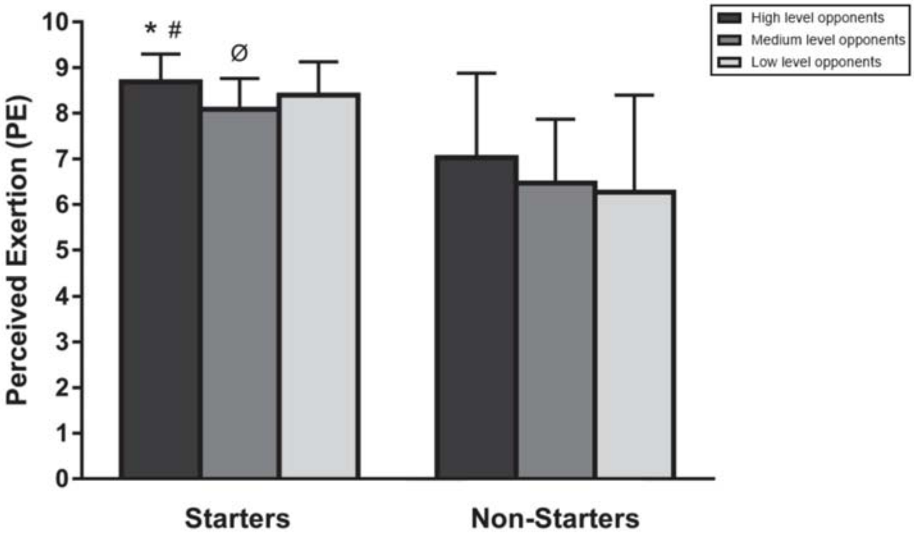

Figure 2. PE for starters and non-starters attending to the level of the opponents. * Significant differences between high- and medium-level opponents ($p < 0.01$), # Significant differences between high- and low-level opponents ($p < 0.01$), Ø Significant differences between medium- and low-level opponents ($p < 0.01$).

Starters declared a lower PE when competing at home than when playing away (8.3 ± 0.8 vs. 8.5 ± 0.6, $p < 0.01$, ES = 0.33, small, Figure 3). However, no significant differences ($p > 0.05$) were observed for non-starters regardless of playing at home or away. A two-way ANOVA revealed no significant differences ($p > 0.05$) in PE in the interaction of the factors "level of the opponents" and "playing at home or away" for either starters or non-starters.

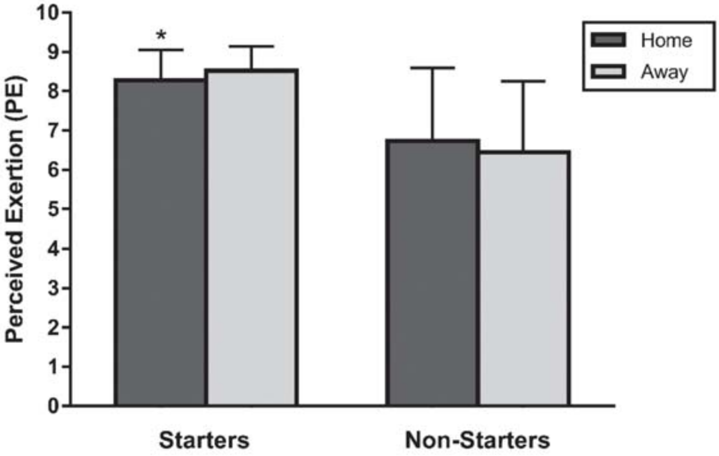

Figure 3. PE for starters and non-starters attending to playing at home or away. * Significant differences between home and away matches ($p < 0.01$).

4. Discussion

The aim of this study was to assess the match PE declared by starter and non-starter junior elite soccer players according to the level of the opponents and playing at home or away. The main findings were: (1) starter and non-starter Spanish junior elite soccer players rated the match as very hard (PE = 8.6 ± 0.7) or hard (PE = 6.7 ± 2.0), respectively; and (2) the competition level of the opponents and playing at home or away affected the match PE declared by starter players; this did not influence the non-starters.

In comparison to English junior elite soccer players that played the entire match (90 min) (PE = 8.4 ± 0.6) [23], starter Spanish junior elite players declared a similar match PE (8.6 ± 0.7). However, these values were higher than the differentiated match PE declared by young professional senior players after playing 90 min (respiratory PE = 6.7 ± 1.3; muscular PE = 6.9 ± 1.6) [13], and by professional soccer players belonging to Qatar´s Stars League that played more than 80 min (≈6.5) [16]. Similarly, non-starter players declared a lower overall PE (6.7 ± 2.0) than young Spanish professional senior soccer players (respiratory PE = 4.6 ± 1.5; muscular PE = 4.1 ± 1.6) [13]. This finding could be explained by the greater high-speed running and sprinting distance imposed on starter players in comparison to non-starter ones in junior soccer players [24]. Therefore, the coaches of elite junior teams should consider that the game is harder for their players than for senior professional players when planning the training strategies of the previous and subsequent weeks. On the other hand, and as expected and found previously in several studies [9,13,25], the starter players declared a significantly higher PE than the non-starters (8.6 ± 0.7 vs. 6.7 ± 2.0, $p < 0.01$; ES = 0.39) due to a greater number of playing minutes. Thus, match playing time can determine the match load and consequently the weekly training load of the players [25].

Despite several studies having assessed match external load variability in soccer players [26–28], few studies have assessed the match-to-match PE variability [13]. In line with Los Arcos et al. [13], the between-match PE variability was lower (coefficient of variation (CV) = 7.6 ± 2.4%) for starters than for non-starters (CV = 26.0 ± 17.6%). This suggests that, in addition to playing time, the match PE declared by each player should be considered in order to design post-match training sessions [13]. Moreover, the variability values were lower in starters (CV, 7.6% vs. ≈14%) but similar in non-starters (CV, 26% vs. ≈25%) in comparison to senior professional soccer players [13]. It seems that elite junior matches are harder and more stable than professional soccer matches. Other investigations, focused on external loading, observed a CV of 18.1 and 37.7% in high-intensity running (19.8–25.2 km·h^{-1}) and sprinting speeds (>25.2 km·h^{-1}), respectively [26].

Several investigations have analyzed the influence of match location, opponent level, and match status on the external load indicators during official matches [5–7], but few studies have investigated the effects of these factors on the PE. While the level of the opponents did not affect the match PE of non-starter players, the starter players of the medium competition level team declared a higher match PE ($p < 0.01$) after the matches played against high-level opponents in comparison to medium- and low-level opponents (Figure 1). Moreover, the match PE was higher when playing against lower level opponents than when playing against medium-level opponents. Similarly, the match location did not influence the match PE of non-starter players, but the starters declared a greater PE ($p < 0.01$) when they disputed the matches away compared to when they played at home (PE = 8.5 ± 0.6 vs. 8.3 ± 0.8). Additionally, no interaction effect on the PE declared by players between the opponent ranking of the teams faced and match location was found. From a practical approach, it would be necessary to analyze the influence of these contextual factors separately.

Attending to the aforementioned findings, these contextual factors only have an individual effect when soccer players participate in a large number of minutes in the match, as it is harder to play against high-level teams and away from home. This suggests that the post-match recovery session should be different according to the level of the adversary team and the match location. The contents of this session should be adapted according to the PE match load declared by each player.

5. Conclusions

In summary, the study analyzed the effects of the level of the opponents and match location on the match PE declared by young elite soccer players. Soccer situational variables only affected the match PE of the players that almost completed the matches. Specifically, starter players declared higher match PE when they played against teams of different competition levels, as well as when the matches were played away. However, there were no significant differences in PE in the interaction of the two mentioned contextual factors for either starters or non-starters. Coaches should consider not only tactical–strategic needs, but also these contextual factors when managing the match playing time of the starter players. Moreover, due to the influence of the situational factors on the PE declared by starters, the coaches should take into account the level of the opponent and the match location when planning post-match recovery strategies, as well as for the distribution of the weekly training load.

Author Contributions: J.R.-G. and D.C. conceived the study. J.R.-G. and D.C. designed the study. J.R.-G. collected data. D.C. analyzed and interpreted the data. J.R.-G., D.C., J.Y., and A.L.A. drafted the manuscript, revised the manuscript, and approved the final version. All authors have read and agreed to the published version of the manuscript.

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Differences in Psychoneuroendocrine Stress Responses of High-Level Swimmers Depending on Autocratic and Democratic Coaching Style

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Abstract: The aim of the present study was to analyse differences in the psychoneuroendocrine stress responses of high-level, young swimmers depending on whether an autocratic and democratic coaching style was applied. Modifications in cortisol and the motivational climate of 18 young swimmers (15.3 ± 1.86 years, 10 females and 8 males) were analysed before and after two training sessions with equivalent training loads but directed by two coaches applying different approaches, i.e., autocratic (A) and democratic (D). The basal testosterone levels of the coaches were also assessed. The basal testosterone concentration was higher in coach A than in coach D; the athletes perceived them as autocratic and democratic, respectively. Swimmers under coach A's instructions showed higher cortisol levels, suggesting higher cortisol production related to coaching style. Furthermore, differences in the motivational climate concerning ego (i.e., athletes comparing their ability with that of other athletes) were observed with coach A, whereas differences in motivational climate concerning the task (i.e., athletes comparing their ability with their own past performance) were observed with coach D. Cognitive variables showed negative perceptions affecting athletes' training experience and performance when they were screamed at or insulted by coach A. There were no gender or age differences in cortisol production or motivational climate. In conclusion, this study suggests that an autocratic coaching style modulates cortisol release in both genders, affecting young elite swimmers' motivational climate and training experience.

Keywords: coaching styles; cortisol; learning; motivational climate; performance

1. Introduction

In competitive sports, a coach plays an important role in the "well-being" and performance of athletes. Dominant and autocratic coaching styles could be a source of stress among athletes, affecting mood states and glucocorticoid response [1]. However, some positive effects in the natural environment were also observed, such as uncertainty reduction or intragroup predictability improvement regarding

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social interactions, especially when the social hierarchy was clearly accepted [2]. Even so, democratic coaching could improve and increase athlete self-confidence, resilience and performance. Coaches should be able to establish contingencies, maintain the intensity of the training process, provide motivation to learn, and offer adequate feedback to achieve better performance [3,4]. Prolonged exposure to aversive stimuli may inhibit the establishment of a balance (e.g., eliciting emotions of frustration, apprehension and anger), affecting athletes' adaptation process to competition [5].

Some coaches prefer to stimulate democratic and active participation in the training process (e.g., encouraging the cooperation and affective expression within the group to achieve their competitive objectives). Previous authors [6] have suggested that positive dialogue, reflexion and good peer affective social interaction promote better learning, a fact that could be an excellent means by which to reach better technical skills and performance. Previous authors also noted how coaches with different leadership styles could become involved in the training of young athletes, i.e., by stopping training when athletes were facing health problems, modulating hormonal activity or showing a negative affective valence to the training load [7].

Competition outcomes and the perception of rivals as aversive increase cortisol production in athletes [8]. Cortisol activates body metabolism to regulate the athlete's adaptation to the environmental demands, stimulating gluconeogenesis and mobilising glucose towards skeletal muscle. Cortisol is also an excellent biological marker of psychosocial stress and hypothalamus-pituitary-adrenal (HPA) axis activity, including experiences of social evaluative threat [9]. Negative moods, psychosocial pressure, face-to-face confrontations or defeat frustration have been related to momentary changes in the concentration of circulating cortisol as well [10].

In autocratic leadership and social status seeking, cortisol and testosterone jointly regulate dominance [9]. Coaches with high testosterone and low cortisol levels may be more likely to be dominant, especially when they have previously shown autocratic behaviours [11]. Thus, an autocratic coaching style would become aversive to athletes during training, leading to increased stress response and, over time, making it more difficult for athletes to approach training as an adaptive challenge [12]. Autocratic coaches and learning based on aggressive behaviour, yelling, negative feedback and training overpressure may play negative roles in young athletes' performance, being linked with discontent and hostility towards subordinates and other components of the group [13]. The aim of the present study was to analyse differences in the psychoneuroendocrine stress responses of high-level young swimmers depending on the implementation of an autocratic or a democratic coaching style. The initial hypothesis was that a democratic coaching style would produce a lower psychoneuroendocrine stress response than an autocratic coaching style.

2. Materials and Methods

2.1. Participants

Eighteen young, high-level swimmers participated in this study (10 females and 8 males), with the following characteristics: age = 15.33 ± 1.86, and 14.60 ± 2.01 years; BMI = 20.88 ± 1.74, and 21.60 ± 2.88 kg/m^2; years of experience = 7.50 ± 5.09, and 7.40 ± 3.75, for male and female candidates, respectively. Two male coaches (age = 30 and 26 years, BMI = 25.06 and 31.17 kg/m^2; with more than 6 years' experience in high-level competition and performance training programs) also participated in the study. Participants were above 85% in the national ranking; they held previous national titles and were preparing to get a podium in the Spanish Championship some weeks after the time of research.

Coach A was considered autocratic because he usually screamed and insulted the athletes when they did not train hard enough; he just paid attention to swimmers who were outstanding and did not care about the preferences of athletes. In contrast, coach D was considered democratic because he never screamed; he just intervened when the athlete was training poorly, and usually contemplated the preferences of athletes [5,11]. Young swimmers were directly asked how they perceived the training styles of coaches A and D, i.e., as autocratic or democratic (see Section 2.2).

2.2. Procedure

First, the procedure was explained to the swimmers' parents. Informed signed consent was collected according to Helsinki's Declaration for studies on under-aged participants. Afterwards, separate meetings were conducted to explain the sampling protocol to the participating coaches and swimmers. They were instructed not to drink, eat, or brush their teeth 30 min before sampling. Also, a medical examination to rule out any psychiatric or physical problems, endocrine diseases, use of the contraceptive pill, or consumption of drugs was performed. This procedure was approved by the Ethical Committee of Universidad de Malaga (Spain) with registration number: CEUMA-35-2018-H.

We analysed the psychoneuroendocrine stress response of swimmers in two training sessions: one directed by the autocratic coach and the other directed by the democratic coach. Prior to starting the training sessions, saliva samples were taken from each of the coaches to measure basal testosterone and cortisol levels (i.e., upon waking on a nonworking day) to determine the individual neuroendocrine basal profile of each coach. Saliva samples of swimmers were taken before and after the two training sessions, as well. The first training session was led by the autocratic coach (coach A). After 48 h of recovery without physical activity, the second training session was led by the democratic coach (coach D). Each training session consisted of 6000 m swimming at equivalent intensity levels, divided into three parts: A warm up, consisting of 2400 m swimming at light aerobic intensity; the principal training, consisting of 3000 m high aerobic intensity swimming with instructions and feedback from the coaches; and recovery, consisting of 600 m low aerobic intensity swimming and free swimming style.

Thirty minutes before each training session (before warm-up to reduce the influence of physical activity on neuroendocrine axes) and 30 min after finishing, saliva specimens were collected using Super•SAL™ devices (Oasis Diagnostics® Corporation, Vancouver, WA, USA). Pure•SAL™ devices, with a visual indicator, allowed confirmation that a homogeneous sample of 1–2 mL was obtained. Cortisol concentrations were determined before and after each training session for each swimmer. All participants' saliva samples were taken between 18:00 and 21:00 h, immediately frozen at −40 °C and stored in the laboratory of the University of Malaga. Samples were centrifuged for 15 min at 3000 rpm and immunoassayed using the Grifols Triturus® (Somagen Diagnostics Inc., Edmonton, AL, Canada) equipment and competitive enzyme immunoassay kits (Diametra, Milan, Italy) with inter-assay coefficients of variation of 10.6% and 8.2%, and 7.4% and 4.6%, sensitivity of 3.28 pg/mL and 0.5 ng/mL, and detection limits of 1000 pg/mL and 100 ng/mL, for testosterone and cortisol, respectively. The samples were immunoassayed twice.

In addition to the collection of saliva samples, psychological variables were measured. The motivational climate perceived factors were determined by the individual sports version questionnaire [14], which analysed the environment generated by coaches A and D. It consisted of 24 items, where 1 applies to "strongly agree" and 5 applies to "strongly disagree" (Cronbach coefficient fluctuated between 71 and 78). The perceived leadership questionnaire [15] about specific coaching styles (i.e., two factors: autocratic or democratic behaviour) with 14 items, where 1 meant "always" and 5 meant "never" (Cronbach coefficient was 79−93), was also applied.

The hormonal baseline (awakening on a resting day) concentrations of each coach to analyse their basal testosterone concentration values indicated that coach A showed higher concentrations of testosterone (150 pg/mL) than coach D (80 pg/mL). Coach A's concentration was considered high (close to 95%, characterised in serum [16]), whereas it was medium for coach D (close to 50%). Serum testosterone and salivary testosterone showed a very high correlation coefficient (i.e., higher than 92) in prior studies [17]. Cortisol concentrations were considered in a low range, i.e., 2.03 and 2.28 ng/mL for coaches A and D, respectively. This hormonal profile fitted with van der Meij et al.'s [11] study about the testosterone-dominance relationship in autocratic leaders.

2.3. Data Analyses

All statistical analyses were performed with the statistical package SPSS® 20.0 (IBM Co, Armonk, NY, USA). Normality was tested by the Shapiro-Wilk test showing the non-parametric distribution of hormonal levels. Hormonal levels were log-transformed to fulfil the homogeneity assumption (non-transformed data were depicted in the figure to facilitate comparison with prior studies), following the statistical procedure performed by Aguilar et al. [10]. Repeated measures ANOVA was used to analyse differences in cortisol levels over time, and the effect size was measured using coefficient η^2_p. A Wilcoxon rank paired test was performed to determine whether there were differences in athlete's hormonal response before and after each training session, and the effect size was measured using Cohen's *d* test. The Mann-Whitney U test was performed to analyse gender or age differences in psychological variables and self-report scales. Post hoc power effect was also performed to detect 1-β error probability. The level of significance was set at $p < 0.05$.

3. Results

Repeated measures ANOVA was performed to analyse swimmers' hormonal level differences over time. Before training, the cortisol levels in both training sessions were similar; however, we found significant differences in cortisol production and a high effect size when swimmers were trained by coach A ($F_{(1,16)} = 3298$, $p < 0.03$, $\eta^2_p = 0.18$). A Wilcoxon ranked paired test was also performed to analyse differences in cortisol concentrations after training. Salivary cortisol concentrations were higher after training with coach A ($Z = -2.17$, $p < 0.03$, Cohen's $d = 0.57$), compared with coach D. Differences between before and after training were also observed in cortisol under coach A's training ($Z = -1.97$, $p < 0.05$, Cohen's $d = 0.86$), showing a higher stress response. The results showed that swimmers under coach A's instructions had greater hormonal activity compared to under coach D (Figure 1). Short sample sizes usually were related to type-II error, but this study showed differences in cortisol and motivational climate with power effects (1-β error probability) over 0.90. The Mann-Whitney U test showed no differences by gender or age in hormonal response and self-determination.

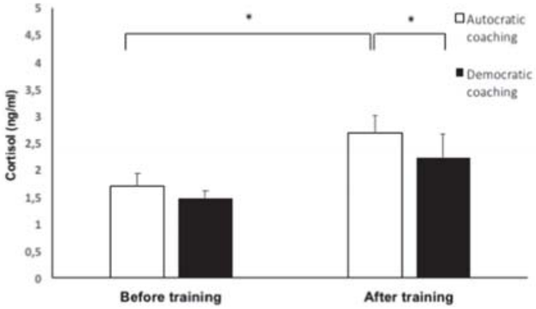

Figure 1. Mean ± SE salivary cortisol depending on coaching style. * $p < 0.05$.

Table 1 shows motivational climate and coaching style perceived in training days. Under coach A's instructions, athletes increased their motivation towards ego ($Z = -3.26$, $p < 0.001$, Cohen's $d = 1.69$). However, under coach D's instructions, swimmers increased their motivation towards the task ($Z = -3.42$, $p < 0.009$, Cohen's $d = 0.64$).

Table 1. Mean ± SD self-informed differences in perceived coaching styles and motivational climate in young swimmers on training days.

Self-Informed Differences inperceived Coaching Styles	Coach A	Coach D	*p*
Autocratic	15.25 ± 3.15	12.13 ± 3.44	0.001
Democratic	26.23 ± 6.94	33.13 ± 6.01	0.001
Motivational ego climate	43.06 ± 9.84	30.25 ± 4.28	0.01
Motivational task climate	39.81 ± 7.59	46.13 ± 5.60	0.009

Swimmers did not consider coach A to be a leader, and his coaching style was perceived as autocratic ($Z = -2.58$, $p < 0.01$, Cohen's $d = 1.69$). In contrast, swimmers perceived coach D as democratic ($Z = -2.14$, $p < 0.009$, Cohen's $d = 0.68$). Self-reports on athletes' experiences when they were trained by both coaches showed higher satisfaction when they were trained by coach D ($Z = -2.07$, $p < 0.038$, Cohen's $d = 0.94$), and they felt embarrassed when coach A shouted and insulted them while giving feedback in the training session ($Z = -2.16$, $p < 0.031$, Cohen's $d = 0.63$).

4. Discussion

The aim of the study was to analyse differences in the psychoneuroendocrine response patterns of high-level young swimmers upon exposure to autocratic and democratic coaching styles. The initial hypothesis was fulfilled, since the democratic coaching style produced a lower psychoneuroendocrine stress response than the autocratic coaching style.

When participants were under pressure (e.g., following coach A's negative feedback), cortisol production increased according to the self-determination theory [18]. The perceived behaviour of their coach affected the athletes' coping styles and well-being; therefore, aversive consequences could be associated with negative autocratic feedback as well. It is important to note that perceiving this activity as unpleasant has physiological, psychological and affective consequences on athlete performance and health [19]. In this line, previous studies have suggested that coach behaviour may influence athlete behaviour, increasing exhaustion, affecting self-confidence, and even resulting in the acceptance of cheating and deception to get their individual objectives [20]. One study with novice dancers suggested that an autocratic style could boost intrinsic motivation [21]. However, in this study, the aggressive, autocratic style was considered least satisfying and effective by the young swimmer sample [22]. Then, there is a need to find a fitting coaching style according to athlete needs.

The perception of being exposed to an autocratic coach showed increased cortisol levels in preactive swimmers before training. This anticipatory response has also been observed prior to competitions and evaluations, where the perception of threat, uncontrollability and uncertainty triggers a defensive response to prepare a person for any possible hazard [23–25]; however, further studies with a large sample size are needed to confirm this. Coach A induced an increase in cortisol concentrations and fostered a climate of "self-demotivation", negatively affecting the training experience of swimmers. These results are consistent with previous studies, where ego-motivation was associated with anxious states and a greater concern for performance [22]. In contrast, coach D was perceived by swimmers as a democratic coach, and his style of teaching fostered a climate of motivation towards the task and positive and personalised feedback. Similar results have been observed by previous studies [26], which pointed out that motivation towards the task at hand was related to the perception of receiving positive feedback and greater social support from the coach. The inclination towards the task, therefore, could depend on the positive perception that the athletes have regarding their own capacity, yielding better adaptive performance than a climate towards ego [27,28].

Training is a long-term process where the acquisition of new technical, physiological and psychological skills depends on the psychophysiological status of the athlete. The periodisation of these physiological and psychological skills in the training load, as well as a correct dosage of technical skill training, would allow the coach to assist in the best progression of the athletes [29,30]. In this line, it is important to highlight the need of a climate of trust and personal security to enhance desire and freedom to learn and communicate without fear of failure. Exchange with the environment causes the reconstruction of cognitive schemes, and failure is an inestimable source in this exchange, because it gives rise to the appearance of cognitive dissonances that cause the reconstruction of cognitive schemes [31]. Skills failure is a good option to learn new strategies and probe different motor skills to find other performance solutions. An environment focused on punishment increases the tendency to hide errors, making it more difficult to acquire new learning based on the search for effective individual solutions. A possible explanation of cortisol increases in the swimmers of the present study was stimuli associated with learning. Some reinforcement contingencies are associated with emotional response;

for example, reward extinction or omission is related to frustration [32]. When young athletes train hard, they expect positive feedback and verbal reinforcement from their coaches. Negative feedback and verbal reinforcements could be perceived as punishment, increasing cortisol production due to stress and frustration [10]. In learning contexts, negative feedback or insults did not help to improve feedback-based tasks, especially under psychosocial stress [33].

Swimmers enjoyed training more and perceived a greater personal effort when they were directed by a close and affective coach, which gave rise to a climate of motivation directed towards the task (Coach D). This subjective perception fits previous research works [34], where motivation was related to the task with a greater perception of enjoyment and satisfaction during training. The athletes considered that their performance was linked to a style of democratic teaching rather than an autocratic style. Previous authors [35] have suggested that the self-evaluation of improvement and competitive performance by objectives were more frequently observed when there was a climate of motivation towards the task. This perception of improvement and performance established by the affective-democratic feedback promoted by coach D and the climate towards the task firmly departed from the perception of failure related to each verbal warning received by coach A, which was related to an ego motivation climate, with negative feedback oriented towards punishment [36]; a tendency of coach A was consistently observed to promote a climate of motivation towards ego and, consequently, a probable impairment of individual self-evaluation, which should be confirmed in subsequent studies.

Limitation of the Study

This study presents some limitations. Firstly, it was very difficult to find high level athletes to be exposure at identical training with equivalent loads in the same days. Results must be considered as new knowledge to be confirmed or refuted in longitudinal research works. Second, hormonal samples were taken between 18:00 and 21:00 h when circadian cycles were decreasing; this must be taken into account to replicate this study in the future. Finally, a testosterone analysis of swimmers would provide us with a better compression of this complex response, as well as a performance analysis of each training session.

5. Conclusions

In conclusion, a direct relationship between autocratic coaching style and higher neuroendocrine activity was observed in young, high-level swimmers. It was confirmed by the momentary increase in circulating cortisol concentration before and after training when young swimmers were under coach A's instructions. Along with the momentary fluctuations of this hormone, there was a greater predominance of an ego-motivational climate, and consequently, a negative influence on related psychological factors (e.g., deterioration of training day experience) or affective factors (e.g., frustration linked to perceived verbal reprobation after failures). An autocratic coaching style could lead to a deterioration in the athlete–coach relationship, and could be unfavourable to the progress of young, promising swimmers over the medium term, affecting their neuroendocrine response patterns, self-confidence and motivational climate.

