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# Training for Optimal Sports Performance and Health

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Edited by

Hadi Nobari and Juan Pedro Fuentes García

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*International Journal of Environmental Research and Public Health*

# **Training for Optimal Sports Performance and Health**



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Editors

**Hadi Nobari**

**Juan Pedro Fuentes García**

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*Editors*

Hadi Nobari  
University of Extremadura  
Spain

Juan Pedro Fuentes García  
University of Extremadura  
Spain

*Editorial Office*

MDPI  
St. Alban-Anlage 66  
4052 Basel, Switzerland

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# Preface to “Training for Optimal Sports Performance and Health”

A complete and regular set of different areas of sports science is required to achieve optimal sports performance. Exercise physiology is one of the most important and practical fields in sports science, as it provides information for strength and conditioning coaches in sports teams. For this reason, identifying the physical demands of sports must be a priority for professional coaches. Each sport’s physical demands differ, with a wide range of skills at varying intensity levels. Athletes are primarily involved in running, kicking, jumping, sprinting, and changing direction; these physical abilities require maximum strength, maximal oxygen uptake, anaerobic power, and the neuromuscular system. In order for athletes and sports teams to achieve optimal performance in these physical indicators, the coach must be able to provide for the athletes and their teams in the following ways: (i) fitness assessments—step-by-step evaluations of athletes in different training phases in the competition season and pre-season; (ii) sport-specific conditioning—a variety of exercises for running at different speeds, acceleration, deceleration, and exercises in simulated field conditions such as small-sided games (SSG); (iii) strength training—including a set of resistance exercises, including weight training, dumbbells, barbells, and functional training; (iv) explosive power, speed, and change of direction development—involving the correct techniques of jumping with one and a pair of legs, jumping on different boxes, sled run, band-resisted running, and agility ladders drills; (v) finally, and most importantly, managing the training load, recovery between the training sessions and match, injury prevention, and the rehabilitation of players through monitoring, which can be of external load to all types of running, metabolic power, and body load, internal load to the rating of perceived exertion or heart rate and, finally, the well-being status of players using various tools such as the Hooper index questionnaire.

Therefore, this Special Issue aims to identify the training strategies that can achieve the desired performance in competitions for athletes, whether individually or as a team, and considering different ages and sports. These strategies can (1) be considered and studied as periodization and programming for training in different periods of the season; (2) maintain the athlete’s performance or lead them towards the desired performance by managing their training load; (3) monitor the athlete’s wellness status to prevent the development of over-training syndrome and non-functional overreaching statuses, as well as to achieve the desired performance and prevent performance distortion; (4) quantify the relationship between fitness assessments with training loads and well-being status; (5) examine the role of different types of training such as SSG, agility, speed, and resistance training in different age groups in different teams.

**Hadi Nobari and Juan Pedro Fuentes García**

*Editors*





Article

# The Effects of Eccentric Contraction Execution Time on the Properties of the Patellar Tendon

Fernando Martínez <sup>1</sup>, Pablo Abián <sup>2</sup>, Fernando Jiménez <sup>1</sup> and Javier Abián-Vicén <sup>1,\*</sup>

<sup>1</sup> Performance and Sport Rehabilitation Laboratory, Faculty of Sport Sciences, University of Castilla-La Mancha, 45071 Toledo, Spain; fermasa83@gmail.com (F.M.); josefernando.jimenez@uclm.es (F.J.)

<sup>2</sup> Faculty of Humanities and Social Sciences, Comillas Pontifical University, 28049 Madrid, Spain; pabian@comillas.edu

\* Correspondence: javier.abian@uclm.es; Tel.: +34-925268800 (ext. 5522)