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Review

Applying Heart Rate Variability to Monitor Health and Performance in Tactical Personnel: A Narrative Review

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Abstract: Human performance optimization of tactical personnel requires accurate, meticulous, and effective monitoring of biological adaptations and systemic recovery. Due to an increased understanding of its importance and the commercial availability of assessment tools, the use of heart rate variability (HRV) to address this need is becoming more common in the tactical community. Measuring HRV is a non-invasive, practical method for objectively assessing a performer's readiness, workload, and recovery status; when combined with additional data sources and practitioner input, it provides an affordable and scalable solution for gaining actionable information to support the facilitation and maintenance of operational performance. This narrative review discusses the non-clinical use of HRV for assessing, monitoring, and interpreting autonomic nervous system resource availability, modulation, effectiveness, and efficiency in tactical populations. Broadly, HRV metrics represent a complex series of interactions resulting from internal and external stimuli; therefore, a general overview of HRV applications in tactical personnel is discussed, including the influence of occupational specific demands, interactions between cognitive and physical domains, and recommendations on implementing HRV for training and recovery insights into critical health and performance outcomes.

Keywords: heart rate variability; HRV; autonomic nervous system; military; human performance; physiological monitoring

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1. Introduction

The health, well-being, and preparedness of tactical personnel are crucial to mission success [1]. In tactical settings, Human Performance Optimization (HPO) programs focus on enhancing and sustaining the individual's ability to perform occupational tasks over time. There are several strategic focus areas that contribute to HPO of tactical personnel, to include physical fitness, health, nutrition, cognitive, and psychosocial. Inherently, military training and operations have remarkably high physical and mental demands that necessitate exceptional levels of fitness [2,3]. To achieve such extreme levels of prowess, bouts of higher training intensities and volume overload are required [4], subsequently allowing the operator to develop hypersensitivity to chronic fatigue and overtraining [5]. However, incomplete recovery from training may also lead to chronic fatigue, which inevitably hinders operator preparedness [6]. Fortunately, scientifically validated approaches in sport and exercise sciences provide the basic concepts and principles for applications of monitoring training loads in tactical settings [7].

The need for accurate, meticulous, and effective monitoring of biological adaptations and systemic recovery is certainly critical for all domains under HPO [8]. In addition, the practice of self-monitoring affords the individual operator a platform for personal insights and real-time biofeedback [9,10], enhancing the opportunity to facilitate in/exteroception capacity and regulation capability [11], implement necessary training modifications or lifestyle adjustments, and mitigate unwanted adaptations (e.g., diminished power output, negative mood states, increased sympathetic tone, skill/knowledge decay) [12]. Monitoring training loads (TL), which can be segmented into external and internal training loads, can be used to manage cumulative TL on an individual and group level [13]. External loads, typically measured by global positioning systems, accelerometers, transducers, or gyroscopes, represent physical work being incurred by the individual, such as, total distance covered, high impacts, bouts of accelerations and decelerations, repetitions, sets, and intensities of resistance training [13,14]. Meanwhile, the internal loads represent the physiological costs incurred by exposure and production of forces during external loads. Internal loads may be monitored objectively through laboratory testing (e.g., blood, blood lactate, saliva, hormone, and metabolic measure) and wearable portable physiological measuring devices (e.g., heart rate monitor), while subjective measures can be monitored by use of rating scales (e.g., rate of perceived exertion (RPE), fatigue scale, sleep quality, etc.) and subjective questionnaires (e.g., Pittsburgh Sleep Quality Index) [7,13–17]. Individuals respond differently to given external loading for numerous reasons (e.g., current fitness levels, genetics, sleep hygiene), and thus it is particularly important to monitor internal loads. In tactical populations, global positioning systems and accelerometers have been used to quantify external loads while laboratory testing, wearable physiological sensors, and subjective measures have been commonly used to assess internal loads [14].

In HPO programs, the utilization of heart rate variability (HRV) provides a non-invasive, practical method for assessing an individual's response to internal load [5,18]. The ability of HRV to afford immediate insight on individual adaptability to environmental demands (i.e., training load) [5,19–22] is particularly important for the strenuous nature of military training [14]. Without proper monitoring and reactive/proactive programming, intense training cycles can lead to maladaptive outcomes (e.g., overtraining or injury) thereby compromising preparedness for occupational tasks [2,23–25]. Ultimately, the use of HRV provides insights on the neurocardiac function of the autonomic nervous system (ANS) [22,26], permitting inferences into overall performer preparedness/workload status and resource availability/efficiency [15,22,27,28]. To ascertain the value of employing HRV in HPO programs, the framework and context pertaining to ANS architecture must be understood. Although literature exists regarding the ANS and HRV methods, limited research has employed HRV in tactical populations. A meta-analysis performed by Tomes and colleagues [29] concluded that HRV was an effective tool for measuring health and performance in tactical personnel. However, despite these findings, literature is lacking that describes various field applications and actionability of HRV data on performance recovery and sustainment. Further, due to the innate uniqueness, there may be special considerations for employing HRV monitoring in tactical settings, which require further investigation. Thus, the purpose of this narrative review is to (1) review the underlying foundations behind the use and limitations of HRV, (2) identify some of the appropriate applications of HRV metrics in tactical settings, and (3) explore the relationship between HRV and stress, occupational performance, cognitive performance, and recovery in tactical environments.

2. Autonomic Nervous System (ANS)

The ANS influences body functions by regulating systemic resource allocation to maintain homeostasis and accomplish goal specific actions in the face of fluctuating internal and external environmental stimuli [30,31]. Practically, it moderates physiological and psychological preparations for and recovery from acute and chronic workloads. This regulatory system's stability and responsiveness is critical for meeting the unpredictable and strenuous occupational demands imposed on military personnel [32]. Moreover, many tactical performance constructs can be directly linked to the efficiency and effectiveness of ANS-modulated systemic responses, such as: endocrine profiles supportive of passing intense selection events [6,24,25]; plasticity to adapt with progressive training overload across critical functional areas (e.g., cardiovascular, musculoskeletal, cognitive) in the development and maintenance of peak operational capabilities [2,3,23,33]; emotional awareness and regulation to sustain before, during, and after a firefight [11,34]; motor control resiliency required to execute mission essential tasks [35]; and the overall system's ability to recover, repair, and improve from various stimuli [32]. Collectively, the ANS is implicated in nearly every aspect of human behavior, with innate links to health and performance [36]. The direct application to high stress, high-stakes vocations only strengthens this connection and elevates the importance of understanding the functional physiology involved.

The primary ANS (excluding enteric) is comprised of the sympathetic nervous system (SNS) and parasympathetic nervous system (PNS), which are associated with arousal (often perceived as stress) or relaxation, respectively [5,37,38]. Variations in cardiac dynamics (e.g., HRV) are the result of intricate interactions between the SNS and PNS [5,38]. The ANS's influence on the cardiovascular system is regulated by a complex network of brain centers (e.g., limbic system, hypothalamus, and medulla) and neural communication pathways (e.g., brainstem and spinal cord) [11]. The medulla integrates sensory input and regulates parasympathetic outflow to help control cardiac modulation in response to arousal, stress, and task demands [5,11,31,38,39]. Primarily, the ANS controls cardiac function through its innervation of the sino-atrial (SA) node (i.e., heart's pacemaker) and atrioventricular (AV) node via the right and left vagus nerves, respectively [5,39–41]. Resultantly, the ANS regulates heart rate, cardiac timing, contractility, and conduction velocity (i.e., mechanisms that facilitate resource delivery). The sympathetic and parasympathetic divisions are often complementary, whereas the SNS generally increases heart rate and vasoconstriction, the PNS generally decreases heart rate and causes vasodilation (Figure 1) [5,38,39]. During fight or flight environmental demands, the need to deliver metabolic substrate for movement overrides the "resting state" variability induced by SA-node conduction delay [42]. However, the SNS and PNS do not always increase and decrease activation in a reciprocal manner, especially during high-stress skill execution [5,35].

Since the ANS is intimately connected with other psychological and physiological systems throughout the body, quantifying responsiveness can provide vital information regarding functional occupational performance capacity or adaptions during/after exercise and training iterations [38]. This is particularly important to understand in tactical personnel, who are frequently or unpredictably exposed to high acute stressors, bouts of intensive physical activity, extreme environments, and varying occupational schedules (i.e., rotating night shifts, multi-day combative engagements), which may directly influence the cardiovascular system [43]. A schematic diagram of SNS and PNS components for various physiological systems is displayed in Figure 1. The acceleratory responses from the SNS, including elevated heart rates, muscular perfusion, bronchial dilation, and pupil dilation are useful for preparing the body's response to situations requiring physical prowess and precise perceptual motor coupling, which are often noted in tactical settings. Chronically elevated SNS responses can have negative consequences, requiring PNS activation or SNS deactivation to ensure adequate recovery from high intensity events and promote overall health and well-being (i.e., reducing symptoms of burnout, post-traumatic stress disorder, or depression) [2,6,32]. Further, sympathetic responses can impede activity and efficiency in specific brain regions, such as the pre-frontal cortex, which is responsible for higher level executive functions, and the motor cortices [34,35,44]. Inhibition of the pre-frontal cortex can negatively affect attention, emotion, and decision-making, while further impeding the fine motor skills needed for occupational tasks of tactical personnel [11,12,34,44,45]. In tactical populations, measuring and monitoring ANS responses, such as HRV, will likely be useful for informing skill efficiency and expertise, individualized training programs, occurrence or likelihood of injury or illness, risks of overtraining, training and recovery statuses, and operational preparedness. An individual's HRV may be described through numerous calculations of raw heart rate data signals, which are deconstructed to quantify the underlying signal components, resonant frequency bands, and glean more value, greater inference, and actionability in the underlying data.

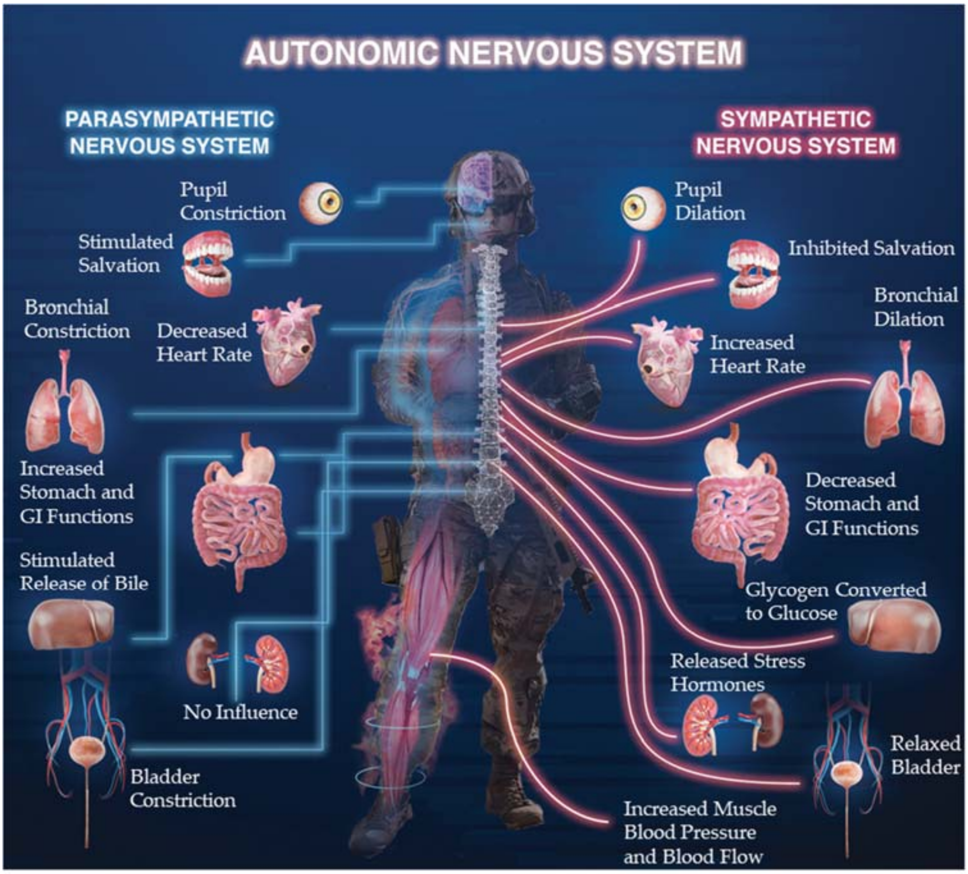

Figure 1. Example layout of the relations between various organs throughout the body and the Autonomic Nervous System.

3. Defining Heart Rate Variability (HRV) and Respective Metrics

The term HRV describes a series of mathematical calculations for modeling psychophysiological responses based on the time between consecutive heart beats, also known as beat-to-beat or inter-beat-intervals (IBI) [5,20,26,46–49]. IBI is the duration between R peaks from consecutive QRS complexes (i.e., ventricular depolarization) across the duration of a contextually relevant time series (i.e., NN or R-R) [5,50–52]. The IBI is typically measured with an electrocardiography (ECG) signal with data sampling frequencies from 250–1000 Hz. Higher sampling frequencies offer more robust data, which is especially important for capturing HRV metrics during dynamic movements (e.g., exercise, room-clearing) and complex cognitive processes (e.g., event based decision making) [5,30,38,50]. The stress (e.g., physical activity, psychological arousal) induced sympathetic activation and parasympathetic (vagal) withdrawal causes the R-R intervals to become shorter, more rapid, and less varied, resulting in a global decrease across most HRV metrics [5,20,22,53]. Generally, sympathetic activation increases HR and decreases IBI variability, while parasympathetic withdrawal decreases HR and increases IBI variability. Typically, PNS modulation occurs rapidly (e.g., <1 s) and possesses a short-lived response (<5–10 s), while the SNS's response is slower (~5 s from stimulation onset to IBI modulation) and persists for longer durations (~10–30 s) [41]. Measuring and analyzing IBI variations across time via HRV metrics can provide quantitative insights on the culmination of psychophysiological responses and further detail a performer's ability to handle internal and external environmental demands (i.e., recovery) [22,53].

With respect to training loads and recovery, the most frequently implemented and practically useful HRV parameters are analyzed using time- and frequency-domain methods [5,20,26]. These commonly used methods represent various ways to view the central tendency (i.e., mean, median, mode), variability, and distribution (i.e., standard deviation) of heart rate over time [5]. HRV parameters consider the average values and overall magnitudes of fluctuations to quantify control of heart rate over time. However, two individuals may have the same average R-R interval and heart rate (HR) responses to an event, but vastly different variability of R-R intervals (see Figure 2 for graphic example). The two most commonly accessible/used time-domain parameters are the standard deviation of the N-N intervals (SDNN) and the root mean square of the differences in adjacent N-N intervals (rMSSD). The SDNN represents a coarse quantification of HRV via autonomic regulation from sympathetic and parasympathetic inputs [21,54], while rMSSD represents parasympathetic activity [20,21,27,54,55]. Unlike SDNN, rMSSD is void of HR slow-wave components, resulting in minimal respiratory influence and a more accurate representation of parasympathetic activity [46,54].

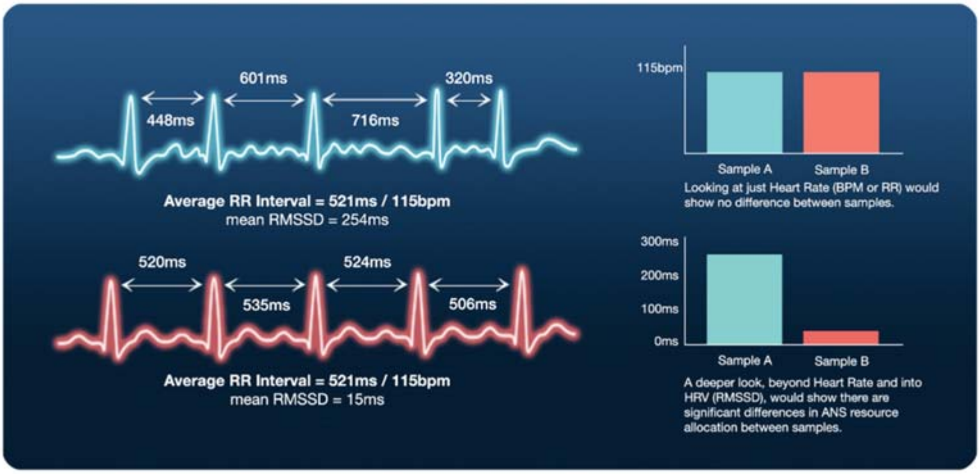

Figure 2. Example of R-R intervals in milliseconds (duration between R peaks from consecutive QRS complexes). Trace 2 (lower, in red) shows little/negligible heart rate variability (HRV). Trace 1 (upper, in blue) shows significant HRV. Data table on right exemplifies importance: average heart rate (HR as bpm) is identical, but when the underlying signal is teased apart, massive differences in root mean squared of successive differences (RMSSD ~ 17 fold) and can be used to infer significant differences in autonomic nervous system (ANS) resource allocation between the same samples.

Frequency domain metrics use mathematical transformations of R-R data to quantify the modulation of a given signal at specific frequency ranges (e.g., power spectral density bands): ultra-low frequency (ULF), very low frequency (VLF), low frequency (LF), and high frequency (HF) [26]. Compared to the time domain, frequency domain HRV metrics tend to offer more detail regarding the underlying signal, its component-based contributions, and their relationship to behavioral outcomes. In clinical settings, ULF ($\leq$0.003 Hz) and VLF (0.0033–0.04 Hz) are often used during long-term recordings ($\geq$24-h) [5,26]. However, athletic and tactical settings may not permit long recordings, thus, typically rely on more biologically relevant and accurate metrics during short-term recordings ($\leq$10-min), such as LF (0.04–0.15 Hz) and HF (0.15–0.4 Hz) [5,56]. The HRV frequency domain metrics represent specific ANS branches [5,26,38]. The LF power is influenced by both SNS and PNS as well as baroreceptors (i.e., blood pressure control) [5,26]. In resting conditions (e.g., sleep), LF reflects the baroreflex, not cardiac sympathetic innervation [26,57], and may not accurately reflect HRV responses to training load and recovery. The HF power band directly reflects parasympathetic activity and can be influenced by the respiratory cycle [26,38] and cognitive workload demands (e.g., emotion, executive functioning, motor control) [11,58,59]. Low (and decreasing) HF is often associated with parasympathetic resource consumption (e.g., mental/physical task), distress (e.g., anxiety, worry), or poor resource efficiency (e.g., non-expertise, lack of recovery), and commonly leads to impaired physical or cognitive performance [23,26,60]. Insights on sleep quality, regeneration, and fatigue can also be obtained through analyzing frequency domain metrics during nocturnal HRV assessments [61,62].

The ratio of LF to HF is often used to represent sympathetic-parasympathetic balance [26,63,64]. This incorrectly assumes that LF solely reflects sympathetic activity, despite being produced by both the SNS and PNS [26,64]. Given those assumptions, a low LF/HF ratio would indicate parasympathetic dominance and a high LF/HF would indicate sympathetic dominance [26]. However, these relations may be influenced by body position, as LF is primarily produced by SNS in a supine position and PNS in a seated position [26,64]. These intricacies make it more difficult to use the LF/HF ratios in assessing training load and recovery [26]. Total power (TP) refers to the sum of all power spectral density components (ULF, VLF, LF, and HF), which reflects the total variance in heart rate pattern over recording durations [21,26,57,61,65–68]. Due to their reflection of general autonomic and parasympathetic activity, TP and HF are effective frequency-domain metrics for interpreting performance efficiency, training adaptation, and recovery/resiliency in the presence of physiological and psychological loads [5,26].

Consequently, in a resting state, increases in HF and TP can indicate desired training adaptations and complete systemic recovery, whereas a decrease in HF and TP typically indicates unwanted training effects, maladaptation, and incomplete recovery [26,34,35]. Mathematically, the modulation of HF to TP (HP/TP) may index the proportion of parasympathetic activity within the entire ANS frequency-domain spectrum. Overall, many HRV metrics exist (Table 1) and represent specific ANS domains, but their applicability and accuracy in specific settings need to be considered prior to their use. Typically, the three variables often considered the most meaningful and actionable are rMSSD, HF, and TP. The normative values for each of these components are dependent on many factors, such as age, gender, fitness, and various demographics. Therefore, context is required when drawing conclusions based on these HRV metrics. For all of these reasons, HRV metrics should be utilized as within-individual changes. Comparing one individual’s HF score to another’s would not provide any personalized insights. Overall, these HRV metrics will initially decrease under persistent conditions of high stress environment, poor sleep hygiene, poor nutrition, inadequate exercise, durations of isolation or harmful relationships. On the contrary, higher HRV values are often associated with positive health states and functionally beneficial adaptations, including cardiovascular fitness and resiliency to training loads or other various stressors.

Table 1. Definitions basic time and frequency domain heart rate variability metrics [27,69].

Time Domains in Short-Term Recordings			
Index	**Definition**	**Interpretation**	**Correlates**
SDNN (ms)	Standard deviation of all R-R intervals	Global quantification of HRV	Total Power
rMSSD (ms)	Root-mean square of successive differences between R-R intervals in a specified time segment	Vagal tone	High Frequency, Parasympathetic activity

Table 1. *Cont.*

Basic Frequency Domains			
Index	**Definition**	**Interpretation**	**Correlates**
VLF (ms^2)	Power in the very-low frequency range (<0.04 Hz)	Hormonal factors and peripheral thermoregulation origination	Parasympathetic activity
LF (ms^2)	Power in the low frequency range (0.04–0.15 Hz)	Baroreflex, arousal	Sympathetic activity, Parasympathetic activity
HF (ms^2)	Power in the high frequency range (0.15–0.4 Hz)	Cardiopulmonary reflex, cognitive regulatory state, dependent on resource availability and interpretation of environmental demands	Parasympathetic activity
LF/HF	Low frequency/high frequency ratio	Sympathetic-parasympathetic balance (assuming known LF)	Sympathetic activity, Parasympathetic activity
Total Power	Total power in the entire frequency range (<0.4 Hz)	General autonomic resource allocation	Sympathetic activity. Parasympathetic activity
HF/Total Power	High frequency/total power ratio	Proportion of parasympathetic to total autonomic resources	Parasympathetic activity

SDNN, standard deviation of the N-N intervals; RMSDD, root mean square of the differences in adjacent N-N intervals; ULF, ultra-low frequency; VLF, very low frequency; LF, low frequency; HF, high frequency.

4. Implementation and Analysis of Heart Rate Variability (HRV) Measures

4.1. How to Measure HRV

Field based HRV monitoring is considered user friendly, objective, and a particularly reliable tool, which can be easily used to manage training loads and facilitate proper recovery [5,53]. Additionally, HRV assessment and monitoring tools have become more affordable and practical to use [26,27]. With a continuously growing number of commercial devices capable of measuring and reporting HRV, it is first important to understand device accuracy [70], then equally necessary to understand their compatibility for use in the field with tactical populations. For example, the placement of the sensors, wiring requirements, duration of battery life, internal memory capacities, device durability (i.e., handling weather conditions and impacts), and screen displays (i.e., ability to be shut off on night missions) should all be considered when selecting devices. Detailed information on wearable sensors capable of measuring HRV in military settings can be found in a recent review article [71], which highlights the pros and cons of several devices. Overall, practical, commercially available wearable sensors tend to lack adequate battery life and internal memory for long duration recordings (24 h cycles), while clinical devices (3-12 lead electrocardiography) provide the most accurate results for long and short durations, particularly at rest [72]. Despite better signal to noise ratios, clinical electrocardiography (ECG) devices may still be susceptible to motion artifact or electrode impedance and lack practicality due to their bulkiness and wiring requirements, making them less suitable for tactical environments.

Practical devices, such as chest straps, clothing garments, and wrist or finger worn photoplethysmography (PPG), are unobtrusive, but their validity and reliability can be significantly impacted by the fit of the device, which should be "snug" to avoid missing data from disconnections [73]. The accuracy of PPG devices is also subject to motion artifact, skin pigment, tattoos, temperature, and pressure placed on the sensor [73]. At rest, certain PPG devices may be accurate [70] but reductions in accuracy are noted as intensity of exercise or movement variability increases [74]. Thus, PPG devices may be useful during sleep/resting stages or mild, continuous exercise, but may not accurately capture HRV during the intense events common to tactical populations. Yet, these devices prove useful for their ability to be conveniently worn on the wrist or finger, with little interruption to daily living, for overnight, upon awakening, or 24 h measurements. However, the utility of these devices in tactical settings requires further exploration. Chest strap devices (e.g., Polar

H10) are typically utilized in a physical training application but can also accurately and reliably be used for HRV assessments [75]. We suggest that chest strap devices are most appropriate during tactically relevant physical maneuvers, while the specific manufacturer to use is subject to their level of accuracy during the intended testing events.

4.2. When to Measure HRV

A measurement's purpose dictates the appropriate schedule of data collection time-points (e.g., time of day, duration, and frequency) [5]. Monitoring HRV may involve periodic resting measures and pre- and post-training assessments across key timepoints to capture autonomic resource distribution, consumption, and restoration before, during, and following specific task demands. Following training, the time required for HRV to return near baseline values may demonstrate the relative ability to recover from training sessions of various intensities and volumes. However, to fully understand the demands of specific occupational tasks (i.e., clearing a house in military or law enforcement) it is important to include HRV monitoring during the task, in addition to baseline and post-event measures. Since time of day, similar to body position, will significantly influence HRV [38,76], it should be matched across repetitive assessments, where able. Otherwise, comparisons across time-points will be difficult to interpret (i.e., unsure if change is due to training loads or fluctuations in diurnal variation). For example, HF increases at night and decreases during the day, emphasizing the importance of both nocturnal and resting awake readings [26,38]. Figure 3 offers a graphical depiction of the opportunities for HRV measurements throughout the day along with context considerations at each time point.

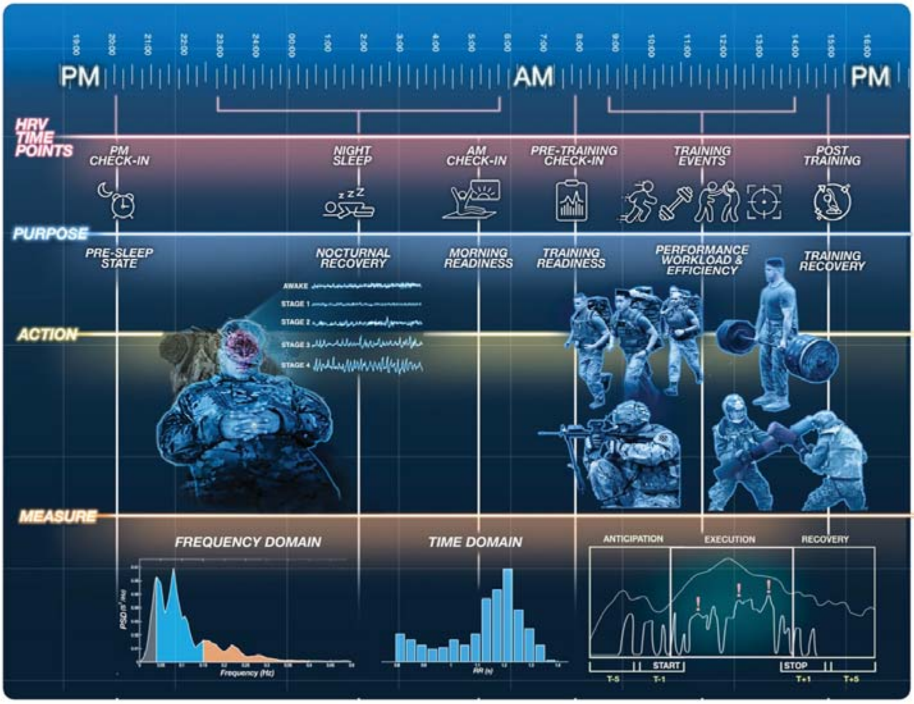

Figure 3. Opportunities within tactical populations for heart rate variability (HRV) collection throughout the day. Pre-sleep HRV assessments inform end of day status, daily resource consumption, and can quantify daily workload. During sleep, HRV provides insight on nocturnal recovery, homeostatic restoration, and daily readiness. Pre- and post-training HRV spot checks may inform acute preparedness for and recovery from training. During events, HRV can provide insights on performance workload and efficiency.

4.2.1. Establishing Baseline HRV Measures

Establishing a resting state, baseline, HRV is important for assessing overall ANS modulation and cardiac vagal tone. Following the principles of resource theory, baseline HRV identifies the pool of available ANS resources affecting the cardiac cycle [77]. The volume of ANS resources utilized while preparing for, completing, and recovering from psychophysiological demands is related to the imposed workload and functional efficiency to accomplish the task [78]. Comparing the HRV during tasks to HRV before and after tasks can be accomplished through normalization, difference scores, and percentage values for indices of training and recovery loads, respectively. When collecting baseline or resting HRV measures, the supine position is recommended, although sitting or standing may be warranted in field exercises (keeping in mind that methodological consistency is critical) [5,79]. Lastly, confounding factors from external (e.g., temperature, noise, and lighting) and internal (e.g., physical and emotional behaviors leading up to testing) need to be controlled and kept consistent as they will affect HRV and the ability to record "true" baseline values.

To establish an individual baseline for consistent daily and actionable assessments, HRV measurements should be recorded over a minimum 1–2 week period [46], before making any adjustments to training load or recovery. The utilization of within-individual HRV metric changes are further improved by quantifying rolling 28-day averages [80] and assessing the individual's deviation from this average. Using the 28-day rolling average, an individual's change in status is identified as the difference between daily mean (or specific event) HRV and the prior 28-day average HRV. Typically, raw or standard scores (Z-score, percentage change) are compared to rolling average measures. With this approach, acute stress or incomplete recovery states may be denoted by reduced rMSSD as compared to that individual's 28-day average, as shown in Figure 4 below where Z-score values are less than zero. Conversely, completed recovery could be indicated by maintained or increased HRV [81] shown in Figure 4 where Z-score vales are greater than zero. However, it should be noted that excessive lengths of incomplete recovery can result in an exaggerated increase in parasympathetic modulation, as an attempt to force systemic recuperation efforts [82,83].

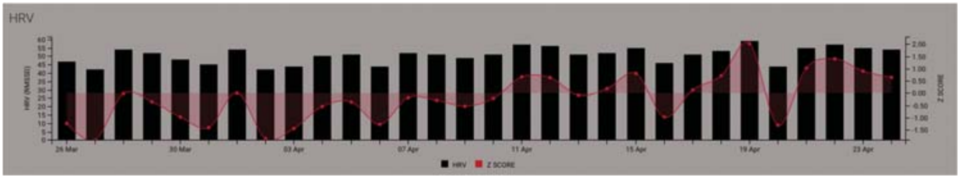

Figure 4. Example of daily heart rate variability (HRV) monitoring of a single individual using a rolling 28-day average to calculate daily Z-scores. Reduced root mean square of the differences in adjacent N-N intervals (rMSSD) and the respective Z-score values below 0 may suggest incomplete recovery, while Z-score values at or above 0 may suggest complete recovery status.

4.2.2. Nocturnal and Morning HRV Measures

Nocturnal and morning HRV indicate different psychophysiological processes; however, both inform recovery and readiness status. In long-term monitoring, HRV quantification represents the amount of recovery in relation to average baseline HRV pre-awakening (i.e., during sleep) [84]. Nocturnal HRV, although not completely understood, is commonly used to assess sleep quality, resource recovery, and systemic adaptations [85]. Moreover, assessments pre/post sleep onset and pre/post awakening provide insights to the systemic resources leftover from yesterday's workload and the subsequent night's recovery, without and with conscious cognitive processes involved [85]. As stated previously, HRV measurements must be done in a consistent fashion for proper utilization for training load and recovery monitoring. Whether an end user chooses nocturnal or post wake measures, they should be done consistently.

4.2.3. Pre-, During-, and Post-Event HRV Measures

Measuring HRV pre-, intra-, and post-task (e.g., physical or cognitive) may indicate autonomic resources roused in the anticipation of completing a task, the resources required to execute said task, and the rate of resource recovery following completion [35]. HRV responses to events, including training simulations, exercise, or real-world applications, may require a resting HRV measurement 5–10 min prior and a recovery HRV measurement 5–10 min after the event (or at least a known baseline value for difference score calculations). Depending on the circumstances, the first 5 min of a 10 min resting assessment may be discarded as part of an acclimation phase, while the remaining 5 min more accurately represents baseline or recovery HRV. Regardless, the timing of pre- and post- event measures should be similar (i.e., resting measurement 5 min before = resting measurement 5 min after). Furthermore, additional timepoints (e.g., 5–10-, 10–15-, and 15–20 min post-event) may provide further information on the time-course of recovery following the event. The same original temporal duration (e.g., 5-min) should be used for all time blocks and may require parsing into multiple sample windows. Body position (e.g., supine, seated, standing) should also be the same for any pre- and post-measurements.

The modulation of HRV measures can be used to assess various tasks, such as the observe-orient-decide-act cycle of an advanced marksmanship/close quarter battle (CQB) assessment or intense training session, but require more precise time stamping and data parsing compared to pre- and post-task measures [35]. Further, during longer duration sessions, behavioral events (e.g., start/stop, breach, contact, endex, exfil) should be used to determine the context-dependent time windows of specific activities being investigated. Delta scores among time periods and rest/baseline represent the autonomic modulation allocated to process changes in environmental states and accomplish relevant behavioral responses. For example, the difference between baseline and pre-task HRV provides insights to biological priming and arousal/anxiety management, while the difference between baseline to during-task HRV indicates the required ANS resources to achieve the cognitive/motor performances [34,69,86]. As a novice develops expertise, the shift toward automaticity of underlying control processes frees up systemic resources, including HRV [87]. Thus, the aforementioned use of HRV may inform the tasks' relative difficulty or unfamiliarity, as well as the individual's level of efficiency and resiliency (i.e., ease of task completion and ability to quickly recover between tasks). For example, smaller changes in HF HRV (i.e., parasympathetic tone), compared to baseline, would suggest greater autonomic resource efficiency and heightened resiliency [11,35,88,89]. This type of HRV perception-action coupling analysis provides a psychophysiological measure of resource allocation, which permits the comparison of performance effectiveness (e.g., resulting outcomes) with resource efficiency (e.g., HRV modulation from baseline) to determine skill automaticity and expertise. Additionally, it enables the tracking of individual progression over time.

4.2.4. Recording 24 h HRV

Continuous measurements over a 24 h period may represent HRV responses to all the various stimuli encountered during typical, daily occupational endeavors. In daily HRV, values below an individual's normative rolling baseline could indicate incomplete recovery from high training volume, high training intensity, onset of illness, chronic stress, or poor sleep [84]. However, more research is necessary to understand the complexities of daily HRV, which can fluctuate greatly due to diurnal variations in circadian sleep patterns/rhythms, body temperature, metabolism, activity [26]. Still, by encapsulating 24 h HRV recordings a more holistic measure of HRV is permitted. The circadian rhythms may best be accounted for by segmenting the HRV data into daily events, such as nocturnal, morning, training, and occupational events. Nonetheless, a single short duration value should not be used interchangeably with 24 h recordings. The 24 h recordings have been shown to be more reliable than 5 min resting measures, which led some to suggest the use of 24 h recordings for intervention studies [26]. Using the 24 h recording may also allow the quantification of overall ANS responses to a range of stimuli, providing more granularity into wholesome psychophysiological states. However, the execution of collecting 24 h HRV data also requires more buy-in from individuals as they will be tasked with wearing the sensor constantly and ensuring adequate battery charging. Thus, it is necessary to consider whether a method will result in high attrition rates, since large quantities of missing data will not permit reliable, actionable inferences.

5. Monitoring Heart Rate Variability (HRV) in Tactical Populations

5.1. Relations between HRV and Stress

Stressors experienced by tactical personnel occur in many forms, such as; physical training, deployments, vigilance, emotions, cognitive strain, environmental conditions, lives at stake, low margins for error, and illness [34]. The high-risk nature of operational demands in military environments, particularly special forces [7,90], influence psychophysiological responses of the ANS to environmental conditions [5,31,38,39]. The acute manifestation and chronic accumulation of stress reduces HRV from baseline, which influences performances on physical and mental tasks [34,35,91]. Although maintaining HRV under strenuous loads may indicate preparedness for the task, acute stress responses of reduced HRV in preparation for an event may not necessarily be maladaptive, especially in high performance scenarios that naturally trigger epinephrine responses to improve performance outcomes [91]. Psychological stress, both acute and chronic, usually results in parasympathetic withdrawal, represented by sympathetic dominance or a decrease in HRV (HF and total power) [21]. This response originates from the brain's perception of the threat or engagement in task workload and is delivered via reduced stimulation of the vagus nerve [92,93]. For example, operators with a high HRV (within and between individuals) are usually better at coping under stressful conditions, expressing greater positive emotions, and performing cognitive tasks more accurately and quickly, whereas operators with low HRV tend to have poor self-regulatory capacity and exhibit by more rigidity and hypervigilance during task responses [44].

5.2. Relations between HRV and Physical Occupational Performance and Training Loads

Studies investigating the effects of physical activity on HRV have exponentially increased in recent years [5,16,38,39,94]. In such models, HRV is typically used to detect overtraining or non-functional overreaching (NFOR), which may be detrimental to overall performance and injury risk [5,21,91], as well as stress and recovery [27,46]. Signs of overtraining and NFOR are primarily associated with long-term sympathetic activity (increased LF and suppressed HF) [5,75], however studies also associate chronic overtraining with long-term parasympathetic dominance [5]. Consistent abnormally high or low HRV values may indicate overtraining and suggest a required reduction in training volume (sets x reps); whereas, a reasonably elevated HRV, representing parasympathetic dominance, may indicate a detraining effect and a need for increased training loads [5]. Previous studies reported associations between cardiac autonomic function, as measured by HRV, and aerobic fitness, external training loads, and anabolic hormone concentrations [2,16,24]. Yet, direct comparisons among individuals requires controlling for confounding variables, such as age, gender, and fitness-levels, by comparing individuals to themselves over time or surrogate groups [5,26].

Conventionally, HRV is mainly directed at capturing internal cardiovascular strain resulting from (or required to accomplish) a given external training load. However, for the tactical athlete, quantifying responses to external workloads and cardiovascular homeostatic balance is only one of many actionable insights HRV can afford. Theoretically, running faster and lifting heavier require more resources through PNS withdrawal and SNS stimulation, which influences cardiac output to become more forceful, rapid, and less variable. The scientific literature supports the link between HRV metrics, aerobic adaptations, and general fitness levels [95–97], although these relationships may be dictated by the individual's characteristics and training intensities and volumes. HRV may be useful for understanding the current state of aerobic fitness and monitoring aerobic training adaptations. However, it is important to note that the HRV is mainly a marker of neurocardiac ANS and its direct relation to neuromuscular performances of maximal strength and power output are unclear [97]. Training programs which base workload adjustments on HRV feedback have demonstrated greater increases in countermovement jump and sprinting abilities [98]. Therefore, despite a lack of direct associations between HRV and neuromuscular capabilities, HRV monitoring may improve training program design by prescribing adequate recovery and training volumes. Thus, it is recommended that HRV monitoring is used in conjunction with other monitoring tools including external and internal loads, as well as measures of neuromuscular capacities such as those described in prior literature reviews [99].

5.3. Relations between HRV and Cognitive/Motor Skill Performance

Mechanistic underpinnings [11,58,100–104] and applied experimental studies [33,35,69,89,105–110] support the use of HRV for assessing, predicting, monitoring, and modulating the cognitive performance domain [59]. Various neural networks obey similar resource theory dynamics and modulate the sympathetic "gas pedal" and parasympathetic "brake." The prefrontal cortex (PFC) and amygdala, two areas that contribute to neurophysiological control of autonomic tone, are directly responsible for producing key outcomes across several cognitive performance domains [11,12,45,59]. Functional magnetic resonance imaging confirms direct causality between and the vagally mediated HF domain of HRV and PFC activity [58,102,104]. Moreover, the PFC is directly implicated in top-down regulation of several cognitive functions critical to tactical training and operations (e.g., attention/situational awareness, decision making, working memory, and emotion regulation), while the amygdala is at the "heart" of the brain's fight or flight response and houses much of the arousal and emotional centers (which typically inhibit PFC control) [12,45]. Neural activity covariation suggests cognitive function is directly linked to the medial visceromotor network—the final common pathway through which cognitive function and emotional response influence autonomic control [12,102]. According to this neurovisceral integration model, low HF HRV is indicative of decreased prefrontal inhibitory influences on sub-cortical structures, like the amygdala, and by extension reduced attentional and emotional self-regulation [58,111]. Motor control regions also possess direct and indirect connections with the PFC and amygdala, such that the efficiency of motor skill acquisition, retention, transfer, and resiliency can be indirectly measured through HRV analysis [12,112]. This indicates an imperative role for the autonomic nervous system (and thus HRV) to affect cognitive function across a wide spectrum of behaviors, from classroom/field education to the performance of tactical skills downrange [113].

When presented with a specific task/stimulus, an individual's HRV response can be altered or vary depending on their knowledge set, skill level, and performance abilities. For the tactical athlete, maintaining emotional (and attentional) control in the face of extreme pressure or danger is vital and influences the entire cognitive performance cascade [12,78]. The ability of an individual to cope with these mental stressors is not only vital to but can alter the psychophysiological mechanisms that control heart rate, and ultimately constrain functionality [12,58,112]. Encountering complex environments that elicit a strong fight or flight response can change self-perception, the current goal-oriented task, and influence attentional/decision making processes, potentially corrupting perception, planning, coordination, and execution of a kinetic response [12,45,114]. Moreover, training and fatigue can also influence within-individual variances across time, such that performance (de)adaptation may be monitored through measuring HRV resource allocation [33,106,115]. For example, during a firefight, a warfighter must pay careful attention to the task at hand, running every bit of relevant visual, auditory, and kinesthetic information through an entire memory registry and decision-making process to rapidly determine the appropriate course of action, then swiftly, yet carefully control their movements to execute the planned response [116,117]. Stress can easily corrupt the entire cascade and this general over-taxation of the system commonly produces a less than desired performance outcome [116,118].

HRV analysis, specifically the fluctuation in HF resource availability and utilization rate/volume, can provide a direct quantification of autonomic efficiency, enabling the measurement of cognitive/motor workload, determination of skill proficiency development across time, and identification of resiliency in the face of other resource demands [33,35,59]. Practically employing HRV in the cognitive performance domain is similar to general physical workload monitoring paradigms, in that, deviations from a resting state or known baseline are compared to time blocks before, during, and after a cognitive or motor demand [69]. Much like physical exercise, decreases in parasympathetic tone (HF HRV) during cognitive/motor skill execution represent resource allocation to accomplish a given goal or task. However, the process is much more temporally sensitive, in that, accurate time-stamping of specific behavioral processes (e.g., anticipation, stimuli presentation, individual responses, post-response recovery) are critical to properly windowing the HRV time-series data [69]. Moreover, while most "physical" HRV paradigms rely on measurement blocks of at least 5 min, many cognitive processes occur on the order of seconds, such that smaller and varying window lengths (e.g., 30, 60, 90 s) may be required to isolate more tactically relevant events (e.g., room-clearing, hand-to-hand combat, calling in a 9-line medevac).

Similar to general resource theory, conservation of autonomic tone (e.g., less disturbance from resting parasympathetic HF) tends to associate with superior performance across numerous facets of perception (attention/emotion regulation, target identification), cognition (working memory, decision making, response inhibition, learning), and action (motor control under duress) [11,35,104,111]. In applied studies, higher resting HRV states (between and within-individual) are most commonly associated with more vagal dominant cognitive control states [11,100], and may indicate a propensity toward superior executive functioning capabilities [59,89,101], while providing resiliency (maintenance of HRV resources from rest to activity/more rapid recovery following) from the negative effects of stress and anxiety [33,69,105,107,119]. Motor performance studies have shown individuals who incur greater deviations from their resting HRV typically demonstrate worse performance on stressful, cognitively challenging motor control tasks [35,106,109]. Additionally, utilizing HRV biofeedback as an educational tool, to promote interoception and self-regulation, has been shown effective at increasing learning rate, reducing performer workload, and increasing performance outcomes [9,10,88,110]. Typically, these paradigms use near-real time HRV markers to help performers identify states during which they may be consuming too many (or the wrong type) of autonomic resources, then measure the affected change from implementing a performance optimization strategy (e.g., mindfulness breathing, cognitive reappraisal).

5.4. Recovering to Restore HRV

Much like training load, recovery loads influence HRV responses and may be used to maintain workload capacities over time [120,121]. Proper recovery following training sessions increases the likelihood of long-term desirable adaptations to stress (i.e., training loads), enhancing both workload capacity (i.e., preparedness and resiliency) and well-being [121,122]. Controlling HRV may be as simple as manipulating respiratory processes, such that heart rate increases during inhalation and decreases with exhalation (3,25). During exercise, both heart rate and respiration increase, causing parasympathetic deactivation or withdrawal, thus increasing overall sympathetic tone (25,26). Conversely, deep breathing or tactical breathing techniques can consciously mitigate parasympathetic withdrawal, thus decreasing the effects of sympathetic tone [123].

Post-exercise recovery has gained a great deal of attention in the past decade, leading to a rise in scientific understanding of accelerated recovery techniques to elicit restored autonomic and cardiovascular homeostasis [124,125]. Flotation using R.E.S.T. (Restricted Environmental Stimulation Therapy) has been effective in increasing sleep quality [126] and

HRV, specifically the parasympathetic marker HF [127]; thus, being effective in facilitating ANS balance, regardless of pre-float autonomic dominance. Whole-body cryostimulation has an immediate and lasting effect on ANS balance that may be observed via improved HRV [128,129]. For example, 3 min whole-body cryostimulation has increased HF, decreased LF, and decreased LF/HF ratios for upwards of 6 h post-cryotherapy [130–132]. Cryotherapy in the evening, after training or competition, has also shown positive effects on sleep quality [129,131,132], which subsequently would improve HRV considering the positive influence of quality sleep hygiene on HRV.

Photobiomodulation (PBM), formerly known as Low-Level Laser Therapy (LLLT) [133], uses red and near-infrared light spectrum wavelengths (600 nm–1100 nm) through light emitting diodes (LED), lasers, or a combination of both, to deeply penetrate the skin and be absorbed by mitochondria of underlying structures (e.g., musculoskeletal tissue) to induce recovery [134,135]. Although limited, the emerging research has shown that whole body PBM may increase sleep quality and the HF of HRV [133,136,137].

Cold Water Immersion consists of 6–20 min immersions in water temperatures below 15 °C (59 °F) [124,125]. The post-exercise effect results in parasympathetic reactivation and sympathetic withdrawal to elicit a restorative effect on autonomic balance and ANS recovery [27,124], resulting in a desirable stress-recovery balance and potentially improving the adaptation to training loads [124].

Many of the aforementioned methods show promise for improving sleep, HRV, and recovery, but require further investigation as to their true efficacy and practicality. However, through the use of a holistic physiological monitoring program, including consistent HRV assessments, it is likely that individual prescription of recovery can be established for the tactical athlete.

6. Conclusions

The key parameters of HRV to be concerned with are rMSSD, HF, and TP. Each provides essential information regarding ANS regulation, making HRV a valuable tool to assess the overall health, wellness, and fitness of tactical personnel [5,27]. Training load is affected by intensity and physiological impact of training, which are represented by the changes in HRV during exercise and recovery [79]. HRV is also influenced by cognitive load, physiological and emotional stress, and environment [18,31,123]. In long-term monitoring, it is a non-invasive method to assess the ANS and make appropriate changes to training and recovery [5]. In short-term monitoring, HRV is useful in assessing responses to acute stimuli, such as a physical or cognitive training events [138]. Since, age, body position, and time of day all influence HRV measurements [5,38,52,139,140], HRV should be highly individualized and assessed relative to the individual. Statistically, time periods before, during, and after activity are compared between one another (and as deltas from accurate baselines or rolling averages) to determine resource modulation in relationship to task performance.

Within high performing populations, HRV assessment and monitoring has recently risen in popularity as one method to foster awareness and avoid overtraining or non-functional overreaching [18,53]. The HRV measures may also be used to understand the differences in occupational requirements among occupational specialties. For example, occupational tasks with high demands for concentration and precision, such as paratroopers, dynamic precision shooters, or medics, require significantly more resources and SNS activity than low-stress, office-based occupational tasks [43]. More traditionally, HRV is clinically used to assess both illness and wellness [27,138], primarily cardiac morbidity and the progression of cardiac death, as well as diabetic neuropathy [5,20], all of which should be confirmed by qualified physicians. More recently, HRV metrics are used to garner further understanding of training workload and physical exercise effects on the body [5,20,46,53]. The information gathered is extremely valuable when assessing expertise or preparedness to train, planning training loads, and prescribing the utilization of recovery tools or techniques [5]. For example, those accustomed to greater levels of physical activity and more experienced in given occupational tasks will have improved overall stress management processes, which may indicate improved capacities for handling certain occupational specialties [43]. The likelihood for facilitating greater awareness of one's own training, recovery, and lifestyle habits, as well as the effect of these habits on their psychophysiology (i.e., HRV), provides another direct benefit of monitoring HRV. Ultimately, HRV measures can be useful for indicating overall health and physical and cognitive preparedness of all tactical personnel, which may be particularly informative for occupational specialties that possess extremely low margins for error in high stress environments (e.g., special forces). Thus, physiological feedback via monitoring HRV would prove useful for addressing the individual's present capability to handle occupational tasks, be it during selection processes or returning to duty following injury or illness but should not be used to diagnose any pathological conditions.

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Review

Effects of Plyometric Jump Training in Female Soccer Player’s Physical Fitness: A Systematic Review with Meta-Analysis

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Abstract: We aimed to assess the effects of plyometric jump training (PJT) on female soccer player’s physical fitness. To this aim, a systematic review with meta-analysis (SRMA) was conducted. The electronic databases PubMed, MEDLINE, Web of Science, and SCOPUS were used. To qualify for inclusion, peer-reviewed studies must have included (i) a PJT programme of ≥2 weeks, (ii) healthy athletes, (iii) a control group, and (iv) physical fitness outcomes (e.g., jump; sprint). Studies were excluded if (i) they incorporated injuried female soccer players, (ii) did not involve PJT or an active control group, (iv) lack of baseline and/or follow-up data. Data was meta-analyzed using the inverse variance random-effects model. Ten moderate-to-high quality studies were included in the analyses, comprising 13 training groups (n = 140) and 10 control groups (n = 110). Small to large (ES = 0.60–2.24; p = 0.040 to <0.001) effects were noted for countermovement jump, drop jump, kicking performance, linear sprint, change of direction speed, and endurance. The moderator analyses (i.e., PJT duration, age groups, competitive level, and soccer experience) revealed no significant differences between groups. In conclusion, PJT may improve the physical fitness of female soccer players. Such improvements might be expected after PJT interventions with six or more weeks of duration, and in players with different chronological ages, competitive levels and soccer experience.

Keywords: human physical conditioning; resistance training; exercise therapy; plyometric exercise; football; sports; athletic performance

1. Introduction

Endurance (cardiorespiratory capacity) is a relevant physical fitness trait in soccer [1]. However, jumping, single and repeated sprinting, change of direction, and kicking are also key proxies of soccer

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performance [2–4]. Indeed, these maximal-intensity single-effort physical fitness traits preceded goal opportunities in competitive leagues [2,5]. For example, sprinting and jumping actions occur in more than 50% of goal situations [2]. Moreover, mean values of acceleration >2.26 m·s^{-2} occur 1.78 times per minute, in addition to sprint distance of 2.87 m per minute, and/or highs-peed running of 6 meter per minute, with greater values during highly-competitive matches [6]. In addition, maximal-intensity short-duration actions such as jumping and sprinting may be associated with team position in a given tournament and/or players' competitive level [4,7,8].

Several training approaches are used among female soccer players to improve jumping, single and repeated sprinting, change of direction and kicking power, as well as endurance attributes [5]. However, plyometric jump training (PJT) may be particularly effective, offering several advantages (e.g., reduced cost; injury prevention) compared to other methods (e.g., traditional resistance training) [9,10]. Additionally, the incorporation of PJT among training practices in soccer might be highly translated into game scenarios. For instance, there is a strong reliance on vertical and horizontal expressions of power during various game scenarios in soccer such as when defending, shooting, and attacking [1,5,11]. In turn, according to the principle of training specificity, soccer players should regularly engage in PJT programs. Indeed, PJT have demonstrated a significant transference effect between jump training exercises and soccer-specific physical performance [12–14].

From a physiological perspective, PJT capitalizes on the stretch-shortening cycle (SSC) where musculotendinous units are eccentrically stretched during the loading or impact phase before concentrically shortening in the push-off or take-off [15,16]. In this regard, PJT results in a wide range of distinct physiological and biomechanical adaptations (e.g., increased motor unit recruitment and rate of force development (RFD) [17–20]. Among soccer athletes (mixed with other sports), after 8 weeks of PJT, significant changes were noted at the muscle fiber level, including myosin heavy chain isoform composition in type I/IIa fibers, increased cross-sectional area, absolute peak calcium-activated force, maximum unloaded and loaded shortening velocity, absolute and normalized peak power, velocity at peak power, absolute force at which peak power was reached, and increased stiffness [21]. In a similar study, increased percentage of type I/IIa and IIa fibers were noted, in line with a decrease in IIx fibres from the vastus lateralis [22]. In another study with soccer players, 8 weeks of PJT combined with resistance training increased electromyography activity (72–110%) in the vastus medialis and rectus femoris muscles during jumping [23]. Another study with soccer players reported significant increases (4–15%; ES = 0.3–1.3) in leg stiffness after 4 weeks of training [24].