**Abstract:** The purpose of this study was to assess the effects of eccentric contraction execution time on the morphological and elastic properties of the patellar tendon (PT) in a six-week, single-leg decline squat (SLDS) exercise training program. In addition, the effects of a six-week detraining period on the same variables were evaluated. Fifty participants were randomized into the control group (CG;  $n = 15$ ), experimental group 1 (EG6s;  $n = 17$ ; eccentric contraction execution time = 6 s) and experimental group 2 (EG3s;  $n = 18$ ; eccentric contraction execution time = 3 s). The thickness and elastographic index (EI) in different regions of interest (ROIs) in the PT were measured after 6 weeks of eccentric training using the single-leg decline squat exercise (three sessions per week, 80% of the eccentric one-repetition maximum) and after 6 weeks of detraining. There was an increase in the thickness of the PT in the different ROIs analyzed in both experimental groups at the end of the training period. Especially worth noting was the increase in the thickness of the PT at the proximal level in EG3s ( $p = 0.001$ ), and the increase at the distal level in EG6s ( $p = 0.001$ ). On the other hand, there was a reduction in EI in EG6S at the end of the intervention program ( $p = 0.021$ ), and both experimental groups increased EI in the three regions of interest analyzed after the detraining period ( $p < 0.01$ ). In conclusion, the execution time of the eccentric contraction in the SLDS exercise determines the anatomical level of the morphological adaptations in the PT. These morphological adaptations are lost after 6 weeks of detraining, producing an increase in tendon stiffness.

**Keywords:** eccentric training; decline squat; patellar tendon; sonoelastography; stiffness

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## 1. Introduction

The patellar tendon (PT) originates from the distal two-thirds of the patella and generally inserts at the most distal end of the anterior tuberosity of the tibia, where it joins the fibrillar expansions of the iliotibial tract [1]. Three vastii and part of the rectus femoris act on the knee joint by way of the vastus aponeurosis, which is likely to have a role in knee pathomechanics [2]. Functionally, the PT is an energy storing structure that transmits muscle-derived forces that produce joint motion and stores and releases energy, which has the capability to enhance power and efficiency in extension [3,4]. The PT stores and releases a great deal of energy during explosive movements and this possibly contributes to the high incidence of patellar tendinopathy in elite athletics [5,6]. PT fibers do not have the same characteristics which can generate different adaptations to mechanical stimulation in the different sections of the tendon; Basso et al. [7] reported that the posterior fibers of the PT are exposed to the greatest strains when the quadriceps tendon is loaded with the knee flexed.

Eccentric exercise is frequently used in physiotherapeutic interventions for the treatment of patellar tendinopathy [8]. Specifically, the eccentric single-leg decline squat (SLDS) exercise has been one of the most widely used because it offers superior results compared

with other exercises [9]. Furthermore, different investigations show the ability of the tendon to morphologically and mechanically adapt its structural components to external load stimuli [10–12]. In this respect, it seems that high loads and longer intervention durations (>12 weeks) may be the most influential variables to produce these modifications [11,13,14].

Movement speed during eccentric training alters important factors involved in muscular adaptations, such as time under tension and muscle activation [15,16]. Burd et al. [17] found that greater muscle time under tension increased mitochondrial and sarcoplasmic protein synthesis. Fast eccentric movement during training has been found most effective for muscle hypertrophy and strength gain [16,18] and slow eccentric movement during training has had larger effects on the elastic and structural properties of the PT [16]. However, there is a need to investigate the different components of the eccentric exercise load on the morphological and mechanical properties of the muscle and tendon structures. Therefore, the aims of this study were: (1) to compare the effects of the execution time of the eccentric contraction (3 s vs. 6 s) on the morphological and elastic properties of the PT after a 6 week SLDEe training program, and (2) to analyze the changes in these variables after a 6 week detraining process.

## 2. Materials and Methods

### 2.1. Participants and General Procedure

Fifty-four healthy physical education students voluntarily participated in this study. Eligible participants were physically active (3–6 h per week of moderate physical activity) and between 18 and 35 years old. All participants scored > 90 points on the Victorian Institute of Sport Assessment-PT questionnaire (VISA-P) in its version translated to Spanish (VISA-P-Sp) [19] to exclude symptoms of patellar tendinopathy, and they were required to keep their normal exercise practices throughout the study. Participants were excluded from the investigation if they: (1) had had an injury in the lower limbs during the last 2 years; (2) had performed strength training in the lower limbs during the last 8 weeks; and (3) practiced basketball or volleyball (sports where jumping is an important action) more than 2 h per week or competitively. The sample size was previously calculated based on existing research [10] which measured the influence of resistance training on PT stiffness. The minimal number of participants required to attain a power of 0.9 and a bilateral alpha level of 0.05 was calculated to be 10 participants per group.