Due to the beneficial effects of PJT, several systematic reviews and meta-analysis (SRMA) have been published evidencing the effectiveness of this training mode to improve distinct power-related attributes in athletes from different sports disciplines including handball [25] and volleyball [26]. Likewise, there is a growing body of experimental evidence examining the effects of PJT on physical fitness attributes in female soccer players [27]; however, this evidence has not yet been comprehensively aggregated. Although a recent SRMA examined the effects of PJT on female soccer, only vertical jump height (i.e., countermovement jump) was analyzed [28]. Another SRMA examined the effects of PJT on male soccer players physical fitness [29], including measures such as jumping, sprinting and strength. However, considering the differences between female and male soccer players [1,5] and their potential different response to PJT [30], it would be adventurous to extrapolate results derived from male to female soccer players.

Given the increased scientific awareness of PJT relevance regarding physical fitness improvement, it was deemed appropriate to aggregate PJT studies conducted in female soccer players in a SRMA to strengthen the level of scientific evidence [31] on this topic. This knowledge can guide practitioners to use PJT routines that are effective in female soccer, avoiding the simple transfer from the knowledge related to male soccer players. Thus, the aim of this SRMA was to assess the effects of PJT on female soccer player's physical fitness (e.g., jump, sprint, kicking ability, change of direction speed; anaerobic performance; endurance).

2. Materials and Methods

This SRMA was conducted and reported in accordance with the PRISMA statement [32].

2.1. Eligibility Criteria

A Participants, Intervention, Comparators, Outcomes, and Study (PICOS) design approach was used to rate studies for eligibility [32]. The respective inclusion criteria adopted in our meta-analysis was as follow: (i) apparently healthy female soccer players, with no restrictions on their playing level or age, (ii) a PJT programme, defined as lower-body unilateral or bilateral bounds, jumps, and hops that commonly utilise a pre-stretch or countermovement stressing the stretch-shortening cycle, (iii) a control group, (iv) at least one measure of physical fitness (e.g., jump, sprint, kicking ability, change of direction speed; anaerobic performance; endurance) before and after PJT, (v) controlled trials. The respective exclusion criteria adopted in our meta-analysis was as follow: (i) female soccer players with health problems (e.g., injuries, recent surgery), (ii) exercise interventions not involving PJT or exercise interventions involving PJT programmes representing less than 50% of the total training load when delivered in conjunction with other training interventions (e.g., high-load resistance training), (iii) absence of active control group, (iv) lack of baseline and/or follow-up data, (v) non-controlled trials. Of note, two previous scoping reviews [27,33] indicated that several otherwise high-quality studies in the field of PJT did not include a randomization design. In order to avoid the exclusion of potentially relevant studies, we considered for inclusion non-randomized study designs, as long as baseline values between groups were similar for the main outcome of the study. In contrast, the inclusion of an active control group was considered essential in order to isolate the effect of PJT from the rest of training methods that female soccer players commonly conduct in their regular training schedule.

Additionally, only full-text, peer-reviewed, original studies written in English were considered, excluding cross-sectional, review papers, or training-related studies that did not focus on the effects of PJT exercises (e.g., studies examining the effects of upper-body plyometric exercises). Retrospective studies, prospective studies, studies in which the use of jump exercises was not clearly described, studies for which only the abstract was available, case reports, special communications, letters to the editor, invited commentaries, errata, overtraining studies, and detraining studies were also excluded from the present meta-analysis. In the case of detraining studies, if there was a training period prior to a detraining period, the study was considered for inclusion.

2.2. Information Sources

We considered previous recommendations from the two largest scoping reviews examining PJT to conduct the literature search [27,33]. Computerized literature searches were conducted in the electronic databases PubMed (comprising MEDLINE), Web of Science Core Collection, and SCOPUS. The search strategy was conducted using the Boolean operators AND as well as OR with the following keywords: "ballistic", "complex", "explosive", "force-velocity", "plyometric", "stretch-shortening cycle", "jump", "training", "female", "women", "football", and "soccer". For example, the following search was adopted using Pubmed: (((((((("randomized controlled trial" [Publication Type]) OR "controlled clinical trial" [Publication Type]) OR "randomized" [Title/Abstract]) OR "trial" [Title]) OR "clinical trials as topic" [MeSH Major Topic]) AND "soccer" [Title/Abstract]) OR "football" [Title/Abstract]) AND "training" [Title/Abstract]) OR "plyometric" [Title/Abstract]). After an initial search (April 2017), accounts were created in the respective databases. Through these accounts, automatically generated emails were received for updates regarding the search terms used. The search was refined in May 2019 and updates were received daily (if available), and studies were eligible for inclusion up to January 2020. Following the formal systematic searches, we conducted additional hand-searches (e.g., personal libraries). One author (RRC) performed the only search.

2.3. Study Selection and Data Collection Process

After the exclusion of repeated article titles, a review of retrieved article titles was conducted. Then, examination of article abstracts followed. Thereafter, full articles were assessed. The reasons to exclude full-text articles were recorded. The data extracted from the included articles was recorded in a pre-form created in Microsoft Excel (Microsoft Corporation, Redmond, WA, USA). Two authors (MSG and JSS) conducted the process independently, and a third author (RRC) resolved disagreements regarding study eligibility.

2.4. Data Items

For the current review, physical fitness was chosen as the main outcome. *A priori*, common measures of physical fitness were considered, but not limited to: (i) jump (i.e., height; distance; flight time; power; reactive strength [i.e., mm.ms; ms.ms]), (ii) sprint (i.e., time; velocity), (iii) kicking ability (i.e., distance; velocity), (iv) change of direction speed (i.e., time; speed), (v) anaerobic performance (e.g., repeated sprint ability mean time; 30-s Wingate test mean power), (vi) endurance (e.g., shuttle-run total time or distance).

In addition to the aforementioned data items, adverse effects were registered, and descriptive characteristics of the PJT interventions (e.g., duration; frequency) and athletes (e.g., age; fitness level) were extracted. A complete description of the additional PJT and athletes' characteristics have been previously published [33].

2.5. Methodological Quality in Individual Studies

The methodological quality of eligible studies was assessed using the Physiotherapy Evidence Database (PEDro) scale, as previously described [34] and interpreted for PJT literature [28,35]. Briefly, studies with ≤3 points were considered of poor quality, 4–5 points as moderate quality, and 6–10 points as high quality. Two of the authors (RRC and MSG) independently scored the articles. Disagreements in the rating between both authors was resolved through discussion with a third author (JSS). Aiming to control the risk of bias between authors, the Kappa correlation test was used to analyse the agreement level for the included studies. An agreement level of k = 0.89 was obtained.

2.6. Summary Measures and Synthesis of Results, and Publication Bias

For analysis and interpretation of results, we followed previous recommendations for PJT meta-analyses [26,28,36,37]. Briefly, using a random-effects model meta-analyses were conducted only if ≥3 studies provided means and standard deviations for the same pre-post PJT parameter (e.g., sprint time), in order to calculate an effect size (ES; Hedges' g ES) alongside their respective 95% confidence intervals (CIs). Data was standardized using post-intervention standard deviation score. Calculated ES were interpreted as previously recommended for sport sciences studies: <0.2, trivial; 0.2–0.6, small; >0.6–1.2, moderate; >1.2–2.0, large; >2.0–4.0, very large; >4.0, extremely large [38]. For studies that incorporated ≥2 intervention groups and only one control group, the sample size in the control group was proportionately divided during analyses [39]. Heterogeneity was assessed using the I^2 statistic, with values of <25%, 25–75%, and >75% considered as representative of low, moderate and high heterogeneity, respectively. The risk of bias was analyzed with the Egger's test [40]. To adjust for publication bias, a sensitivity analysis was conducted using the trim and fill method [41], with L0 as the default estimator for the number of missing studies [42]. Subgroup analyses to assess the potential influence of PJT duration (number of weeks), training frequency (number of sessions per week), total number of training sessions, in addition to the age, the expertise level of the participants (i.e., moderate-level vs. high-level players), and the athletes years of soccer experience were performed according to the median split technique. Analyses were performed using specialized software (Comprehensive Meta-Analysis; version 2; Biostat, Englewood, NJ, USA). Statistical significance was set at $p < 0.05$.

3. Results

3.1. Study Selection

The electronic search process identified 7206 studies (2261 from PUBMED, 2280 from SCOPUS, and 2665 from WOS), plus 24 studies through other sources. Duplicate studies were then removed (n = 4761). Study titles and abstracts were screened with a further 2020 studies removed. Accordingly, full-text versions of 449 studies were screened. From these, 224 studies did not include an appropriate study design (e.g., control group), 188 studies did not include soccer players or female soccer players only, and 27 were excluded for different reasons (e.g., no measure of physical fitness provided). The remaining 10 studies [43–52] were included in the SRMA (Figure 1). The included studies involved 13 individual experimental groups and 140 participants, and 110 participants in the 10 control groups. The characteristics of the participants and PJT interventions are indicated in the Tables 1 and 2. Briefly, the age of the participants was between a mean of 13.4 to 26.6 years, with a fitness level that varied from recreationally trained to professional athletes. The PJT interventions lasted between six up to 12 weeks. Almost all studies (except one) incorporated PJT during the in-season period. A complete description of the physical fitness measures used in the meta-analyses is provided in Table 3.

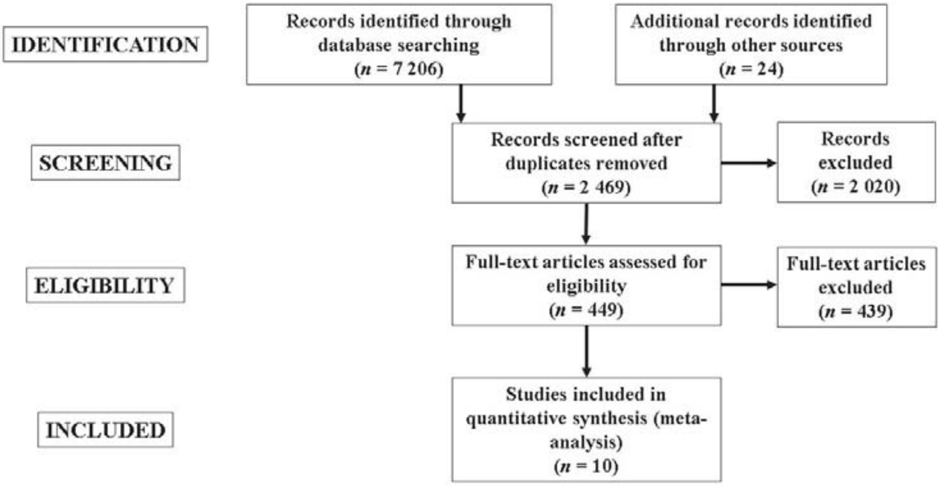


Figure 1. Flow diagram of the search process.

3.2. Methodological Quality

Using the PEDro checklist, five studies achieved 4 or 5 points and were classified as being of “moderate” quality, while five studies achieved 6–10 points and were therefore considered as being of “high” methodological quality (Table 4).

Table 1. Characteristics of included study participants and of PJT programs.

Authors	Treat	Age (y) *	SPT	Fit	Freq	Dur	Int	BH (cm)	NTJ	Tply
Chimera et al. 2004	WD	20.0	No	Moderate-high	2	6	Max (RSI; quickness)	45	3940 + 1680 s	B + V + C + A + L + Turn
Fischetti et al. 2019	WD	26.6	Yes	High	3	12	Max (height; distance; velocity; RSI)	60 (hurdle) 50 (stands)	3240	B + C + V + H
Ozbar et al. 2014	WD	18.3	Yes	Moderate	1	8	NR	20 to 60 (hurdle)	1210	H + V + L + D + U + B + A + C
Ramirez-Campillo et al. 2018	WD	22.1	No	Moderate	1-2	8	Max (height; distance, RSI)	5 to 35 (optimal (RSI)	810	U + B + C + A + V + H + Turn + fast SSC + slow SSC
Ramirez-Campillo et al. 2016a	WD	23	No	Moderate	2	6	Max (height; distance; RSI)	20	1440	V + H + U + B + C + A
Ramirez-Campillo et al. 2016b	WD	22.4	No	Moderate-high	2	6	Max (height; distance; RSI)	NA	1440	U + B + V + H + C + A
Rosas et al. 2017	WD	23.6	No	Normal	2	6	Max (height; distance; RSI)	40	1440	V + H + U + B + C + A
Rubley et al. 2011	ID	13.4	No	Moderate-high	1	12	NR	30	1680	U + H + B + V + L + A + C
Sedano et al. 2009	WD	22.8	Yes	High	3	12	Max (height; distance; velocity; RSI)	50 to 60	3240	V + H + B + C + A
Siegler et al. 2003	ID	16.5	No	Normal-moderate	1–3	10	Max (height; distance; quickness)	18 to 42	1046 + 70 s + 900 m	B + V + A + C + H + U + L + Back + D

Note: abbreviations descriptions ordered alphabetically. *: mean values reported for experimental and control groups; A: acyclical (non-repeated); B: bilateral; C: cyclical (repeated); D: diagonal; BH: box height (for those drills that required the use of a box, not necessarily applied to drop jumps); Com: combined PJT with another type of drill; Fitness level: classified as in the recent review by Ramirez-Campillo et al. [28], (i) NR, (ii) high encompasses professional/elite athletes with regular enrolment in national and/or international competitions or highly trained participants with ≥10 training hours per week or ≥6 training sessions per week and a regularly scheduled official or friendly competition, (iii) moderate encompasses non-elite/professional athletes, with a regular attendance in regional and/or national competitions, between 5–9.9 training hours per week or 3–5 training sessions per week and a regularly scheduled official or friendly competition, (iv) normal encompasses recreational athletes with <5 training hours per week with sporadic or no participation in competition; Freq: PJT frequency (sessions per week); H: horizontal; I: intensity; ID: insufficiently described, when the PJT treatment description omitted the reporting of any of the following: duration, frequency, intensity, type of exercises, sets, repetitions; IS: in-season; L: lateral; Max: maximal, involving either maximal effort to achieve maximal height, distance, RSI, velocity, or another marker of intensity; NA: non-applicable; NR: non-reported; NTJ: number of total jumps; PJT: plyometric jump training; PO: progressive overload, in the form of either volume, intensity, or a combination of these; RBR: rest time between repetitions (only when the PJT programs incorporated non-repeated jumps); RBS: rest time between sets; RBTS: rest between training sessions; Rep: replace, denoting if the athletes replace some common drills with PJT drills; RSI: reactive strength index; RT: resistance training; SPT: indicates if the participants had previous systematic experience with PJT; SSC: stretch-shortening cycle; Surf: type of surface used during the intervention; T: type of drill; TP: training period; Tply: type of PJT drills used; U: unilateral; V: vertical; Vo: volume; WD: well described, when treatment description allowed for adequate study PJT replication, including the reporting of duration, frequency, intensity, type of exercises, sets, and repetitions.

Table 2. Characteristics of PJT programs.

Authors	Com	RBS (s)	RBR (s)	RBTS (Hours)	Tsurf	PO	TP	Replace	Taper
Chimera et al. 2004	No	30 to 120	NR	NR	NR	Vo + T	OS	A	No
Fischetti et al. 2019	No	240	30 to 60	48 to 72	Hard synthetic floor	Vo	IS	Yes	Yes
Ozbar et al. 2014	No	60 to 300	NR	168	NR	Vo + I + T	IS	No	No
Ramirez-Campillo et al. 2018	No	30 to 60	5 to 15	48 to 168	Combined (grass, land, dirt)	Vo	IS	Yes (6%)	Yes
Ramirez-Campillo et al. 2016a	No	60	15	48 or more	Grass	Vo	IS	Yes	No
Ramirez-Campillo et al. 2016b	No	60	15	72 or more	Grass	Vo	IS	Yes	No
Rosas et al. 2017	No	60	15	48 or more	Grass	Vo	IS	Yes	No
Rubley et al. 2011	Cutting drills	NR	NR	168	NR	T	IS	No	No
Sedano et al. 2009	No	30 to 300	NR	48 to 72	Hard synthetic floor	Vo	IS	Yes	Yes
Siegler et al. 2003	RT + sprints	NR	NR	48 to 144	Grass	Vo + T	IS	Yes	Yes

Note: abbreviations descriptions ordered alphabetically. *: mean values reported for experimental and control groups; A: acyclical (non-repeated); B: bilateral; C: cyclical (repeated); D: diagonal; BH: box height (for those drills that required the use of a box, not necessarily applied to drop jumps); Com: combined PJT with another type of drill; Fitness level: classified as in the recent review by Ramirez-Campillo et al. [28], (i) NR, (ii) high encompasses professional/elite athletes with regular enrolment in national and/or international competitions or highly trained participants with ≥10 training hours per week or ≥6 training sessions per week and a regularly scheduled official or friendly competition, (iii) moderate encompasses non-elite/professional athletes, with a regular attendance in regional and/or national competitions, between 5–9.9 training hours per week or 3–5 training sessions per week and a regularly scheduled official or friendly competition, (iv) normal encompasses recreational athletes with <5 training hours per week with sporadic or no participation in competition; Freq: PJT frequency (sessions per week); H: horizontal; I: intensity; ID: insufficiently described, when the PJT treatment description omitted the reporting of any of the following: duration, frequency, intensity, type of exercises, sets, repetitions; IS: in-season; L: lateral; Max: maximal, involving either maximal effort to achieve maximal height, distance, RSI, velocity, or another marker of intensity; NA: non-applicable; NR: non-reported; NTJ: number of total jumps; PJT: plyometric jump training; PO: progressive overload, in the form of either volume, intensity, or a combination of these; RBR: rest time between repetitions (only when the PJT programs incorporated non-repeated jumps); RBS: rest time between sets; RBTS: rest between training sessions; Rep: replace, denoting if the athletes replace some common drills with PJT drills; RSI: reactive strength index; RT: resistance training; SPT: indicates if the participants had previous systematic experience with PJT; SSC: stretch-shortening cycle; Surf: type of surface used during the intervention; T: type of drill; TP: training period; Tply: type of PJT drills used; U: unilateral; V: vertical; Vo: volume; WD: well described, when treatment description allowed for adequate study PJT replication, including the reporting of duration, frequency, intensity, type of exercises, sets, and repetitions.

Table 3. Study groups and their physical fitness.

Author (Year)	Test	PJT, Before $^{\alpha}$			Control, Before			PJT, After			Control, After		
		Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*
Chimera et al. 2004	Jump (DJ 45-cm; cm)	17.89	2.29	9	18.17	2.24	9	18.89	2.45	9	18.5	2.06	9
Fischetti et al. 2019	Jump (CMJ; cm)	33.6	5.5	14	32.1	6.4	14	36.8	5.8	14	32.7	5.7	14
	CODS (T-test; s)	8.8	0.3	14	8.9	0.3	14	8.5	0.3	14	8.8	0.4	14
Ozbar et al. 2014	Jump (CMJ; cm)	39.8	4.5	9	35.4	4.6	9	46.8	2.2	9	37.9	3.9	9
	Vertical jump power (CMJ; w)	3480	643.2	9	2492.2	432.1	9	3855.2	536.6	9	3080.2	420.4	9
	Linear sprint (20 m; s)	3.7	0.3	9	3.9	0.4	9	3.4	0.2	9	4	0.5	9
Ramírez-Campillo et al. 2018 (1 PJT/week)	Jump (CMJ; cm)	28.5	6.9	8	28.8	4.9	3	31.5	7.5	8	29.9	5.1	3
	Jump (DJ 20 cm; cm)	27.2	5.9	8	28.7	4.3	3	30.9	7.8	8	29.3	4.6	3
	Kicking ability (Instep kick; km·h^{-1})	65.1	9	8	67.3	7.2	3	70.6	8.9	8	68.9	7.5	3
	Linear sprint (15 m; s)	3.28	0.1	8	3.42	0.2	3	3.01	0.1	8	3.45	0.2	3
	CODS (Meylan test; s)	4.94	0.2	8	4.96	0.2	3	4.57	0.2	8	4.95	0.4	3
	Endurance (Yo-yo test level 1; m)	573	237	8	606	175	3	628	224	8	612	179	3
Ramírez-Campillo et al. 2018 (2 PJT/week)	Jump (CMJ; cm)	27.4	4.3	8	28.8	4.9	4	30.1	4.7	8	29.9	5.1	4
	Jump (DJ 20 cm; cm)	27.7	5.8	8	28.7	4.3	4	31.3	6.6	8	29.3	4.6	4
	Kicking ability (instep kick; km·h^{-1})	63	9.5	8	67.3	7.2	4	68.9	11	8	68.9	7.5	4
	Linear sprint (15 m; s)	3.43	0.1	8	3.42	0.2	4	3.1	0.1	8	3.45	0.2	4
	CODS (Meylan test; s)	5.12	0.3	8	4.96	0.2	4	4.74	0.3	8	4.95	0.4	4
	Endurance (Yo-yo Test IR1; m)	630	192	8	606	175	4	690	203	8	612	179	4
Ramírez-Campillo et al. 2016a (placebo)	Jump (CMJ; cm)	28.7	5.1	10	25.9	4.1	5	30	5.3	10	25.9	3.2	5
	Vertical jump power (CMJ; w)	1940	338	10	1979	211	5	2037	354	10	1914	249	5
	Jump (DJ; mm·mm^{-1})	1.36	0.4	10	1.4	0.66	5	1.36	0.4	10	1.39	0.6	5
	Linear sprint (20 m; s)	3.87	0.3	10	3.99	0.2	5	3.74	0.26	10	3.98	0.14	5
	CODS (Illinois test; s)	18.8	1.2	10	19.4	0.8	5	18.2	0.9	10	19.3	0.5	5
	Endurance (20 m mult-stage shuttle run test; min)	7.8	1.5	10	7.4	1.9	5	8.3	1.3	10	7.5	1.8	5
	Anaerobic performance (Test RAST mean; s)	7.08	0.6	10	7.35	0.5	5	6.78	0.53	10	7.2	0.31	5
Ramírez-Campillo et al. 2016a (creatine)	Jump (CMJ; cm)	27.3	5.2	10	25.9	4.1	5	28.9	4.6	10	25.9	3.2	5
	Vertical Jump Power (CMJ; w)	1969	250	10	1979	211	5	2108	278	10	1914	249	5
	Jump (DJ; mm·mm^{-1})	1.33	0.3	10	1.4	0.6	5	1.46	0.3	10	1.39	0.6	5
	Linear sprint (20 m; s)	3.98	0.4	10	3.99	0.2	5	3.85	0.37	10	3.98	0.14	5
	CODS (Illinois test; s)	19.3	1.1	10	19.4	0.8	5	18.8	0.8	10	19.3	0.5	5
	Endurance (20 m mult-stage shuttle run test; min)	8	1.6	10	7.4	1.9	5	8.5	1.3	10	7.5	1.8	5
	Anaerobic performance (RAST mean; s)	7.48	1	10	7.35	0.5	5	7.08	0.88	10	7.3	0.31	5
Ramírez-Campillo et al. 2016b	Jump (CMJ; cm)	26.7	5.5	19	26.6	4.8	19	29.4	5.8	19	26.6	4.3	19
	Jump (CMJA; cm)	30.3	6.5	19	29.2	5.5	19	32.6	6.5	19	28.9	5.1	19
	Jump (DJ 40 cm; cm·ms^{-1})	0.119	0.04	19	0.101	0.03	19	0.144	0.04	19	0.107	0.03	19
	Linear sprint (30 m; s)	5.69	0.31	19	5.72	0.28	19	5.4	0.32	19	5.82	0.31	19
	CODS (Illinois test; s)	19.48	0.9	19	19.79	1	19	18.73	1	19	19.93	0.9	19
	Endurance (20 m mult-stage shuttle run test; min)	8.4	1.9	19	8.6	1.6	19	9.1	1.2	19	8.6	1.1	19

Table 3. *Cont.*

Author (Year)	Test	PJT, Before $^{\alpha}$			Control, Before			PJT, After			Control, After		
		Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*
Rosas et al. 2017 (placebo)	Jump (CMJ; cm)	24.8	3.4	8	28.9	5.8	5	26.4	3	8	29.4	6.3	5
	Vertical jump power (CMJ; w)	1974	259	8	2003	341	5	2140	250	8	1989	272	5
	Jump (DJ 40 cm; cm)	1.24	0.4	8	1.33	0.3	4	1.67	0.6	8	1.33	0.5	4
	Linear sprint (20 m; s)	3.89	0.4	8	3.78	0.3	4	3.77	0.4	8	3.83	0.4	4
	CODS (Illinois test; s)	18.5	0.3	8	18.7	0.4	4	18.2	0.4	8	18.7	0.4	4
	Endurance (20 m shuttle run; min)	7.1	1.1	8	7.9	1.8	4	7.5	1	8	7.9	2	4
	Anaerobic performance (RAST mean; s)	7.49	0.8	8	7.18	0.9	4	7.18	0.6	8	7.17	0.8	4
Rosas et al. 2017 (beta-alanina)	Jump (CMJ; cm)	28.1	3.5	8	28.9	5.8	4	30.6	3.1	8	29.4	6.3	4
	Vertical Jump Power (CMJ; w)	1944	340	8	2003	341	4	2122	318	8	1989	272	4
	Jump (DJ 40 cm; cm)	1.11	0.2	8	1.33	0.3	5	1.53	0.5	8	1.33	0.5	5
	Linear sprint (20 m; s)	3.92	0.2	8	3.78	0.3	5	3.8	0.1	8	3.83	0.4	5
	CODS (Illinois test; s)	18.9	0.7	8	18.7	0.4	5	18.6	0.8	8	18.7	0.4	5
	Endurance (20 m shuttle-run; min)	7.9	1.7	8	7.9	1.8	5	8.5	1.7	8	7.9	2	5
	Anaerobic performance (RAST mean; s)	7.61	0.5	8	7.18	0.9	5	7.1	0.5	8	7.17	0.8	5
Rubley et al. 2011	Jump (CMJA; cm)	39.6	8.2	10	39.4	8.3	6	47	8.1	10	39.6	8.2	10
	Kicking ability (m)	25.9	2.6	10	27.6	2.5	6	33	3.7	10	23.3	3.7	6
Siegler et al. 2003	Jump (CMJA; cm)	37.65	0.15	17	36.46	3.68	17	39.37	4.69	17	39.19	4.45	17
	Vertical jump power (Wingate 30 s; kg·m·min^{-1})	10.36	2.38	17	9.59	0.92	17	10.68	2.2	17	9.78	1.36	17
	Linear sprint (20 m; s)	3	0.15	17	2.89	0.13	17	2.9	0.13	17	2.85	0.13	17
	Endurance (LIST; s)	646	167.5	17	1064	195.2	17	1040	157.33	17	1115	157.51	17
	Anaerobic performance (Wingate 30 s; kg·m·min^{-1})	7.27	0.49	17	7.76	0.6	17	7.37	0.64	17	7.73	0.78	17
Sedano-Campo et al. 2009	Jump (CMJ; cm)	25.6	1	10	26.2	0.9	10	29.3	1	10	25.9	0.9	10
	Jump (DJ 40 cm; cm)	24.9	1.1	10	27.1	1	10	28.9	0.8	10	25.6	0.9	10
	Kicking ability (km·h^{-1})	70	2.4	10	75.8	1.5	10	78.3	2.1	10	74.1	1.1	10

$^{\alpha}$: before and after values denotes the mean ± standard deviation for each group before and after the intervention, respectively. Note: abbreviations descriptions ordered alphabetically. CMJ: countermovement jump; CMJA: countermovement jump with arm swing; CODS: change of direction speed; DJ: drop jump; LIST: Loughborough intermittent shuttle test; RAST: repeated anaerobic sprint test; SD: standard deviation.

Table 4. Physiotherapy Evidence Database (PEDro) scale ratings.

	N° 1 *	**N° 2**	**N° 3**	**N° 4**	**N° 5**	**N° 6**	**N° 7**	**N° 8**	**N° 9**	**N° 10**	**N° 11**	**Total ****
Chimera et al. 2004	1	1	0	1	0	0	0	1	0	1	1	5
Fischetti et al. 2019	1	1	0	1	0	0	0	1	1	1	1	6
Ozbar et al. 2014	0	1	0	1	0	0	0	1	0	1	1	5
Ramirez-Campillo et al. 2018	1	1	1	1	0	0	1	0	0	1	1	6
Ramirez-Campillo et al. 2016 a	0	1	1	1	0	1	1	1	0	1	1	9
Ramirez-Campillo et al. 2016 b	0	1	1	1	0	0	0	1	0	1	1	7
Rosas et al. 2018	0	1	1	1	1	1	1	1	0	1	1	9
Rubley et al. 2011	0	0	0	1	0	0	0	1	0	1	1	4
Sedano-Campo et al. 2009	0	1	0	1	0	0	0	1	0	1	1	5
Siegler et al. 2003	0	0	0	1	0	0	0	1	0	1	1	4

*: PEDro scale items number; **: the total number of points from a possible maximal of 10. A detailed explanation for each PEDro scale item can be accessed at https://www.pedro.org.au/english/downloads/pedro-scale. In brief: item 1, eligibility criteria were specified; item 2, participants were randomly allocated to groups; item 3, allocation was concealed; item 4, the groups were similar at baseline; item 5, there was blinding of all participants regarding the plyometric jump training programme being applied; item 6, there was blinding of all coaches responsible for the application of plyometric jump training programme regarding its aim toward the improvement of physical fitness; item 7, there was blinding of all assessors involved in measurement of physical fitness attributes; item 8, measures of at least one key fitness variable were obtained from more than 85% of participants initially allocated to groups; item 9, all participants for whom fitness variables were available received the treatment or control condition as allocated or, data for at least one key fitness variable was analysed by "intention to treat"; item 10, the results of between-group statistical comparisons are reported for at least one key fitness variable; and item 11, point measures and measures of variability for at least one key fitness variable are provided.

3.3. Meta-Analysis Results for Countermovement Jump

Seven studies provided data for CMJ, involving 10 experimental and seven control groups (pooled $n = 182$). There was a significant effect of PJT on CMJ (ES = 0.71; 95% CI = 0.20 to 1.23; $p = 0.007$; $I^2 = 62.9\%$; Egger's test $p = 0.224$; Figure 2A). The relative weight of each study in the analysis ranged from 6.9% to 13.3%.

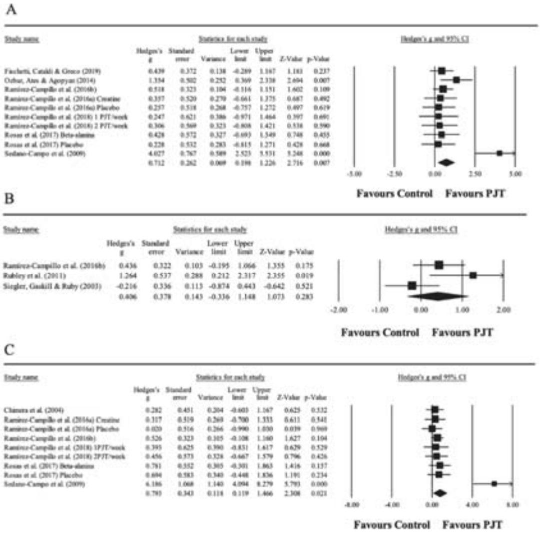


Figure 2. Forest plot of changes in countermovement jump (**A**), countermovement jump with arms awing (**B**), and drop jump performance (**C**) in athletes participating in plyometric jump training (PJT) compared to controls. Values shown are effect sizes (Hedges's g) with 95% confidence intervals (CI). The size of the plotted squares reflects the statistical weight of the study.

No significant sub-group difference (between-group $p = 0.188$) was found when PJT interventions with ≤6 weeks (5 study groups; ES = 0.40; 95% CI = −0.01 to 0.80; within-group $I^2 = 0.0\%$) were compared to PJT interventions with >6 weeks (5 study groups; ES = 1.18; 95% CI = 0.09 to 2.28; within-group I^2 = 81.1%).

Similarly, no significant sub-group difference (between-group p = 0.664) was found when PJT interventions with players ≤22.8 years old (4 study groups; ES = 0.62; 95% CI = 0.17 to 1.07; within-group I^2 = 0.0%) were compared to PJT interventions with players >22.8 years old (6 study groups; ES = 0.84; 95% CI = −0.04 to 1.71; within-group I^2 = 76.6%).

Moreover, no significant sub-group difference (between-group p = 0.080) was found when PJT interventions with high-level players (4 study groups; ES = 1.41; 95% CI = 0.26 to 2.56; within-group I^2 = 85.3%) were compared to PJT interventions with moderate-level players (6 study groups; ES = 0.30; 95% CI = −0.14 to 0.75; within-group I^2 = 0.0%).

Furthermore, no significant sub-group difference (between-group p = 0.420) was found when PJT interventions conducted on players with ≤5.7 years of soccer experience (4 study groups; ES = 1.42; 95% CI = −0.05 to 2.89; within-group I^2 = 83.8%) were compared to PJT interventions conducted on players with >5.7 years of soccer experience (5 study groups; ES = 0.40; 95% CI = −0.01 to 0.80; within-group I^2 = 0.0%).

3.4. Meta-Analysis Results for Countermovement Jump with Arm Swing

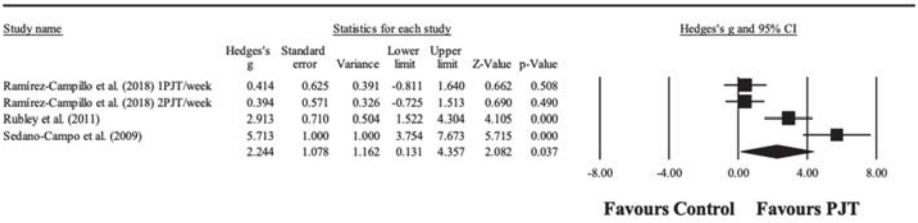
Three studies provided data for CMJA, involving three experimental and three control groups (pooled n = 88). There was a non-significant effect of PJT on CMJA (ES = 0.41; 95% CI = −0.34 to 1.15; p = 0.28; I^2 = 65.3%; Egger's test p = 0.452; Figure 2B). The relative weight of each study in the analysis ranged from 25.4% to 37.8%.

3.5. Meta-Analysis Results for Drop Jump

Six studies provided data for DJ, involving nine experimental and six control groups (pooled n = 154). There was a significant effect of PJT on DJ (ES = 0.79; 95% CI = 0.12 to 1.47; p = 0.021; I^2 = 73.1%; Egger's test p = 0.063; Figure 2C). The relative weight of each study in the analysis ranged from 6.3% to 14.0%.

3.6. Meta-Analysis Results for Kicking Performance

Three studies provided data for kicking performance, involving four experimental and three control groups (pooled n = 59). There was a significant effect of PJT on kicking performance (ES = 2.24; 95% CI = 0.13 to 4.36; p = 0.037; I^2 = 89.4%; Egger's test p = 0.040; Figure 3). After the trim and fill method, the adjusted values remained as the observed values. The relative weight of each study in the analysis ranged from 22.8% to 26.2%.



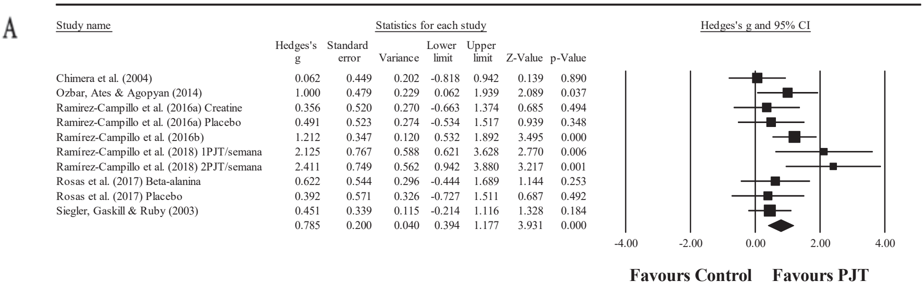
Study name	Statistics for each study						
	Hedges's g	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Ramírez-Campillo et al. (2018) 1PJT/week	0.414	0.625	0.391	-0.811	1.640	0.662	0.508
Ramírez-Campillo et al. (2018) 2PJT/week	0.394	0.571	0.326	-0.725	1.513	0.690	0.490
Rubley et al. (2011)	2.913	0.710	0.504	1.522	4.304	4.105	0.000
Sedano-Campo et al. (2009)	5.713	1.000	1.000	3.754	7.673	5.715	0.000
	2.244	1.078	1.162	0.131	4.357	2.082	0.037

Figure 3. Forest plot of changes in kicking performance, in athletes participating in plyometric jump training (PJT) compared to controls. Values shown are effect sizes (Hedges's g) with 95% confidence intervals (CI). The size of the plotted squares reflects the statistical weight of the study.

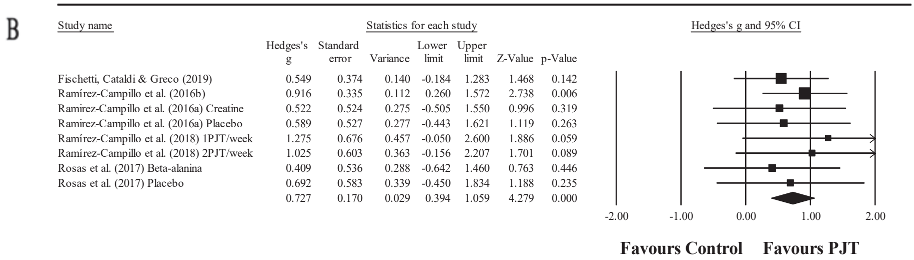
3.7. Meta-Analysis Results for Linear Sprint

Seven studies provided data for linear sprint performance, involving 10 experimental and seven control groups (pooled n = 186). There was a significant effect of PJT on linear sprint performance

(ES = 0.79; 95% CI = 0.39 to 1.18; $p < 0.001$; $I^2 = 38.2\%$; Egger's test $p = 0.257$; Figure 4A). The relative weight of each study in the analysis ranged from 5.4% to 15.3%.



Study name	Hedges's g	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Chimera et al. (2004)	0.062	0.449	0.202	-0.818	0.942	0.139	0.890
Ozbar, Ates & Agopypan (2014)	1.000	0.479	0.229	0.062	1.939	2.089	0.037
Ramirez-Campillo et al. (2016a) Creatine	0.356	0.520	0.270	-0.663	1.374	0.685	0.494
Ramirez-Campillo et al. (2016a) Placebo	0.491	0.523	0.274	-0.534	1.517	0.939	0.348
Ramirez-Campillo et al. (2016b)	1.212	0.347	0.120	0.532	1.892	3.495	0.000
Ramírez-Campillo et al. (2018) 1PJT/semana	2.125	0.767	0.588	0.621	3.628	2.770	0.006
Ramírez-Campillo et al. (2018) 2PJT/semana	2.411	0.749	0.562	0.942	3.880	3.217	0.001
Rosas et al. (2017) Beta-alanina	0.622	0.544	0.296	-0.444	1.689	1.144	0.253
Rosas et al. (2017) Placebo	0.392	0.571	0.326	-0.727	1.511	0.687	0.492
Siegler, Gaskill & Ruby (2003)	0.451	0.339	0.115	-0.214	1.116	1.328	0.184
	0.785	0.200	0.040	0.394	1.177	3.931	0.000



Study name	Hedges's g	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Fischetti, Cataldi & Greco (2019)	0.549	0.374	0.140	-0.184	1.283	1.468	0.142
Ramírez-Campillo et al. (2016b)	0.916	0.335	0.112	0.260	1.572	2.738	0.006
Ramirez-Campillo et al. (2016a) Creatine	0.522	0.524	0.275	-0.505	1.550	0.996	0.319
Ramirez-Campillo et al. (2016a) Placebo	0.589	0.527	0.277	-0.443	1.621	1.119	0.263
Ramírez-Campillo et al. (2018) 1PJT/week	1.275	0.676	0.457	-0.050	2.600	1.886	0.059
Ramírez-Campillo et al. (2018) 2PJT/week	1.025	0.603	0.363	-0.156	2.207	1.701	0.089
Rosas et al. (2017) Beta-alanina	0.409	0.536	0.288	-0.642	1.460	0.763	0.446
Rosas et al. (2017) Placebo	0.692	0.583	0.339	-0.450	1.834	1.188	0.235
	0.727	0.170	0.029	0.394	1.059	4.279	0.000

Figure 4. Forest plot of changes in linear sprint performance (**A**) (**upper**) and change of direction speed performance (**B**) (**bottom**), in athletes participating in plyometric jump training (PJT) compared to controls. Values shown are effect sizes (Hedges's g) with 95% confidence intervals (CI). The size of the plotted squares reflects the statistical weight of the study.

No significant sub-group difference (between-group $p = 0.167$) was found when PJT interventions with ≤6 weeks (six study groups; ES = 0.62; 95% CI = 0.24 to 1.00; within-group $I^2 = 0.0\%$) were compared to PJT interventions with >6 weeks (four study groups; ES = 1.30; 95% CI = 0.41 to 2.20; within-group $I^2 = 63.9\%$).

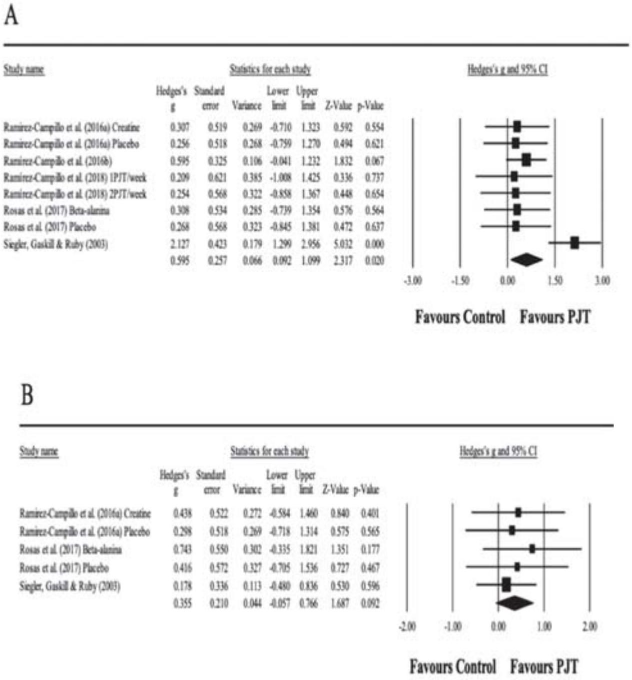
Similarly, no significant sub-group difference (between-group $p = 0.545$) was found when PJT interventions with players ≤21.45 years old (five study groups; ES = 1.02; 95% CI = 0.25 to 1.79; within-group $I^2 = 65.3\%$) were compared to PJT interventions with players >21.45 years old (five study groups; ES = 0.75; 95% CI = 0.33 to 1.16; within-group $I^2 = 0.0\%$).

3.8. Meta-Analysis Results for Change of Direction Speed

Five studies provided data for CODS performance, involving eight experimental and five control groups (pooled $n = 144$). There was a significant effect of PJT on CODS performance (ES = 0.73; 95% CI = 0.39 to 1.06; $p < 0.001$; $I^2 = 0.0\%$; Egger's test $p = 0.813$; Figure 4B). The relative weight of each study in the analysis ranged from 6.3% to 25.8%.

3.9. Meta-Analysis Results for Endurance

Five studies provided data for endurance performance, involving eight experimental and five control groups (pooled $n = 150$). There was a significant effect of PJT on endurance performance (ES = 0.60; 95% CI = 0.09 to 1.10; $p = 0.020$; $I^2 = 53.7\%$; Egger's test $p = 0.328$; Figure 5A). The relative weight of each study in the analysis ranged from 10.0% to 17.3%.



A

Study name	Hedges's g	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Ramirez-Campillo et al. (2016a) Creatine	0.307	0.519	0.269	-0.710	1.323	0.592	0.554
Ramirez-Campillo et al. (2016a) Placebo	0.256	0.518	0.268	-0.759	1.270	0.494	0.621
Ramirez-Campillo et al. (2016b)	0.595	0.325	0.106	-0.041	1.232	1.832	0.067
Ramirez-Campillo et al. (2018) 1PJT/week	0.209	0.621	0.385	-1.008	1.425	0.336	0.737
Ramirez-Campillo et al. (2018) 2PJT/week	0.254	0.568	0.322	-0.858	1.367	0.448	0.654
Rosas et al. (2017) Beta-alanina	0.308	0.534	0.285	-0.739	1.354	0.576	0.564
Rosas et al. (2017) Placebo	0.268	0.568	0.323	-0.845	1.381	0.472	0.637
Siegler, Gaskill & Ruby (2003)	2.127	0.423	0.179	1.299	2.956	5.032	0.000
	0.595	0.257	0.066	0.092	1.099	2.317	0.020

B

Study name	Hedges's g	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Ramirez-Campillo et al. (2016a) Creatine	0.438	0.522	0.272	-0.584	1.460	0.840	0.401
Ramirez-Campillo et al. (2016a) Placebo	0.298	0.518	0.269	-0.718	1.314	0.575	0.565
Rosas et al. (2017) Beta-alanina	0.743	0.550	0.302	-0.335	1.821	1.351	0.177
Rosas et al. (2017) Placebo	0.416	0.572	0.327	-0.705	1.536	0.727	0.467
Siegler, Gaskill & Ruby (2003)	0.178	0.336	0.113	-0.480	0.836	0.530	0.596
	0.355	0.210	0.044	-0.057	0.766	1.687	0.092

Figure 5. Forest plot of changes in endurance performance (**A**) and anaerobic performance (**B**), in athletes participating in plyometric jump training (PJT) compared to controls. Values shown are effect sizes (Hedges's g) with 95% confidence intervals (CI). The size of the plotted squares reflects the statistical weight of the study. Split figure and place (**B**) under (**A**) to fit on the page.

3.10. Meta-Analysis Results for Anaerobic Performance

Three studies provided data for anaerobic performance, involving five experimental and three control groups (pooled $n = 89$). There was a non-significant effect of PJT on anaerobic performance (ES = 0.36; 95% CI = −0.06 to 0.77; $p = 0.092$; $I^2 = 0.0\%$; Egger's test $p = 0.121$; Figure 5B). The relative weight of each study in the analysis ranged from 13.5% to 39.2%. Of note, from a pool of 48 potential moderator analyses, due to a limited number of studies (i.e., <3 per moderator), only six moderator analyses were possible (as indicated above).

3.11. Adverse Effects

None of the included studies reported soreness, pain, fatigue, injury, damage, or adverse effects related to the PJT intervention.

4. Discussion

Our findings revealed that PJT is effective in improving CMJ and DJ, kicking performance, speed on sprint and COD tests and endurance performance in female soccer players. A discussion of such findings follows below.

4.1. Countermovement Jump

A moderate increase in CMJ was observed. The positive effect of PJT on CMJ was previously noted in female athletes [28,35]. Similarly, a meta-analysis in male soccer players revealed also a significant effect of PJT on male athletes' vertical jump height performance [29]. Although the moderate ES can be considered consistent across the included studies, caution should be taken since one study [47] out of 10 resulted in an extremely large (ES = 4.027) effect, even though the sample was composed by Spanish National Women's First Division players who are supposed to be highly trained. Similarly, Stojanović et al. [35] showed a large effect in female athletes, but also warned about the presence of one study (out of seven) reporting uncommon large gains (ES = 5.10) in CMJA. In both cases, discrepant effects can inflate results and reveal the necessity of performing more highly controlled studies to reach firm conclusions about the effectiveness of PJT on CMJ with and without arm swing in female soccer players. Nonetheless, after a moderator analysis for the current SRMA, excluding the study that yielded extremely large CMJ improvements [47], a significant improvement in CMJ (ES = 0.48; $p = 0.002$) was still observed. Therefore, PJT seems effective in the improvement of CMJ in female soccer players. Such improvement can be explained by the improved specific neural activation patterns and enhanced SSC utilization after training [17], even though CMJ can be considered a slow SSC movement [53].

4.2. Countermovement Jump with Arm Swing

Although two out of three studies demonstrated an improvement in CMJA performance, in contrast to CMJ, the CMJA increase was not significant. Although three studies provided data for the current meta-analysis, complying with previous recommendations for robust meta-analysis [54–56], the interpretation of results derived from such a limited number of studies should be performed with caution. Firstly, in the study of Ramirez-Campillo et al. [44] and Rubley et al. [46] the participants in the experimental groups had an increase in performance, while the soccer players in the control groups reduced their performance. In contrast, in the study of Siegler et al. [48] the participants from both the PJT and control groups exhibited an improved performance, with a greater improvement observed in the control group, which may explain the negative ES in Figure 2B for the Siegler et al. study [48]. Of note, in the study of Rubley et al. [46], one PJT session was applied each week, in the study of Ramirez-Campillo et al. [44] two sessions per week, whereas in the Siegler et al. study [48] up to three sessions were applied each week. According to a previous meta-analysis in female soccer players, PJT interventions with two or more sessions per week produced a moderate effect on vertical jump height (ES = 0.8), while those with less than two sessions per week produced a large effect (ES = 1.47). Is unlikely that overtraining occurred in the Siegler et al. study [48], since an improvement in performance was observed (similar to the control group). However, the possibility that a reduced frequency of PJT may induce an optimization of the training process, with optimal and efficient training volumes, particularly in female soccer players [50], should be further explored.

The lack of a significant increase in CMJA among female soccer players after PJT is in contrast with the findings from a previous meta-analysis in male adult soccer players, were a significant effect of PJT was noted for vertical jump height [29]. Further, among youth male soccer players a PJT meta-analysis revealed a significant increase in vertical jump height (i.e., CMJ and CMJA) [57]. Future studies may elucidate if different biological mechanisms or if differences in methodological issues between published studies (e.g., training duration; testing procedures) underlay the apparent different responses to PJT between male and female soccer athletes.

4.3. Drop Jump

A moderate improvement was noted for DJ (ES = 0.43; $p = 0.012$). Improvements in DJ (i.e., reactive strength) may be associated with several neuromechanically factors [17,58]. Of particular relevance may be the increased musculotendinous stiffness associated to DJ improvements after PJT [17,59]. Such an increase may be associated with a better running economy [60], particularly at high speeds [61], commonly occurring during female soccer matches [5]. In addition, a better DJ performance may reflect an improved ability to tolerate greater eccentric forces and improved concentric RFD [17,20,62,63] which may contribute to several key soccer-specific movements such as change of direction ability, to better tolerate landings after a jump, and explosive actions such as sprinting capabilities [5,59,64,65].

Of note, the improvement in DJ performance in our meta-analysis (ES = 0.43) was lower to that observed in a previous meta-analysis that analyzed the effects of PJT on DJ performance (ES = 0.66) [30]. Moreover, in the aforementioned meta-analyses females improved less their jumping performance (ES = 0.5) compared to males (ES = 0.8) [30]. Moreover, male soccer players showed a large improvement in DJ performance after a PJT intervention (ES=1.6), while female athletes showed only a moderate improvement (ES=0.6) [44]. Independent from the magnitude of the response to PJT in female compared to male soccer players, future studies should elucidate the mechanisms underlying the physical fitness improvements in soccer players after PJT.

4.4. Kicking Performance

A very large improvement was noted for kicking performance. Kicking performance is a key ability in soccer, involved in key moments of a match, such as passing actions and scoring goals [5]. Indeed, shooting performance is related with match success and league positioning [66–68]. Therefore, improvements in kicking performance after PJT may be important for female soccer players. Such improvement after PJT might be mediated through neuromechanically adaptations, including increased force and rate of force development [17,69,70]. However, from three studies that provided data for kicking performance, one [47] yielded an improvement value that was unusually high (ES = 5.71). Such result may be related to low SD values reported by the authors, probably due to the high performance level (Spanish National Women's First Division) of the athletes involved in the study. Independent of this, the fact that high-level athletes achieved an improvement in kicking performance of ~Δ12% [47] after PJT should be highlighted. In the study of Rubley et al. [46] a very large improvement was noted (ES = 2.91; Δ22%). Such improvement may not be uncommon among youth players. Indeed, in the study of Rubley et al. [46], youth (age = 13.4 y) athletes were included. In a previous work [71] with youth (male) soccer players (age = 11.8 y), an improvement of ES=1.83 was noted for kicking performance, after a PJT with a comparable load (i.e., 8-16 sessions) as in the study of Rubley et al. [46]. In this sense, soccer skills (e.g., kicking) may be particularly improved at certain age [72], such as in the study by Rubley et al. [46], whereas adult (female) athletes may achieve comparatively less improvements (ES = 0.42–0.45) [50]. When the total volume of PJT was compared between studies, values of 810 jumps [50] 1680 jumps [46] and 3240 jumps were noted. [47]. Whether such volume difference may explain the different magnitude of observed improvements deserves to be examined in future PJT research with female soccer players. Such research may allow to stablish optimal volumes for both increased performance and reduced injury risk [10,73].

4.5. Linear Sprint

Linear sprint performance obtained a moderate improvement after PJT. Considering the high frequency of short-distance (<30 m) sprints occurring in soccer matches, improving sprinting ability may increase the probability to winning ball possession and stand out from other players [5,74]. Interestingly, the studies analyzed in this SRMA provided data for 15 m to 20 m sprints (one study provided data for 30 m sprint). Therefore, improvements in linear sprinting probably reflect improved acceleration capabilities [75–77]. Considering the relevance of the force-velocity spectrum parameters (i.e., force, power and velocity) during sprinting among soccer players [78], it seems plausible that an optimization of the force-velocity spectrum may help to explain the improvements in sprinting after PJT. Indeed, PJT may provide positive adaptations in the force-velocity spectrum [79,80].

Compared to male soccer players, there are some contrast findings in the literature. For example, as in the current meta-analysis, a previous meta-analysis conducted with youth male soccer players observed significant improvements in linear sprint performance after PJT in distances between 5 m, 10 m, 20 m, 30 m, but not 40 m [57]. On the other side, a previous meta-analysis conducted with adult male soccer players observed non-significant improvements in linear sprint performance after PJT in distances between 5, 10, 15 and 30 m, with a significant improvement noted only for 20 m [29]. Future studies may elucidate how sex and maturity might interact to moderate the effects of PJT on linear sprint performance among soccer players.

4.6. Change of Direction Speed

A moderate improvement in COD was observed after PJT. Considering the relevance of COD performance among female soccer players (e.g., 1,336–1,529 movement changes during a match) [81], such improvement may reflect an advantage during competitive matches (e.g., increased goal chances) [2]. Neuromechanically adaptations such as greater muscle activation of the knee flexors (eccentric phase) and extensors (concentric or propulsive phase) [17,82,83] may favor COD improvements [64]. This may allow greater absorption of forces and increase ground reaction force production required in task execution of the COD [84]. Additionally, improvements in plyometric ability may also explain the improved COD capability [85].

The improvement in COD performance among female soccer players is in contrast with the result observed in male soccer players. Indeed, no improvement was noted in COD assessed with the *t* test and the zig-zag test in a previous PJT meta-analysis conducted in male soccer players [29]. Of note, in our meta-analysis female soccer players were assessed for COD performance with the *t* test and the zig-zag test, although also with the Illinois agility test, the latter requiring longer distances. Future studies may elucidate if differences in measurement protocols or sex-related differences might explain the apparently contrast COD performance changes after PJT between male and female soccer players.

4.7. Endurance

Endurance performance obtained a moderate improvement after PJT. Similarly, a meta-analysis revealed a significant effect of PJT on male soccer players endurance capability [29]. Improvements in endurance capacity has shown significant correlations with repeat sprint ability (RSA) [86], the latter positively associated with match-play performance [87]. These improvements could be explained due to better phosphocreatine resynthesize rate [87]. Therefore, increments of absolute strength would result enhancement in economy running due to a reduced recruitment of higher threshold motor units, producing a more economical behavior [88]. Resistance training methods with an emphasis on PJT and eccentric contractions could be advantageous for female soccer players to improve neuromuscular performance such as maximal strength, tendon stiffness and rate of force development.

4.8. Anaerobic Performance

Regarding anaerobic performance, most studies addressed the effects of PJT on performance during the repeated-sprint ability (RSA) test, and no meaningful result was obtained while comparing the experimental groups with controls. Though maximal sprinting speed is one of the performance composites of anaerobic performance during repeated sprints [89] other factors affecting recovery between sprints and neuromuscular performance maintenance are key to improve RSA [89–91]. Therefore, PJT may have limited potential to improve anaerobic performance, especially when endurance and fatigue-resistance factors need to be changed in order to induce adaptation. However, compared to controls, the PJT groups achieved an ES = 0.36 ($p = 0.092$). This effect probably is related to the significant and moderate effect of PJT on maximal linear sprinting ability in female soccer players.

Current findings are difficult to compare with those obtained in male soccer players, as most PJT studies that included RSA assessments included youth male soccer players. In one of such studies, youth males (age, 12.7 years; APHV, −1.3), after 8 weeks of jump training, improved (2.1–2.5%; ES = 0.2–1.6) best and total RSA times [92]. Also, in male youth soccer players (age, 13.6 years; Tanner stage II-III) small (0.7–0.8%) improvements were noted in best and mean RSA times after 6 weeks of training [93]. Relatedly, in young adult soccer players (age, 18.4 years), 8 weeks of training induced an improvement (1.4%; ES = 0.85) in mean RSA with COD time (1.4%; ES = 0.85) and related fatigue index (27.8%; ES = 0.91) [94]. Future studies may elucidate if differences in measurement protocols (i.e., RSA vs RSA-COD), players biological maturity or sex-related differences might explain the apparently contrast findings between our results and those previously reported for RSA changes after PJT for male soccer players.

4.9. Potential Limitations

Some potential limitations of this SRMA should be acknowledged. Firstly, additional analyses were not always possible as <3 studies were available for at least one of the moderators. Additionally, the use of the median split technique may induce residual confounding and reduced statistical power [95]. Moreover, a meta-regression was not possible due to reduced number of studies available. Further, even though the included studies did not specify any adverse events associated with the PJT intervention, it is unclear if there was an attempt by the researchers to comprehensively record all possible adverse events. Therefore, future studies are encouraged to describe with more detailed data about possible injuries, pain and/ or any other potential adverse effects, as this would expand our knowledge on the safety of PJT. Finally, only two of the included studies recruited youth female soccer players, and their biological maturity was not described. Considering the potential of biological maturity as a moderator of the effects of PJT on youth female physical fitness adaptations [96–98], future studies should strive for the inclusion of biological maturity description among youth female soccer players.

4.10. Practical Applications Derived from the Systematic Review

According to the results of our meta-analysis female soccer players should incorporate PJT programs into their regular training schedules in order to achieve small-large improvements in several physical fitness measures of key relevance in soccer, including jumping, sprinting, kicking, change of direction speed, and endurance. According to our systematic review results, PJT is effective in both youth and adult female soccer players, (age range: 13–27 years), with or without previous experience in PJT, from amateur to professional level. The PJT among the included studies in this meta-analysis proved to be safe, with no injuries reported. Indeed, PJT may be considered an integral part of neuromuscular training programs focused on injury prevention [9,99].

Regarding the characteristics of effective PJT interventions, it seems that a training frequency of 1-3 sessions per week, during 6–12 weeks, with a maximal- near-maximal intensity, is an adequate stimulus to boost physical fitness. Most studies incorporated some form of drop-like jump, although all the studies included different types of jump drills into their programs. The total number of jumps range between 800 up to 5620 jumps. Caution is warranted when high volume of PJT is prescribed. Indeed, a high volume of PJT may increase the injury risk among female soccer players [73]. As a moderate volume of PJT may be as effective compared to a program with a greater volume [50,100] a moderate-volume of PJT is advised, particularly during initial stages of PJT, in those unexperienced with PJT, poor technical ability, and reduced ability to cope with the eccentric forces associated to jumping drills. In line with the volume of PJT, most studies in the meta-analysis did not incorporated a taper. Such strategy may allow to boost performance before important competitions, thus its incorporation may be adequate, particularly with a high-volume PJT program [101].

Rest between sets range from 30 s up to 300 s, and for inter-repetition rest values from 5 up to 60 s were noticed. As 30 s and 120 s of inter-set rest are equally effective in order to allow significant physical fitness improvements after PJT among soccer players [102], and 15 s of inter-repetition rest is adequate to recover between maximal jumping efforts [103], in order to reduce the total duration of a PJT session, coaches may consider values from the low spectrum of above mentioned rest intervals. The minimum inter-session rest was 48 h among the included studies, which seems a common an adequate minimal recovery time between sessions.

A grass surface was the most common type of surface used among the included studies, which seems to be in line with the training principle of specificity, as soccer players usually train and compete on this type of surface. All the studies included in this meta-analysis incorporated a progressive overload, either in the form of volume, type of drill (e.g., two leg, progressing toward one leg), intensity, or a combination of these. Of note, most studies incorporated PJT during the in-season period, demonstrating that during such period of the season significant improvement in relevant physical fitness measures are possible among female soccer players, which may boost performance during important competitive match dates, thus increasing chances to achieve a better league positioning [4].

5. Conclusions

Several key physical fitness traits for female soccer players may be improved after PJT, including CMJ, DJ, kicking performance, linear speed, COD speed, and aerobic endurance performance, without changing anaerobic performance. Such improvements may be expected after PJT interventions with 6 or more weeks of duration, and among players with different chronological age, competitive level and soccer experience.

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Review

Sex Differences in Swimming Disciplines—Can Women Outperform Men in Swimming?

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Abstract: In recent years, the interest of female dominance in long-distance swimming has grown where several newspaper articles have been published speculating about female performance and dominance—especially in open-water ultra-distance swimming. The aim of this narrative review is to review the scientific literature regarding the difference between the sexes for all swimming strokes (i.e., butterfly, backstroke, breaststroke, freestyle and individual medley), different distances (i.e., from sprint to ultra-distances), extreme conditions (i.e., cold water), different ages and swimming integrated in multi-sports disciplines, such as triathlon, in various age groups and over calendar years. The influence of various physiological, psychological, anthropometrical and biomechanical aspects to potentially explain the female dominance was also discussed. The data bases Scopus and PUBMED were searched by April 2020 for the terms 'sex–difference–swimming'. Long-distance open-water swimmers and pool swimmers of different ages and performance levels were mainly investigated. In open-water long-distance swimming events of the 'Triple Crown of Open Water Swimming' with the 'Catalina Channel Swim', the 'English Channel Swim' and the 'Manhattan Island Marathon Swim', women were about 0.06 km/h faster than men. In master swimmers (i.e., age groups 25–29 to 90–94 years) competing in the FINA (Fédération Internationale de Natation) World Championships in pool swimming in freestyle, backstroke, butterfly, breaststroke, individual medley and in 3000-m open-water swimming, women master swimmers appeared able to achieve similar performances as men in the oldest age groups (i.e., older than 75–80 years). In boys and girls aged 5–18 years—and listed in the all-time top 100 U.S. freestyle swimming performances from 50 m to 1500 m—the five fastest girls were faster than the five fastest boys until the age of ~10 years. After the age of 10 years, and until the age of 17 years, however, boys were increasingly faster than girls. Therefore, women tended to decrease the existing sex differences in specific age groups (i.e., younger than 10 years and older than 75–80 years) and swimming strokes in pool-swimming or even to overperform men in long-distance open-water swimming (distance of ~30 km), especially under extreme weather conditions (water colder than ~20 °C). Two main variables may explain why women can swim faster than men in open-water swimming events: (i) the long distance of around 30 km, (ii) and water colder than ~20 °C. Future studies may investigate more detailed (e.g., anthropometry) the very young (<10 years) and very old (>75–80 years) age groups in swimming

Keywords: gender difference; sex gap; swimming performance; swimming stroke; holistic approach

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1. Introduction

Swimming is a specific sports discipline which can be performed in a range of styles, usually referred to as ’strokes’ [1–4], over different lengths [5,6] and in both pools (i.e., indoor, outdoor) of different lengths (mainly 25 m and 50 m) and in open water (i.e., sea, lake, river) [7–10]. The different swimming strokes are butterfly [1], backstroke [2], breaststroke [3], freestyle [4] and individual medley referred to the combination of the four different strokes [11]. In addition to these individual events, four swimmers can take part in either a freestyle or medley relay. In pool-swimming competitions, the distances for butterfly, backstroke, breaststroke and freestyle usually include 50 m to 200 m, whereas individual medley is held over 200 m and 400 m [11]. In freestyle, the 800 m and the 1500 m are further race distances in pool-swimming [4,12]. Indoor-swimming events with a defined time limit (i.e., 12 h) are also held [13]. In open-water swimming, master swimmers most often compete in 3000 m [14], whereas 5 km [6,10], 10 km [10,15] and 25 km [10,15] races were held for elite swimmers. Open-water swimming events of different lengths in lakes and seas are held as solo swims [7]. Swimming is also part of multi-sports races like triathlons over different distances like the Olympic Distance triathlon [16,17], the half-Ironman [18], the Ironman [16,19] and longer triathlon race distances than the Ironman distance [20].

In recent years, the interest of female dominance in long-distance swimming grew where several newspaper articles were published speculating about the female performance and dominance especially in open-water ultra-distance swimming. In one newspaper article, the history of female performance in open-water swimming started with Gertrud Ederle in the ’English Channel Crossing’ and the female dominance in ’Manhattan Island Marathon Swim’. The author discussed the problem of comparing the fastest men and women in contrast to all men and women, leading to a different finding regarding male or female dominance. Particularly, the fastest men beat the fastest women, but that the average woman was faster than the average man [21]. In addition, another newspaper article describing the sex difference in swimming and running discussed the aspect of the fastest women and men [22]. A further newspaper article cites Steven Munatones, one of the world’s top experts on open-water swimming, reporting that the average female time was 33 min faster than the average male time in the 135 years of the ’English Channel Crossing’ [23]. It seemed that women were better in long-distance open-water swimming. A further newspaper article reported that the female swimmer Sarah Thomas was the first person in the world to cross the ’English Channel’ four times in a row without stopping [24]. In a further newspaper article, women were described winning also ultra-endurance races in cycling and running ahead of all men [25]. Moreover, a newspaper article reported that the fastest women ever were faster than the fastest men ever in both the ‘Catalina Channel Swim’ and in the ’Manhattan Island Marathon Swim’ held in the USA [26].

These descriptions lead to the intention to review existing literature to confirm or disprove these statements of female dominance in open-water swimming. Since sex-related differences including anthropometric characteristics, swimming energy, as well as stroking parameters have been previously reported [27], it would also be interesting to examine this ’sex gap’ as translated into swimming performance. Such information would have both theoretical and practical relevance. From a theoretical point of view, the sex difference in human performance has been a major topic in exercise physiology, and thus, researchers working in this area would benefit from new knowledge on sex differences in swimming. From a practical perspective, coaches usually working with both sexes could use sex difference in swimming to optimize the training of their athletes. Therefore, the aim of the present research was to review original studies on sex differences in swimming performance with regards to age, swimming strokes and race distance.

2. Method

The data bases Scopus and PUBMED were searched on April 2020 for the terms 'sex–difference—swimming'. The search in Scopus led to 239 entries, the search in PUBMED to 558 entries. We excluded case studies, case reports, animal studies, studies with divers and rowers and studies with patients. Although the newspaper articles primarily reported about outstanding female achievements in open-water long-distance swimming, we consider in this narrative review all scientific results upon differences between the sexes in swimming for all swimming strokes (i.e., butterfly, backstroke, breaststroke, freestyle and individual medley), distances (i.e., from sprint to ultra-distances), conditions (i.e., cold water), ages (i.e., youth and master swimmers) and swimming integrated in multi-sports disciplines such as triathlon.

3. Findings

3.1. Pool-Swimming

Swimming competitions are held in pool-swimming in short-course (i.e., 25 m or 25 yards) and long-course (i.e., 50 m) pools from 50 m to 200 m in the four different strokes such as butterfly, backstroke, breaststroke and freestyle [28,29], where freestyle races were also held for 400 m, 800 m and 1500 m [29]. Apart from distance-limited swimming races, also time-limited swimming events (i.e., 12 h) are performed in pool-swimming [13]. Studies investigated different populations such as elite swimmers competing at national and international level for different strokes and distances, youth and master swimmers [1–3,8,29–31]. In pool-swimming, it seemed that the sex difference varied with the distance of the events [29,30,32]. For elite swimmers competing in different strokes such as freestyle [28], butterfly [31], breaststroke [30] and individual medley [30], the sex difference decreased with increasing race distance. In long-distance pool-swimming such as a 12-h-swim, women were able to achieve a similar performance to men. In the 'Zurich 12-h Swim' held in Switzerland, the annual best performance did not differ between males (~38.3 km) and females (~34.4 km), respectively [8]. For master swimmers competing at the FINA World Championships in different age groups for different strokes and distances, men were faster than women for all strokes, distances and age groups except in 50–800-m freestyle for age groups 80–84 and 85–89 years [4], in 50–200-m butterfly for age group 90–94 years [1], in 200-m and 400-m individual medley for age groups 85–89 und 90–94 years [11], in 50–200-m breaststroke for age groups 90–94 and 95–99 years [3] and in 50–200-m backstroke for age groups 85–89, 90–94 and 95–99 years [2] where women achieved a similar performance than men. The disparate findings were explained by differences in performance level, race distance, stroke, age and sample size.

3.2. Open-Water Swimming

In addition to pool-swimming events, open-water swims were held as individual swims (i.e., solo swims in Channel Crossings without drafting) or competitions in long-distance swimming events up to 25 km in open-water swimming [10,15] where swimmers are allowed to swim in a group and where drafting is allowed. In addition, there were open-water swimming events of different lengths in lakes and seas [7] where drafting is also allowed.

In open-water swimming, the water temperature may be of importance for female dominance. For official swimming competitions held in heated pools, the FINA has established that water temperature shall be at 25 °C to 28 °C [33]. Apart from swimming competitions held in heated pools, swimming events were also held in open water such as rivers, lakes and seas. For open-water swimming events sanctioned by the FINA (i.e., World Cup races in 5 km, 10 km and 25 km), the FINA has established that the water temperature should be a minimum of 16 °C and a maximum of 31 °C [34]. In Channel Crossings like the 'English Channel Swim', swimmers face, however, a water temperature of 15 °C at the end of June, increasing to 18 °C by the beginning of September [35]. Water temperature seemed to have an influence on the performance of the swimmers. For open-water swimmers competing in the ‘Marathon Swim in Lake Zurich’, a 26.4 km open-water ultra-swim held in Switzerland, performance of the top swimmers was negatively related to water temperature [9].

Female performance was investigated for different solo swims where drafting is not possible. In these studies, different groups of different performance levels were investigated. It seemed that women were able to achieve a similar performance to men in solo swims in long-distance open-water swimming such as individual Channel Crossings [7,36,37], where water temperatures were generally below 20 °C. In lake swimming such as ‘Marathon Swim Lake Zurich’ with water temperatures at or warmer than 20 °C, women achieved a similar performance to men [9].

However, depending upon the investigated sample (i.e., the fastest woman/man, the three fastest women/men, the five fastest women/men, the ten fastest women/men, all women/men, the annual fastest women/men, the annual three fastest women/men), women were able to outperform men in long-distance open-water swimming [36–38]. In the ‘English Channel Crossing’, the overall female swim time of 13:16 h:min was not different compared to the overall male swim time of 13:35 h:min between 1875 and 2011 [7]. Although the fastest male swim time (6:57 h:min) during this period was 6.7% faster than the fastest female swim time (7:25 h:min), the sex difference in performance of the top three times was ~8.9% [7]. The fastest annual swim speed did not differ between men (~0.89 m/s) and women (~0.84 m/s) [8]. In the ’Triple Crown of Open Water Swimming’ with ’Catalina Channel Swim’, ’English Channel Swim’ and ’Manhattan Island Marathon Swim’, overall women were ~0.06 km/h faster than overall men [31]. However, women were ~0.07 km/h slower than men when considering the annual five fastest swimmers [37]. Analyses were also performed for the single events of the ’Triple Crown of Open Water Swimming’. In the ’Catalina Channel Swim’, the fastest woman ever was faster than the fastest man ever (~22 min) [36]. The three fastest women were faster than the three fastest men (~20 min), however, the difference reached no statistical significance [36]. The ten fastest women were ~1 min faster than the ten fastest men, however, also here, the difference reached no statistical significance [36]. However, the annual fastest women (~10:51 h:min) were ~52.9 min (~16%) faster than the annual fastest men [36]. In a further open-water event held in the USA, women were faster than men. In the ’Manhattan Island Marathon Swim’, the ten fastest women were ~12%–14% faster than the ten fastest men [38]. Open-water swimming is also held for master swimmers competing in 3000 m. In master swimmers competing at the FINA World Championships, men were faster than women for all age groups except age groups 75–79, 80–84 and 85–89 years where women achieved the same performance like men [14]. Not only the age, but also the distance may be of importance. It seemed that in shorter open-water swimming events with a higher water temperature, women have a disadvantage compared to men. In the ’Marathon Swim in Lake Zurich’, the male record was 2.3% faster than the female record. For the annual winners, men were ~11.5% faster [9].

Based on these observations, in most cases analyzed, there were no sex differences in performance during open-water swimming. In some instances, women were faster than men. However, a variety of parameters such as water temperature and distance can influence the outcome. Depending upon the sample size (i.e., the fastest woman/man, the three fastest women/men, the five fastest women/men, the ten fastest women/men, all women/men, the annual fastest women/men, the annual three fastest women/men) and the statistical approach (i.e., comparison of groups or comparison of changes over time), women were faster than men in this specific sports discipline.

3.3. Ice Swimming

Since 2009, ice swimming for 1 mile and 1 km is a new discipline in open-water swimming [39,40]. In this swimming discipline, water temperature must be colder than +5 °C. One may assume that female performance may be better when water temperature is very low. Performances of women and men were investigated for ’Ice Mile’ and ’1 km Ice event’ where the fastest men were faster than the fastest women in both events [41]. Obviously, women had no advantage in this cold water. In the ’Ice Mile’, variables such as calendar year, number of swims, water temperature and wind chill showed no relation to swimming speed for both women and men. Water temperature was not correlated to swimming speed in either 'Ice Mile' or '1 km Ice event' for both women and men [41]. Therefore, the limited data concerning ice swimming demonstrate that men have an advantage, compared to women, in this specific condition. Regarding the results from long-distance open-water swimming, the length of the event may be decisive. Future studies may investigate the body composition of open-water long-distance swimmers and ice swimmers. There may be a difference in body fat in the competitors in the two disciplines.

3.4. Age

Age is an important aspect regarding the sex difference in swimming performance. This aspect was investigated for different age groups, strokes and swimming distances. An analysis of sex differences in swimming speed for the top-10 World ranking (i.e.,1st–10th place), age group (25–89 years), and event distance from the world's top ten swimming times of both women and men in the World Championships showed that the sex difference in swimming speed increased with world record place and age [42]. Very recent studies investigated the performance trends and sex difference in swimming performance in master swimmers competing in the FINA World Championships in pool-swimming in freestyle [4], in backstroke [2], in butterfly [1], in breaststroke [3], in individual medley [11] in 3000-m open-water swimming [14] and for youth swimmers [43]. In butterfly [1], in breaststroke [3], in backstroke [2], in freestyle [4] and in individual medley [11], women were able to reduce the gap to men in different age groups. In 3000-m open-water swimming, however, women were not able to reduce the sex difference to men [14]. Consequently, the existing sex difference regarding all swimming strokes is evident during the youngest (i.e., 25 to 29 years) age groups, while for nearly all the rest of the age groups (i.e., 30 years and older), women tend to reduce this sex gap.

In addition, for youth swimmers, age is of importance. When boys and girls from the age of 5 to 18 years for 50 m to 1500 m from the all-time top 100 *U*.S. freestyle swimming performances were investigated, the top five girls were faster until the age of ~10 years than boys. After the age of 10 years, however, boys were increasingly faster than girls until the age of ~17 years [43]. Overall, female swimmers can beat male swimmers under the age of ~10 years and achieve almost the same performance as men in the highest age groups (i.e., older than ~75–80 years) depending upon the distance and the stroke.

A few studies have investigated the age effect in open-water swimming. The age of peak performance increased over calendar years in long-distance open-water swimming. In the 'Manhattan Island Marathon Swim', the age of the annual three fastest swimmers increased between 1983 and 2013 from ~28 to ~38 years for women and from ~23 to ~42 years for men [38]. In the 26.4 km open-water ultra-swim 'Marathon Swim in Lake Zurich', Switzerland, the mean age of the finishers during the period 1987–2011 was ~32.0 years for men and ~30.9 years for women. The mean age of finishers and the age of winners increased across the years for both sexes [9].

3.5. Sex Difference and Swimming Strokes

Some studies investigated the aspect of sex difference for different strokes. In pool-swimming competitions, athletes perform in the four strokes (i.e., butterfly, backstroke, breaststroke and freestyle) [28] as well as in the combination of all four strokes as individual medley [11]. There seem to be changes in the sex difference for the swimming strokes depending upon the distance and the performance level. For both 200-m and 400-m freestyle and individual medley, no sex difference was found between neither the two distances, nor between the two swimming strokes [44]. The sex differences were ~9.7% and ~7.1% in individual medley and ~10.1% and ~6.1% in freestyle, respectively [45]. For elite male and female butterfly and freestyle swimmers at national level, the sex difference in peak swimming speed was lower in butterfly than in freestyle [31,46]. For national and international breaststroke and freestyle swimmers, the sex differences in swimming speed increased over time for national swimmers, but not for international swimmers for freestyle, while the sex difference remained stable for both national and international breaststroke swimmers [30].

The disparate findings were explained by the different performance levels, the different distances and strokes and the different sample sizes.

3.6. Performance Level and Sex Difference in Performance

In some studies, the changes in sex difference over time were investigated for different levels of athletes (i.e., national level, international level) [44,47]. In swimmers competing at national and international level, the sex-related difference in swimming speed was greater for freestyle than for breaststroke in 50-m to 200-m race distances for national swimmers, but not for international swimmers. For both groups, the sex-related difference for both freestyle and breaststroke swimming speeds decreased with increasing race distance. The sex-related differences in performance were greater for freestyle than for breaststroke for swimmers at national level, but not for swimmers at international level [47]. The disparate findings were explained by differences in performance level, distance, stroke and sample size.

3.7. Changes in Swimming Performance Over Years

Some studies investigated the change in performance over calendar years [8,48]. There seem to be differences between sexes, disciplines, performance level and distances [8]. In the past, it has already been suggested that women would soon perform better than men in swimming. In 1977, it was reported that women were gaining on their male counterparts at the rate of 0.45% a year in the 100-yard freestyle [48]. It was assumed that with that rate of improvement national level women may catch up with male counterparts by the year 2003. Likewise, in the 1650-yard freestyle, women were gaining on men, but at a slower rate of improvement of ~0.155%. It was assumed that it would take the women ~51 years to catch up to the men. The authors found that race times in women were improving at a rate faster than race times in men, but at some time in the future the rate of growth would probably stabilize for both sexes [48]. While at present that assumption, regarding the specific race distance (i.e., 100 yards), has not been completely fulfilled, it remains to be seen if a performance plateau would allow women to outperform men.

Regarding newer studies, female performance has improved over calendar years in some instances [8]. These analyses of changes in performance over the years have been performed for elite pool swimmers [5], for open-water swimmers [7,8] and for master swimmers competing in freestyle [4], backstroke [2], butterfly [1], breaststroke [3], individual medley [11] and in 3000-m open-water swimming [14]. In pool-swimming, performance was improved for most distances in both elite and master swimmers in backstroke [49], freestyle [31,46], breaststroke [30], butterfly [1,50], individual medley [11,30] and in 3000-m open-water swimming [14].

Some studies have investigated open-water swimming and showed that performance changed over years. For women and men crossing the 'Catalina Channel' between 1927 and 2014, performance decreased nonlinearly in the annual fastest men and women [36]. In the 'Manhattan Island Marathon Swim', race times of the annual three fastest women and men did not differ between sexes and remained stable across the years [38]. In the 'Maratona del Golfo Capri-Napoli', race times of the annual fastest swimmers decreased linearly for women and for men from 1954 to 2013 from 39.2% to 4.7% [51]. For the annual top three swimmers, race times decreased linearly between 1963 and 2013 for women and for men from 38.2% ± 14.0% to 6.0% ± 1.0% [51]. In the 'English Channel Crossing', the performance of the annual top three swimmers showed no changes either both females or males over the last 36 years and the sex difference remained unchanged at ~12.5% over the years [7]. In the 'English Channel Crossing', performance increased progressively for both sexes, but was lower for female than for male athletes from 1900 to 2010 [8].

A different kind of events was the FINA races which were not held as solo events and swimmers could draft. For elite male and female swimmers competing at the FINA World Cup events of 5 km, 10 km and 25 km events, swimming speed of the annual ten fastest women decreased at 5 km and at 25 km, while it increased at 10 km. For the annual ten fastest men, peak swimming speed decreased at

5 km, while it remained unchanged at both 10 km and 25 km [10]. In the FINA 10 km competitions (i.e., World Cup races, European Championships, World Championships and Olympic Games) held between 2008 and 2012, swimming speed of the fastest women and men showed no changes across the years. Performance of the top ten female swimmers per event remained stable across calendar years. The top ten male swimmers per event showed a decrease in performance over years, even though swimming speed in the first race (i.e., January 2008, 1.40 m/s) was slower than swimming speed in the last race (i.e., October 2012, 1.50 m/s) [50]. The disparate findings were explained by the different performance levels, the different distances and strokes, the different ages, the different periods of time and the different sample sizes.

3.8. Swimming in Multi-Sports Disciplines Like Triathlons

Swimming is the first segment of a triathlon event, followed by cycling and then running [52]. Several studies investigated the trends in performance and the sex difference in performance in swimming in triathlons of different lengths such as the Olympic distance triathlon (i.e., 1.5 km swimming, 40 km cycling and 10 km running) [16,17,53], the Ironman distance triathlon (i.e., 3.8 km swimming, 180 km cycling and 42.195 km running) [16] and ultra-triathlon distances longer than the Ironman distance [20,54,55].

In Olympic distance triathletes competing in the 'Zürich Triathlon' in Switzerland from 2000 to 2010, the sex difference in swimming was 15.2% for the top five triathletes overall [17]. For the world's best triathletes at the ITU (International Triathlon Union) World Triathlon Series during the 2009–2012 period including the 2012 London Olympic Games, swim times and the sex difference in swimming remained unchanged [53].

For longer triathlon distances than the Olympic distance, sex difference has been investigated for the Ironman distance [19,56,57] and longer triathlon distances from 2× to 10× the Ironman distance [20,57]. It was shown that women improved swimming performance and closed the gap to men. In 'Ironman Hawaii', the overall top ten men finishers improved their swimming performance between 1983 and 2012. The sex difference remained unchanged over the years at ~12.5% [19]. For the annual three best finishers in 'Ironman Hawaii', the sex difference decreased nonlinearly in swimming between 1978 and 2013 [57]. In 'Isklar Norseman Xtreme Triathlon' held over the Ironman distance, athletes swim at a water temperature of ~13–15 °C. Men were faster than the women in cycling, but not in swimming, running or overall race time. Across years, women improved their performance in swimming and both women and men improved their performance in cycling and in overall race time. In running, however, neither women nor men improved [3].

Different findings were, however, reported for longer triathlon distances. In Double Iron ultra-triathlon (i.e., 7.6 km swimming, 360 km cycling and 84.4 km running), men (2:36 h:min) were ~8 min faster than women (2:44 h:min) [20]. For triathlon distances from the Ironman distance in 'Ironman Hawaii' to the Double Deca Iron ultra-triathlon distance (i.e., 76 km swimming, 3600 km cycling and 840 km running), the sex difference in performance showed no change with increasing race distance with the exception for the swimming split where the sex difference increased with increasing race distance for the three fastest ever [57].

Regarding triathlon swimming performance, in most cases, the sex difference tends to remain unchanged over the years. However, as previously mentioned, women can outperform men in specific triathlon races under more extreme conditions (i.e., water temperature of ~13–15 °C).

3.9. The Change in Sex Difference Over Years

In the same way where swimming performance can change over years, also the sex difference in swimming performance may change over years [10,44,58,59]. In some instances, women reduced the gap to men [59,60], in others not [10,28,44,53]. In these studies, pool-swimmers of sub-elite and elite level [28,44], open-water long-distance swimmers [59], master swimmers up to very high ages and swimmers in triathlons [20,54,55] were analyzed. In pool-swimming, the changes in sex difference differ over time regarding the distance, the age and the discipline [14,28,31,47]. According to this information, the sex difference in relation to pool-swimming and open-water swimming performance is largely dependent on the parameters analyzed in this review (i.e., competition level, swimming stroke and distance).

Several studies have examined the variation in sex difference over calendar years in open-water swimming events. In some events, the sex difference decreased, and the women reduced the gap to men [32,39,56], but in others not [9]. In women and men crossing the 'Catalina Channel', the sex difference for all women and men decreased linearly between 1927 and 2014 from 52.4% to 7.1% [36]. The decrease of sex difference was linear suggesting that women continuously reduced the sex difference to men [36]. In the 36 km 'Maratona del Golfo Capri-Napoli', the sex difference for the annual fastest swimmers, decreased linearly from 39.2% to 4.7% from 1955 to 2013 [51]. For the annual three fastest swimmers, the sex difference in performance decreased linearly from 38.2% ± 14.0% to 6.0% ± 1.0% from 1963 to 2013 [51]. Again, in this event, the linear change in both race times and sex differences indicates that women could achieve men's performance or even to perform better than men in the near future in this event [51]. In 'La Traversée Internationale du Lac St-Jean' (32 km) held between 1955 and 2012 in Canada, the sex difference remained unchanged over years for the annual fastest women and men at 8.8% [58]. For the annual three fastest women and men, the sex difference decreased across years (1975–2011) from 14.4% ± 11.0% to 3.7% ± 1.4% [58]. Overall, most studies found that women reduced the gap to men over years in open-water swimming.

Differences were found in open-water swimming where women are allowed to draft behind men. For elite male and female swimmers competing in 5 km, 10 km and 25 km open-water FINA World Cup races, elite female swimmers improved their performance in 10 km, but impaired performance in 25 km, leading to a linear decrease in sex difference in 10 km and a linear increase in sex difference in 25 km. The linear change in sex differences suggests that women will improve in the near future in 10 km, but not in 25 km [10]. In elite open-water swimmers competing at FINA 10 km races, the mean sex difference in performance for the fastest swimmers was stable across years [50].

In long triathlon races, women were not able to close the gap to men. For triathlon races longer than the Ironman race distance, the sex difference in swimming showed no change over years in either Double Iron ultra-triathlon [54] or in 'Ultraman Hawaii' [55]. In Double Iron ultra-triathlon races, the swimming times remained unchanged across years with an unchanged sex difference for the annual three fastest women and men [20]. Potential explanations as to why sex differences decreased over the years or not could be the selected period of time [44,53,58], the level of the investigated athletes (i.e., annual fastest, annual ten fastest, national level, international level, etc.) [44,58], and/or whether the swimmers were solo swimmer, competing in a drafting race or triathletes. When the sex difference showed no change over time, the investigated period of time was most likely too short [53].

3.10. The Influence of Swimwear on Performance

Sex differences among swimmers using a wetsuit have also been investigated [61]. A wetsuit can be mainly used in the swim split in triathlon races [61] or in long-distance open-water swimming races [62]. In triathletes, the effect of a wetsuit on lighter female swimmers was no different than the effect on heavier male swimmers [61]. Swimming with or without a wetsuit shows a difference between the sexes [63,64]. When swimming speed was compared among women and men with or without wetsuit over different distances, wearing a wetsuit improved swimming speed for both women and men, but the benefit of the use of wetsuits depends on additional factors such as race distance. Women may be favored from wearing a wetsuit more than men in longer ultra-distance races of open-water swimming [63]. It has also been shown that high-tech swimsuits gave more pronounced advantage to men than women and for low resistance as compared with high resistance swimming strokes [64]. Yet, the new rules on swimsuits, in effect since 2010, should also be considered when analyzing swimming performance.

4. Potential Explanations for The Female Dominance in Long-Distance Open Water Swimming and Age Group Swimmers

Based on these results, we may conclude that women were able to outperform men in swimming in solo, long-distance swimming events held in water temperatures between 15 °C and 20 °C. Women were also able to achieve the same performance as men in all distances and disciplines of pool swimming at younger ages (i.e., younger than ~10 years) and older ages (i.e., from ~80 years onwards).

Two main variables may explain why women can outperform men in open-water swimming: (i) the long distance of ~30 km, (ii) and water colder than ~20 °C. Potential explanations for the finding that women can achieve a better performance than men can be attributed to differences in anthropometric characteristics such as body composition [65], body weight [66], body fat [67–70], lean body mass [71,72], body height [73,74], muscle thickness [75] and muscle size [72]. Other possible explanations were differences in swimming biomechanics such as kinematic parameters [76], arm coordination and arm–leg coordination [76–78], energetic cost, differences in swimming economy and swimming efficiency [79–81], gliding [82], body roll [83], shoulder flexibility [84], trunk flexibility [85], knee flexibility [86,87], propelling efficiency [88]. Beyond that, additional differences in motivational aspects [89–96], physiology [97–100] and biochemistry [101–103], recovery [104] and injury prevalence [105,106] can be pointed out. We found, however, no potential explanation for the youth swimmers.

A very likely explanation that women were faster than men in specific open-water ultra-distance events is the fact that female swimmers have more body fat than male swimmers [67] leading to better insulation against the cold and better buoyancy for long swimming distances [69]. It is well known that sex is associated with waist-to-hip ratio and body fat percentage in swimmers [107]. Due to the higher body fat, female swimmers have a different body shape compared to male swimmers. Male swimmers have a more central distribution of fat when compared to females, where body fat is built up in the region of legs [108]. In nonstationary swimming with changing velocity, water around the swimmer is set in motion which can be thought of as an added mass of water. Female swimmers have a lower added mass and relative added mass than male swimmers suggesting that sex differences in body shape may be associated with added mass [109].

In ultra-distances of open-water swimming, different anthropometric characteristics such as body height, body mass index (BMI), length of arm and training characteristics (e.g., swimming speed) were associated with performance for men. For women, swimming speed during training was associated with performance, but not anthropometric characteristics. Considering all variables for men, BMI and swimming speed during training were related to race time, but not for women [110]. Differences in anthropometric characteristics do exist between female and male swimmers [111,112]. It was shown that differences in height, arm span, skinfold thicknesses (e.g., triceps, subscapular, crista iliaca, ileo-spinal, abdominal, thigh, leg, sum of skinfolds), bi-acromial-bi-iliac index, bone body mass, muscle and fat, ectomorphy and endomorphy exist [111]. In another study, elite female youth swimmers had greater skinfolds at triceps, suprailiac and abdominal site. Endomorphic somatotype was twofold greater among elite female compared to elite male youth swimmers [112].

Although women could outperform men in certain swimming disciplines, in general, elite men were faster than elite women [113]. The sex gap in swimming performance seemed to remain stable in shorter distances. In Olympic Trial swimming from 1972 to 2016, the performance gap in swimming remained at ~8% [114]. The plateau during these ~40 years in the performance gap highlighted the role of biologic background (e.g., longer limbs, larger muscle mass, greater aerobic capacity and lower fat mass) on race time. Current evidence indicates that women will not swim as fast as men in Olympic events, which justifies sex segregation in these individual sport disciplines [114]. Men have an advantage of larger body size and muscle mass, a superior ventilation function and anaerobic and aerobic energy transfer systems. It is well known that male swimmers have a higher maximum oxygen uptake than female swimmers among both younger [97] and older age groups [98–100]. It is also known that ventilation functions including forced vital capacity (FVC), forced expiratory volume in one second (FEV1), FEV1/FVC and mandatory minute ventilation (MMV) were superior in male athletes to those in females [115]. Furthermore, the average diameter of muscle fibers was larger in men than in women [116]. Therefore, it is not a surprise that male youth swimmers show increased power values in both their legs and arms [117]. Finally, although male swimmers have a higher muscle mass, maximum handgrip isometric strength values correlated with swimming race time, especially in female swimmers [118]. Compared to men, women have an enhanced ability to oxidize fat, superior hydrodynamics and more even pacing, which provide advantage, especially during prolonged swimming [113]. Regarding pacing in pool- swimming, an effect of sex on lap time in master swimmers competing in 100, 200, 400 and 800-m freestyle at the World Championships suggested greater changes of pacing in women than in men [12].

Apart from anthropometric and/or physiological differences, psychological differences may also explain female dominance in certain swimming disciplines [91–95,119–121]. Female swimmers show differences to male swimmers regarding mental toughness [92]. Competitive female swimmers were emotionally secure, physically healthy and reasonably contented with their present social status. For these women, the emotional, social, and physical costs were definitely worth the sacrifices [120].

Effects of cultural and sociopolitical norms and outdated stereotypes (i.e., reduced opportunity to participate and compete in sports) that influence the number of female competitors, particularly in earlier years, should also be considered. Women have traditionally been under-represented in sports. In the 19th century, women were engaged in non-competitive recreational activities, but not in competitive sport. In 1971 in the United States, less than 7% of high school varsity athletes were female. Title IX was set in the United States in 1972 with the aim of providing equal treatment in sports, regardless of sex and increasing the number of women in sports [122]. Even as recently as 1979 in Brazil, it was illegal for women to play football [123]. According to Capranica et al., there were still some countries in the 2012 London Olympic Games that did not have a female in their delegation [124]. There was an increase in the number of female athletes in all sports over the last century, including swimming, that likely contributed to the reduction in the gap in performance between the sexes [125].

A potential explanation for the improved performance of older women, especially in pool-swimming, is the fact that the age of peak performance has increased in women since the 1980s. When 116 years from the first Olympic Games (1898) to the 2014 Olympic Games were analyzed, regarding the ages at which peak performance was observed, peak performance ages in women have increased consistently since the 1980s in all the athletic events examined (i.e., track and field, swimming, rowing and ice-skating events). When the age of peak female performance increased, it became similar to the age of male performance in many events. In the last 20 to 30 years, the age of peak female athletic performance increased, but not the age of male athletic performance [126]. Age in open-water swimmers may also be of importance. In the 'Manhattan Marathon Island Swim', the age of peak performance of ultra-distance swimmers has changed across the last decades, with the fastest swimmers getting older between 1983 and 2013. During this period, the age of the three fastest swimmers raised from 28 to 38 years in women and from 23 to 42 years in men [38]. The fact that female competitors were younger than their male counterparts likely had an effect on performance. Age was also of importance in other endurance athletes such as ultra-marathon runners [127,128]. In recent studies investigating ultra-marathoners [128] and athletes competing in tower running [129], women were able to close the gap to men [128] or to even outperform men [130]. In 50- and 100-mile ultra-marathoners, the sex difference in running performance decreased with increasing age and was smaller in the longer (100 miles) compared to the shorter (50 miles) distance [128]. This finding may be explained by the lower participation of women in longer ultra-marathon races. Also, in tower running (stair climbing), women were able to beat men in specific situations, e.g., in smaller buildings with less than 600 stairs for younger (30–59 years) and older (>69 years) age groups, in buildings with 1600–220 stairs for older ages (>69 years) and in buildings with more than 2200 stairs for younger (<20 years) and older (60–69 years) age groups [130]. With increasing age, experience may improve and race tactics may become better.

A further aspect was the use of technical wetsuits in pool-swimming. In the 2009 FINA World Championship held in Rome, a total of 43 world records were set. Men set new world records in 15 of those events, whereas women did the same in 17 events. Each of the men's world records and 14 of the 17 women's records still stood. In the past, these world records had not been broken in such a short period of time. There was much speculation that full-body, polyurethane, technical swimsuits were the reason for the improvement in world records. Further analysis led the FINA to institute new rules on 1st January 2010, that limited the types of technical swimsuits that could be worn by athletes. No long-course world record has been broken since then [129]. A problem in this field was, however, the fact that most of the considered studies used different numbers of subjects, different populations, different analyses leading to different significance levels. This problem cannot be eliminated, but it must be considered that depending on the sample and the analysis used, a significant difference between female and male performance can result or not.

5. Conclusions

The collective data presented in this review indicate that existing sex differences in swimming performance showed a generally diminishing trend that was more profound during the longer pool-races, for all swimming strokes. Age-related variations were also reported in both pool and open-water swimming, as sex difference mainly remained for the younger age groups. However, female athletes in very young age groups (10 years and younger) and very old age groups (75–80 years and older) outscored their male counterparts. When analyzing triathlon swimming performance, the sex gap remained stable during shorter and longer swim distances, except for events under more extreme water temperatures where women can outperform men. Finally, the sex difference in pool-swimming performance over time showed variations depending on the swimming stroke, distance and the competitive level. Regarding open-water swimming over time, women seemed to continuously narrow the gap to men, especially in specific long-distances, where the assumption of outperforming men existed. In summary, women tended to decrease the existing sex differences in specific age groups (i.e., younger than 10 years and older than 75–80 years) and swimming strokes or even overperform men in long-distance open-water swimming (distance of around 30 km), especially under extreme weather conditions (water colder than ~20 °C). Future studies may investigate body compositions of the different age group swimmers in order to explain the sex differences for specific sports disciplines such as pool-swimming and long-distance open-water swimming.

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Case Report

Self-Selected Pacing During a World Record Attempt in 40 Ironman-Distance Triathlons in 40 Days

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Abstract: The present case study analyzed performance, pacing, and potential predictors in a self-paced world record attempt of a professional triathlete to finish 40 Ironman-distance triathlons within 40 days. Split times (i.e., swimming, cycling, running) and overall times, body weight, daily highest temperature, wind speed, energy expenditure, mean heart rate, and sleeping time were recorded. Non-linear regressions were applied to investigate changes in split and overall times across days. Multivariate regression analyses were performed to test which variables showed the greatest influence on the dependent variables cycling, running and overall time. The athlete completed the 40×Ironman distances in a total time of 444:22 h:min. He spent 50:26 h:min in swimming, 245:37 h:min in cycling, 137:17 h:min in running and 11:02 h:min in transition times. Swimming and cycling times became slower across days, whereas running times got faster until the 20th day and, thereafter, became slower until the 40th day. Overall times got slower until the 15th day, became faster to 31st, and started then to get slower until the end. Wind speed, previous day's race time and average heart race during cycling were significant independent variables influencing cycling time. Body weight and average heart rate during running were significant independent variables influencing running performance. Cycling performance, running performance, and body weight were significant independent variables influencing overall time. In summary, running time was influenced by body weight, cycling by wind speed, and overall time by both running and cycling performances.

Keywords: swimming; cycling; running; ultra-endurance; recovery

1. Introduction

Triathlon races of the classical Ironman distance (i.e., 3.8 km swimming, 180 km cycling, and 42.195 km running) [1] and ultra-triathlon races with multiple times the Ironman distance such as Double Iron ultra-triathlon (i.e., 7.6 km swimming, 360 km cycling, and 84.4 km running), Triple Iron ultra-triathlon (i.e., 11.4 km swimming, 540 km cycling and 126.6km running), Quintuple Iron ultra-triathlon (i.e., 19 km swimming, 900km cycling, and 221 km running) and Deca Iron ultra-triathlon (i.e., 38 km swimming, 1800 km cycling, and 422 km running) are of increasing popularity [2,3].

Pacing during endurance and ultra-endurance performance is very important for a successful race outcome. Different pacing strategies are known such as: negative pacing (i.e., the athlete becomes faster during the performance); all-out pacing (i.e., limited to extremely short performances of ≤30 s); positive pacing (i.e., the athlete becomes slower during the performance); even pacing (i.e., the performance is constant over time); parabolic-shaped pacing (i.e., speed increases and decreases); and variable pacing (i.e., change of negative and positive pacing) strategies [4].

In longer triathlon races, pacing has been investigated for both the cycling and running split in elite female and male Ironman triathletes, where both women and men adopted a positive pacing during both cycling and running splits [5]. In recent decades, the length of the triathlon race distances has increased. While distances of 2× to 10× Ironman are regularly held, only one official race of 30×Ironman-distance triathlons was ever held in autumn 2013 [6]. To date, only one athlete went beyond the 30 days in a self-paced event completing 33 Ironman-distance triathlons in 33 days also held in summer 2013 [7].

Ultra-endurance athletes are pushing their limits to go for the ultimate limit in endurance performance [8]. In ultra-running, the limit has most probably achieved with crossing a continent [9]. In the 'Trans Europe Foot Race 2009', a small sample of a few dozens of ultra-marathoners covered the distance of 4487 km from South Italy to North Cape [10]. However, the total distance of 4487 km within 64 days [11] was outperformed in 2016 by the French ultra-marathoner Patrick Maladin with 10,000 km in 100 days (99 days 4 h 12 min) with the daily performance of a 100 km ultra-marathon [12]. However, in 2019, he broke his own record while he crossed the United States of America from New York to Los Angeles (4801 km) within 46 days and Canada from Vancouver to Halifax (5931 km) within 56 days to complete the overall distance of 10,732 km within 102 days 18 h and 48 min [13].

Triathletes push their limits as much as the ultra-runners with completing daily an Ironman-distance triathlon for as many days as possible. While the longest scientifically verified self-paced event was 33 Ironman-distance triathlons in 33 days in summer 2013 [7], we present here the pacing in the first and only athlete to complete a scientifically verified (all data collected and open available for detailed analysis) self-paced world record attempt to finish 40 Ironman-distance triathlons within 40 days held in autumn 2019. We also investigated potential predictor variables for split disciplines (i.e., cycling and running) and overall performance.

2. Materials and Methods

2.1. Ethical Approval

This study was approved by the Institutional Review Board of Kanton St. Gallen, Switzerland, with a waiver of the requirement for informed consent of the participant as the study involved the analysis of publicly available data. Heart rates [14] and body weight [15] are free available from the blog of the athlete. The study was conducted in accordance with recognized ethical standards according to the Declaration of Helsinki adopted in 1964 and revised in 2013.

2.2. Athlete

The athlete (36 years old, body mass 74 kg, body height 1.78 m, body fat percentage 7.3%) is a professional world class level ultra-endurance athlete competing in ultra-marathon running and ultra-triathlon races. Until the start of the event, he had finished 17 official ultra-triathlon races. These were 7 Double Iron ultra-triathlons, 4 Triple Iron ultra-triathlons, 1 Quadruple Iron ultra-triathlon (4× Ironman in 4 days), 1 Quintuple Iron ultra-triathlon, 1 Deca Iron ultra-triathlon as 10× Ironman in 10 days and 1 Deca Iron triathlon with 38 km swimming, 1800 km cycling and 422 km running. Apart from the official races, he finished 2 self-paced ultra-triathlons (5× Ironman-distance in 5 days and 20× Ironman-distance in 20 days). The personal best times for the official races are: 19:42:57 h:min:s (Double Iron ultra-triathlon—official world record June 2019), 32:57:01 h:min:s (Triple Iron ultra-triathlon), 53:08:27 h:min:s (4×Ironman-distance triathlons in 4 days), 72:55:29 h:min:s (Quintuple Iron ultra-triathlon), 234:31:15 h:min:s (Deca Iron ultra-triathlon), and 108:48:58 h:min:s (10× Ironman-distance triathlons in 10 days). He has won 5 races and come to second place at 7 times. Most of his personal best times were the second fastest times ever achieved behind the world record. His personal best time in a single Ironman is 9:50:30 h:min:s in 2013.

Apart from the ultra-triathlons, he competed in 13 ultra-marathons. He has completed 'TransGranCanaria' ultra-run three times in 2015, 2016 and 2018 (i.e., 125 km, 7500 m elevation gain, best time 16:53:55 h:min:sin 2018), 'Vaarojen Marathon' in 2011 (86 km, 2325 m elevation gain), 'Axtrail Ultra Trail Aldeias Do Xisto' in 2012—Utax (82 km, 5000 m elevation gain), 'The North Face Ultra-Trail Du Mont-Blanc®' in 2013 - Tds® (119 km, 7250 m elevation gain), 'Lanzarote Ultra-Trail' (84 km, 2674 m elevation gain), 'Haanja Ultra100' (100 km, 1125 m elevation gain) in 2014, 2015 and 2017—he won all those three races, with the best time 8:03:48 h:min:s.

In the last three years before the planned event of 40 Ironman-distance triathlons in 40 days, his training consisted primarily of swimming, cycling, running, and body strength workouts. On average, he invested annually ~1100 h of training with ~900 km of swimming, ~12,000 km of cycling and ~5000 km of running. He developed his own training principle where his training units have short transitions. With this principle, he saves time and trains the transition from one discipline to the other. As an example, he starts his training day with running 11 km, changes to swimming 4.4 km, changes then to running 13 km, followed by cycling for 1.5 h. He likes to split the distance. In order to run 84.4 km (two marathons) in a Double Iron ultra-triathlon in 6 h, he needs to run long distances, but by splitting. As an example, when running 64.1 km, he starts with running 16.4 km, after a break of 1 h, he runs 15.4 km, after a small break he runs 20.3 km, and after a further short break 12 km. With this principle, he can run the same distance a few days later in three (21.7 km, 25.9 km, and 16.3 km) compared to four runs.

2.3. Event, Equipment, Support, and Measurements

The concept of the event was a self-paced world record attempt to achieve as the first athlete in the world 40 Ironman-distance triathlons in 40 days. The event was held in Fuerteventura (Las Playitas, Canary Island). The challenge started 28th of September, 2019. The athlete followed no specific pacing strategy. Every day of the event, he tried to swim, cycle and run as fast as possible with the consideration that he must be able to compete the following day in the same manner. He always selected the specific speed in all three disciplines according to his subjective feelings without monitoring of heart rate or split times. He tried to get through the transition zones and food points as fast as possible. The athlete is a very experienced competitive athlete to listen to body signals and tries to prevent potential problems such as overheating, dehydration, lack of energy, muscle overload, muscle stiffness, general fatigue, or lack of sleep. Based upon his previous experience he knows himself very well, and most of the time he can notice and understand these signals. He monitored his body weight daily (every morning) using Garmin Index Smart Scale [16]. The energy expenditure was measured using the heart rate method with Garmin Forerunner 935 [16].

The swim took place in the 50-m outdoor pool at Playitas resort [17] where the water temperature was ~26–27 °C. He used every day a sleeveless wetsuit [18]. The bike course was one loop of 180 km with an elevation of 2079 m (measured by Google maps and Garmin Forerunner 935). The bike loop was open to car traffic. He used a Trek® Concept 7.5 triathlon bike with regular Bontrager® race wheels [19]. The run took place at Playitas resort with 12.5 laps of 3.375 km each (measured by Google maps and Garmin Forerunner 935). He started from the highest point of the lap and had to then complete 12 full laps. The last half lap consisted in the descent to sea level. He measured the elevation by running two full laps and divided the whole elevation (54 m) by two resulting in 27 m elevation difference per running lap. Running shoes were mostly different types of Saucony® shoes (i.e., Kinvara 9, Kinvara 10, Everrun, Type A8, Fastwitch 8 and Fastwitch 9) [20]. One day, he used also Hoka® One Tracer's shoes [21] but changed them during the run because he felt uncomfortable.

The athlete started the swim for Days 1–29 at 07:40 a.m. After turning the clock one hour back at the end of October, he started at 07:30 a.m. This procedure was chosen in order to be able to run as long as possible in daylight due to the change from summer to winter time. The weather was very windy, mostly sunny and basically without rain. Two times was some rain. The average high temperature in October in Fuerteventura is ~26 °C, and no more than two days with little rain can be expected [22]. Daily temperatures (i.e., the daily highest air temperatures) were obtained from a local weather report [23]. All times (i.e., swimming, cycling, running, transition, overall, and sleeping time) were measured using a Garmin® watch (Forerunner 935) and by the support crew using a stop watch CHRO301. Official times were those times measured and recorded by the support crew.

During the 40 days he had a team of 25 persons. They worked in shifts and were responsible for timing, nutrition, and material. In swimming, he had always two persons where one was counting the laps. During the swim, he did not eat anything, only had a bit of a sports drink. After the swimming, he ate one big bowl of cereal or rice porridge with cinnamon and cactus jam. Then, the crew changed where two new persons accompanied him by car on the bike. During the 180 km, he had six food stations where he made a short stop for food. In case of very hot weather, 3–4 additional aid stations were added as drink stations where he passed without stopping.

During cycling, he drank mostly water with minerals and vitamins [24], sports drinks, iced tea, sometimes one cup a coffee, pure 100% orange juice, non-alcoholic beer and lots of pure water. On the bike course, he always consumed salt tablets (www.enervit.com). Between the food stations, he consumed lots of gels. At the food stations, he consumed croissants with cheese, lots of muesli, fruits (i.e., watermelon, kiwi, melon, grapes), fried eggs, pancakes with Nutella®, very many different sweets (i.e., chocolate, biscuits, Enervit® protein bars). At the last food station and also right after finish of the cycling, he ate a bowl of Kellogg's® cereals with orange juice.

During the run, he had two persons as support crew where one was following him by bike and supported him with food and drinks and the other one was located at the food station to prepare nutrition. On the run course, he stopped only for comfort breaks and changes of shirts or running shoes. During the run, he consumed Coca Cola® (about 1,4 l/day), iced tea (about 0.7–0.8 l/day), lots of watermelon and some biscuits. During the run, he always consumed salt tablets [25]. Overall, during the 40 days, he consumed about 700 salt tablets.

After the run, he always had a bowl of Kellogg's® and often ate lots of sweets. Then 30 min later, he had dinner at Playitas restaurant (Swedish table). He ate pasta, rice, fries with different sauces, bread with cheese and jam, and lots of different homemade cakes. An hour later, he had a massage of about one hour. Before and after the massage, he ate bread with egg butter, omelette with cheese, sweets, nuts and raisins and/or crispbread with peanut butter and he drank lots of iced tea and mineral water. During the night, he ate food he brought with him from the restaurant (e.g., pasta, cakes etc.). In the morning, he drank coffee, ate sweets and Kellogg's®, and sometimes he took one RedBull® before swimming. He was sleeping in an apartment of Playitas resort. The sleeping time was measured using Garmin® Forerunner 935.

During the challenge, he faced many different problems. At the beginning, he had problems with weight loss because he was not used to expending so much energy per day. Body mass loss was too fast in the first days and the loss continued without stopping toward the half of the event. In the first days, he had also problems with the hot weather (temperature was over 30 °C) and on the 6th day he felt overheated at the end of the bike. He took 25 min in T2 zone to recover using ice and consuming cold drinks. At the beginning of the challenge, he had problems with the bike position since he was not used to sit on the saddle for such a long time. After six days he got used to it. During the challenge, he had only three blisters. To avoid blisters, he used finger socks and Vaseline. The mentally hardest days (self-reported by the athlete and not utilizing a standard measurement) were days 6, 12 and 18 and were mainly caused by very difficult wind conditions (i.e., strength or direction). He was physically in good conditions and had no muscular problems, However, these three days, it took him a lot of willpower to cope with the strong winds while cycling. By the end of these days, he felt mentally tired (self-reported by the athlete and not utilizing a standard measurement).

The second half of the challenge, he had problems with muscles that caused some discomfort during the run. During the 40 days, he suffered from different muscular problems, but no overuse injuries. The athlete considered the second half of the challenge mentally and physically easier than the first half. Mentally because after passing day 20, he was doing something new since his previous challenge was 20 Ironman-distance triathlons in 20 days. So, every step forward was pushing his limits farther. The first part was harder because at the beginning he was not used to it and he got under great stress. Also, he needed some time to get used to the hot weather. When some days the temperature dropped a bit, he started to feel a bit chilly. The harshness of the whole challenge was hidden in the weather conditions (i.e., wind) with the direction and/or the strength of the wind. At first, both body and mind needed time to adjust to the wind. As the days went on, he got mentally stronger. He believed that when he had passed the previous point (20 days), it would become mentally easier and he could compete faster.

2.4. Statistical Analysis

Exploratory analysis of the data was performed, reporting mean, standard deviation, minimum, maximum, and coefficient of variance (CV) of the variables. A paired t-test was applied to compare average heart rate of cycling and running. Repeated measures ANOVA was applied to test the time effect over performance with the average of every 10 race days. Non-linear regressions (2nd and 3rd order polynomial) were applied to investigate changes in split (swimming, cycling, and running) and overall times. Multivariate regression analyses were performed to test which variables had the greatest influence on the dependent variables. We ran three models: model 1 = cycling performance as the dependent variable, model 2 = running performance as the dependent variable, and model 3 = overall performance as the dependent variable. All procedures were conducted using Statistical Software for the Social Sciences (IMB® SPSS v25. Chicago, Il, USA) and GraphPad Prism (GraphPad Prism v8. San Francisco, CA, USA). The statistical significance was set at $p \leq 0.05$.

3. Results

The athlete completed the 40x Ironman distances in a total time of 444:22 h:min. He invested 50:26 h:min in swimming, 245:37 h:min in cycling, 137:17 h:min in running and 11:02 h:min in transition times. Performance in swimming and cycling became reduced over the days, whereas running performance improved until the 20th day and became reduced to the 40th day (Figure 1). Second-order non-linear regressions showed the best fit for swimming, but low or non-fitting models for cycling, running and overall race time. The coefficients of variance (CV) were higher in running, followed by cycling and swimming (Table 1). Regarding overall times, the last two days were the slowest ones, which were also the slowest regarding running and cycling performance. A third-order polynomial non-linear regression was the best fit for overall performance ($R^2 = 0.732$). Ten-day race time average showed a significant time effect for swimming ($F = 17.6$; $p < 0.001$), running ($F = 7.5$; $p = 0.011$) and overall performance ($F = 4.2$; $p = 0.034$), but no for cycling (Figure 2).

Table 1. Split and overall times (minutes), coefficient of variance (CV) and goodness of fit in 2nd order polynomial regression (R^2) for 40 Ironman-distance triathlons for 40 consecutive days.

Times	Mean (±SD)	Min–Max	CV	R^2
Swimming split time	75.6 (1.4)	71.8–79.9	1.9	0.63
Cycling split time	368.4 (11.3)	347.0–399.8	3.1	0.00
Running split time	205.9 (9.5)	192.6–230.0	4.6	0.41
Overall time	666.5 (21.0)	628.5–720.0	3.1	0.13

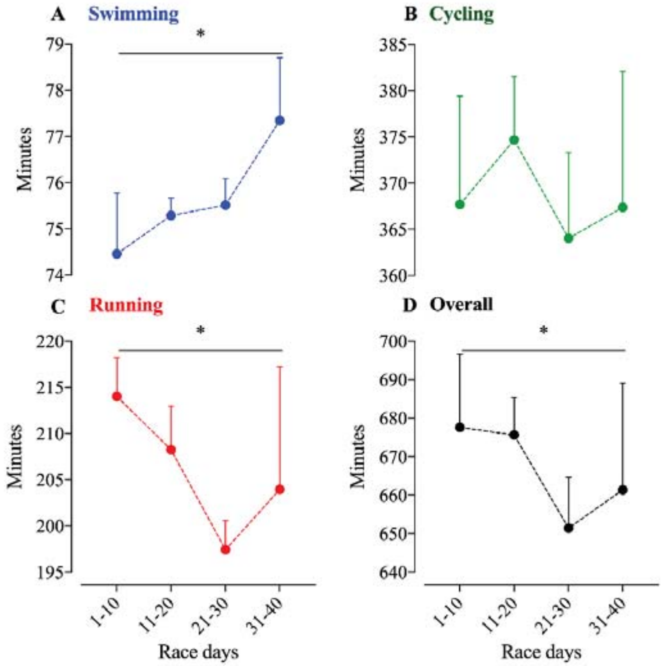

Figure 1. Ten-day average times of swimming, cycling, running, and overall time in 40 Ironman-distance triathlons. *: statistical significance ($p < 0.05$) for time-effect. (**A**): Swimming time; (**B**): Cycling time; (**C**): Running time; (**D**): Overall time.

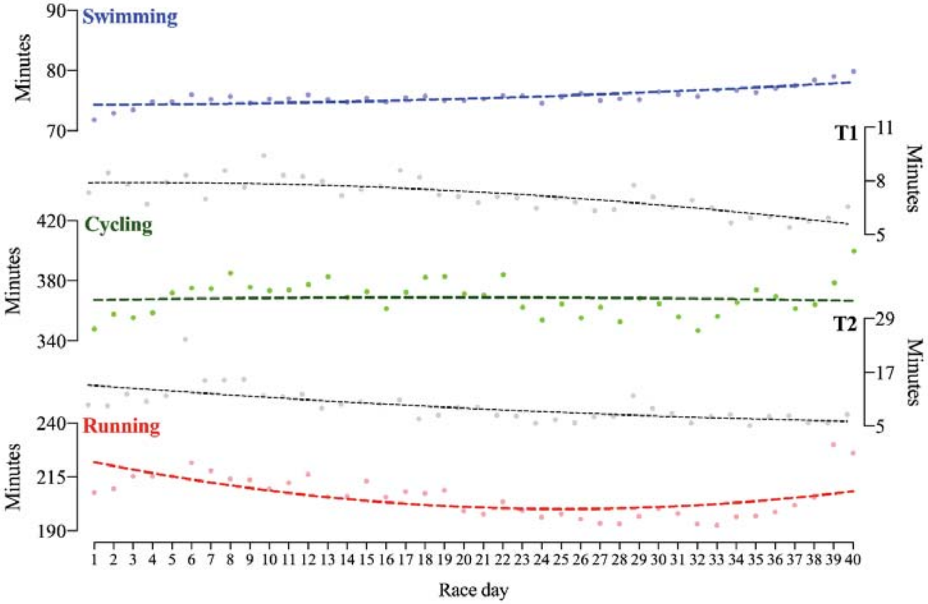

Figure 2. Times of swimming, cycling, running, and transition in 40 Ironman-distance triathlons in 40 days.

Physiological variables are shown in Table 2. Times of the event increased (got slower) until day 15, improved to day 31 and then again increased until the end indicating a sinusoidal pattern (Figure 3A). Average heart rate was significantly higher in running in comparison to cycling ($t_{39} = 21.7$; $p < 0.001$) (Figure 3B). Morning body weight reached the highest peak on the 6th day with 77.8 kg and the lowest peak on the 23rd and 36th day with 73.6 kg (Figure 2C). Wind speed and temperature showed erratic behaviors throughout the days (Figure 3D), with wind speed going up to 52.5 km/hon the 35th day, and the temperature reaching 35 °C on the 3rd day. Energy expenditure was higher in the first 10 days, and then varying from ~7500 to 8000 kcal/day in the following days (Figure 3E). Finally, with the exception of the first night with only 232 min of sleep, sleep duration varied between ~370 and ~480 min/night throughout the race days (Figure 3F).

Table 2. Total energy expenditure, average heart rate, sleep time and body weight during 40 Ironman-distance triathlons for 40 consecutive days.

Measured Variables	Mean (±SD)	Min–Max
Energy expenditure (total kcal)	7780.2 (319.5)	7159.0–8815.0
Average heart rate in cycling (bpm)	106.9 (6.3)	99.0–131.0
Average heart rate in running (bpm)	116.5 (5.6)	108.0–136.0
Sleep duration (min)	405.5 (37.6)	232.0–478.0
Morning body weight (kg)	74.8 (1.3)	73.6–77.8

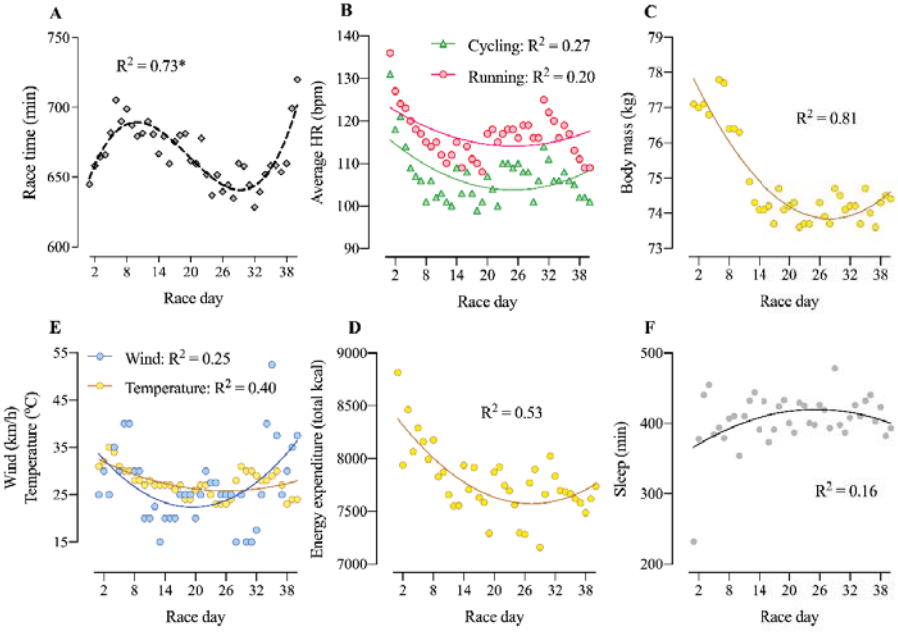

Figure 3. Non-linear regression for overall performance, weather and physiological variables in 40 Ironman-distance triathlons in 40 days. *: third-order polynomial fit; all other regressions were second-order polynomial fits. (**A**): overall performance; (**B**): average heart rate (HR) during cycling and running; (**C**): body mass; (**D**): wind speed and average temperature; (**E**): energy expenditure; (**F**): sleep minutes between race days.

Table 3 presents the results of the four multivariate regression models. The first multivariate regression model (swimming model) used swimming performance as the dependent variable and showed statistical significance ($p = 0.010$) with an adjusted R^2 of 0.28. The only significant independent variables to influence swimming performance was wind speed ($p = 0.005$), and a trend ($p = 0.062$) for body weight. The second multivariate regression model (cycling model) used cycling performance as the dependent variable and showed statistical significance ($p < 0.001$) with an adjusted R^2 of 0.78. Wind speed, overall time in the previous day and average heart race during cycling were the significant independent variables ($p < 0.05$) to influence cycling performance. The third multivariate regression model (running model) used running time as the dependent variable and showed statistical significance ($p < 0.001$) with an adjusted R^2 of 0.69. Body weight and average heart rate during running were the significant independent variables ($p < 0.05$) to influence running performance. The fourth multivariate regression model (overall model) used overall time as the dependent variable and showed statistical significance ($p < 0.001$) with an adjusted R^2 of 0.98. Cycling performance, running performance and body weight were the significant independent variables ($p < 0.05$) to influence overall time.

Table 3. Results of the multivariate regression models to test which variable has the highest influence in swimming, cycling, running and overall time in 40 Ironman-distance triathlons in 40 days.

	Independent Variables	Standardized β	*p*-Value
Model 1 Swimming	Sleep time	−0.09	0.59
	Body weight	−0.46	0.06
	Wind speed	0.47	0.01
	Overall time previous day	0.10	0.57
	Temperature	−0.18	0.40
Model 2 Cycling	Swimming time	−0.01	0.96
	Sleep time	0.04	0.68
	Body weight	−0.08	0.57
	Wind speed	0.29	0.01
	Overall time previous day	0.51	<0.01
	Temperature	0.12	0.33
	Average heart rate	−0.61	<0.01
Model 3 Running	Swimming time	0.221	0.12
	Cycling time	0.11	0.53
	Sleep time	−0.13	0.25
	Body weight	0.64	<0.01
	Wind speed	−0.01	0.97
	Overall time previous day	0.08	0.61
	Temperature	0.19	0.25
	Average heart rate	−0.50	0.02
Model 4 Overall	Swimming time	0.04	0.29
	Cycling time	0.60	<0.01
	Running time	0.45	<0.01
	Sleep time	−0.01	0.71
	Body weight	0.14	0.01
	Wind speed	−0.05	0.14
	Overall time previous day	0.02	0.69
	Energy expenditure	0.00	0.99
	Temperature	0.02	0.59

4. Discussion

In this self-paced world record attempt, we found that (i) performances in cycling and running were predictive for overall time, (ii) overall time was predictive for cycling performance of the following day, (iii) wind speed was predictive for swimming and cycling time, (iv) body mass was predictive for running and overall time, (v) heart rate was predictive for cycling and running time, (vi) temperature and sleep time were not predictive to performance, and (vii) the highest variation occurred in the marathon split times.

4.1. Cycling and Running were Predictive for Overall Race Time, But not Swimming

An important finding was that both cycling and running times were predictive for overall race time. While the cycling split is highly predictive in a single Ironman-distance race [26,27], both cycling and running are predictive in longer triathlon distances [28,29]. The contribution of swimming to overall performance in an ultra-triathlon is lower than for cycling and running [30]. A study investigating the performance level and race distance on pacing in ultra-triathlons (i.e., Double, Triple, Quintuple and Deca Iron ultra-triathlon) showed that the fastest ultra-triathletes spent relatively more time in swimming and cycling and less time in running, highlighting the importance of the role of the latter discipline for the overall ultra-triathlon performance [28]. Running seems, however, also important in a single Ironman-distance race. When split times (i.e., swimming, cycling, and running) and overall race times of 343,345 athletes competing between 2002 and 2015 in 253 different Ironman-distance triathlon races were analyzed, it was shown that the fastest Ironman-distance triathletes were the relatively fastest in running and transition times [31]. Similarly, a study investigating Triple Iron ultra-triathletes showed that running rather than cycling performance seemed to be the most important predictor for overall race time [32]. Overall, both cycling and running seemed of importance in ultra-endurance multi-stage triathlon races, but not swimming.

4.2. The Aspect of Experience and Age

The present athlete was able to perform during 40 days an Ironman-distance triathlon with an average race time of ~11 h. per day with 10:28 h:min for the fastest and 12:00 h:min for the slowest day. His personal best time in Double Iron ultra-triathlon was in June 2019 an official world record and his personal best time in 10× Ironman in 10 days was the second fastest time in history. His personal best time in a single Ironman is 9:50:30 h:min:s in 2013. Since then, he never competed again in a single Ironman triathlon.

It is well known that previous experience is important for an ultra-endurance performance such as an Ironman [33] or an ultra-triathlon [34–37]. When all female and male ultra-triathletes who had finished at least one Double Iron ultra-triathlon, one Triple Iron ultra-triathlon, one Quintuple Iron ultra-triathlon, and one Deca Iron ultra-triathlon, it was shown that fast race times in shorter ultra-triathlon races (i.e., Double and Triple Iron ultra-triathlon) were more important than a large of number finished races in order to achieve a fast race time in a longer ultra-triathlon (i.e., Quintuple and Deca Iron ultra-triathlon) [34]. In successful finishers in a Deca Iron ultra-triathlon, both the number of finished Triple Iron ultra-triathlons and the personal best time in a Triple Iron ultra-triathlon were related to overall race time [36]. Furthermore, the athlete is at the best age (36 years) for a fast Ironman triathlon performance since the most competitive age for male athletes is ~35 years for a top performance in an Ironman triathlon [38,39].

4.3. Sinusoidal Change in Overall Race Time

A further finding was the sinusoidal change in overall time with a decrease in performance until the 15th day, an improvement in performance up to the 31st day, and a decrease until the end of the event. To date, only one study investigated the pacing in a multi-stage triathlon longer than a Deca Iron ultra-triathlon. In the first and only Triple Deca Iron ultra-triathlon held until now, the daily performance remained unchanged across the 30 days (i.e., even pacing) in the eight male finishers [6]. Different intrinsic and extrinsic factors such as motivation, weather, course, nutrition etc. [40–42] might explain the difference between the finishers in a Triple Deca Iron ultra-triathlon and the athlete in the present case study.

4.4. The Aspect of Heart Rate and Energy Expenditure

Average heart rate was significantly higher in running in comparison to cycling splits, and heart rate during the split performances was predictive for the performance in the corresponding split.

It is well known from laboratory studies with triathletes that heart rate and energy expenditure is higher in running compared to cycling [43]. While the measurement of heart rate with a wrist-worn device might be reliable [44], the reported energy expenditure from these devices should be interpreted with caution [45], given their potential bias and error [46], the current wrist-worn activity trackers are most likely not accurate enough [26]. However, these devices might be suitable for the use in interventions of behavior change as they provide feedback to user on trends in energy expenditure [46]. Energy expenditure reported by wrist-worn devices differs between different sports disciplines (e.g., walking and running) with a general increase as exercise intensity increased [46]. Interestingly, heart race correlated with cycling and running performance, but not the measured energy expenditure with the wrist-worn device. However, this is the only way to get a note about the energy expenditure under such conditions.

4.5. Energy Expenditure, Loss in Body Mass and Body Weight as Predictor in Performance

Mean energy expenditure per Ironman-distance was ~7780 kcal and the athlete lost a total of ~4 kg of body mass. Although the determination of energy expenditure using a wrist-worn device might be questionable, the mean energy expenditure per Ironman was practically the same as has been reported for an athlete in a 10× Ironman with 7544 ± 913 kcal per day when using a portable heart rate monitor [47]. For a single Ironman triathlon, energy expenditure is, however, higher at ~10,036 ± 931 kcal [48] to ~11,009 ± 664 kcal [49]. The difference might be explained that intensity during a single Ironman triathlon is considerably higher than when finishing every day an Ironman-distance triathlon during multiple days. This is evidenced due to this specific athlete's daily performance as compared to their personal best in a single Ironman-distance event.

A loss in body mass in a single Ironman triathlon [50,51] and an ultra-triathlon [52–55] is a common finding where the loss in body mass in an Ironman triathlon is related to a loss in skeletal muscle mass [50]. This loss in muscle mass is due to depletion in muscle glycogen stores [50]. In ultra-endurance triathletes competing in longer races than an Ironman triathlon, mainly body fat is lost. The loss in body mass in a Deca Iron ultra-triathlon was due to a loss in body fat [54] and in Triple Iron ultra-triathletes the loss in body mass was related to the loss in body fat [55]. While the loss in body mass in a single Ironman triathlon is ~2 kg for male athletes [50,51], the present athlete lost only ~4 kg during 40 days. Obviously, he was well experienced to preserve his body mass during these 40 days. However, the absolute loss in solid body mass might have been higher since total body water increases in a multi-stage triathlon such as a Deca Iron ultra-triathlon [47,56]. In a Deca Iron ultra-triathlon, both body mass and fat mass decrease [36,47] whereas lean body mass [36] and total body water increase [47]. The increase in lean body mass is due to the increase in total body water [57]. While multi-stage ultra-triathletes seem to become overhydrated, finishers in a single Ironman are hypo- to dehydrated [58]. Overall, multi-stage ultra-triathletes seem able to preserve their body mass. In a Deca Iron ultra-triathlete, the overall loss in body mass was ~1 kg although the energy deficit was ~11,480 kcal during the 10 days [47].

Body mass was predictive for running and overall time. For a single Ironman triathlon, body mass as an anthropometric variable is not predictive for race time, but rather previous experience such as the personal best time in an Olympic distance triathlon and in a marathon [59]. In the present athlete, body mass decreased through the event and we might assume that the loss in body mass was 'ergogenic'. Studies investigating marathoners [42] and 100 km ultra-marathoners [60] showed that a loss in body mass was related to faster race times. A study investigating marathon runners showed a relationship between running speed during training and percent body fat [61].

4.6. The Influence of Environmental Factors (e.g., Wind Speed and Temperature) and Recovery

It was shown that wind speed was predictive for swimming and cycling, but the daily highest temperature was not related to performance. It is well known that environmental conditions such a wind and heat have an influence on ultra-endurance performance [62]. While it is known that wind has an influence on ultra-cycling performance [63], one might assume that also heat might impair ultra-endurance performance [64]. Triathletes competing under thermal stress conditions in the 'Ironman Hawaii' reached a state of hyperthermia during the marathon [65].It was observed that overall time was predictive for cycling performance of the following day and sleep time was not related to performance. It seems that the athlete was able to find his best way of intensity and recovery since it is well known that sleep deprivation would lead to an impaired athletic performance [66].

4.7. Limitations

Although this case study provides a lot of physiological and technical details, some limitations must be acknowledged. The daily measurement of body composition using a bioelectrical impedance analysis or radiological procedures like BIA [67], DEXA [68], or MRI [69] would help monitor changes in body composition. The inclusion of the last two days with very slow overall times (outlier) might have had an influence on the analysis of the data. Missing data for humidity [70], wind direction [71] and nutrition [72] also might have had an influence on the outcome of the analysis. We only used the daily maximum of air temperature and wind speed which might have had an influence on our analysis.

4.8. Practical Applications

This case study shows that it is possible to perform an Ironman-distance triathlon daily for 40 days. The findings in this study may help any athlete intending to outperform the performance with a faster time per Ironman or to finish more consecutive Ironman-distance triathlons.

5. Conclusions

In summary, a fast running performance is significantly influenced by a low body weight, a fast cycling performance is significantly influenced by both a low wind speed and by the previous day's effort, and a fast overall performance is significantly influenced by both fast running and fast cycling performances. This study could be of assistance for coaches and athletes preparing for similar challenges. More in-depth physiological responses (i.e., body composition measures, sleep quality, inflammation, oxidative stress) to such a challenge with or without a follow-up could be the next step. A highly trained professional triathlete with extensive previous experience is able to finish daily an Ironman-distance triathlon on consecutive 40 days where cycling and running performance are highly predictive of overall time.

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