Participants were randomly divided into three groups: (1) the control group (CG;  $n = 15$ ;  $21.3 \pm 2.8$  years;  $67.22 \pm 12.25$  kg;  $1.72 \pm 0.09$  m and  $95.8 \pm 1.9$  score in the VISA-P-Sp; 3 participants were lost during the follow-up of the study) with no intervention program; (2) experimental group 1 (EG6s;  $n = 17$ ;  $21.2 \pm 2.2$  years;  $67.14 \pm 12.37$  kg,  $1.72 \pm 0.09$  m and  $95.4 \pm 2.1$  score in the VISA-P-Sp; 1 participant was lost during the follow-up of the study) which followed the intervention program performing the SLDEe in 6 s; and (3) experimental group 2 (EG3s;  $n = 18$ ;  $21.3 \pm 2.5$  years;  $67.68.27 \pm 11.85$  kg,  $1.70 \pm 0.10$  m and  $96.1 \pm 2.4$  score in the VISA-P-Sp) which followed the intervention program performing the SLDEe in 3 s (Figure 1).

All participants were informed about the objective and methods of the investigation and signed the informed consent forms before the start of the study. Ethical approval was obtained from the Ethics Committee of Clinical Research at the Hospital Complex in Toledo (Spain) (62-100615) according to the principles of the latest version of the Declaration of Helsinki.

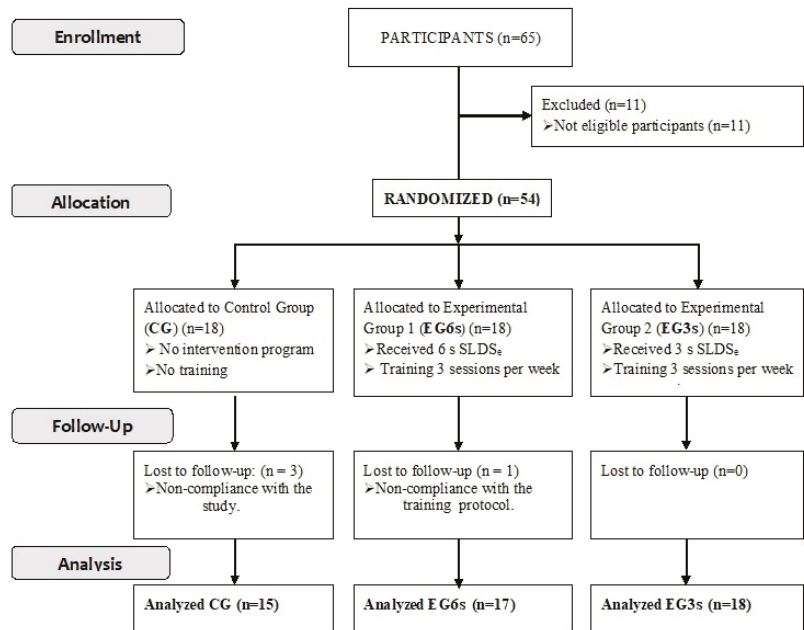


Figure 1. Participants and inclusion procedure.

## 2.2. Experimental Design

A three-group parallel repeated measures design was carried out to examine the thickness and elastography index adaptations of the PT in response to two SLDS<sub>e</sub> training programs performed with two execution times (6 s vs. 3 s). In week −1, the participants were selected and the VISA-P-Sp questionnaire was filled out. In week 0, two sessions were carried out. In session 1, the first assessment (PRE) was made before the intervention program (these data were used as baseline measures) and the anthropomorphic measures were recorded. Participants were then notified of their allocated training group and familiarized with the eccentric SLDS<sub>e</sub> exercise and with the 5 repetition maximum (RM) test of their corresponding exercise-specific training program (no training, 3 s of SLDS<sub>e</sub> or 6 s of SLDS<sub>e</sub>). In session 2 (72 h after session 1), the 5 RM test of the SLDS<sub>e</sub> exercise for EG6s and EG3s in the dominant limb was performed in order to determine the training load.

From weeks 1 to 6 the participants completed a 6 week eccentric program. Each session was carried out with the principal investigator and two collaborators to ensure adherence and the correct technique. In week 7, the second measurement was performed at the end of the intervention program (POST-1) and in week 14, the third measurement was performed, 6 weeks after the intervention finished (POST-2) to evaluate the consequences after a 6 week detraining period (Figure 2).

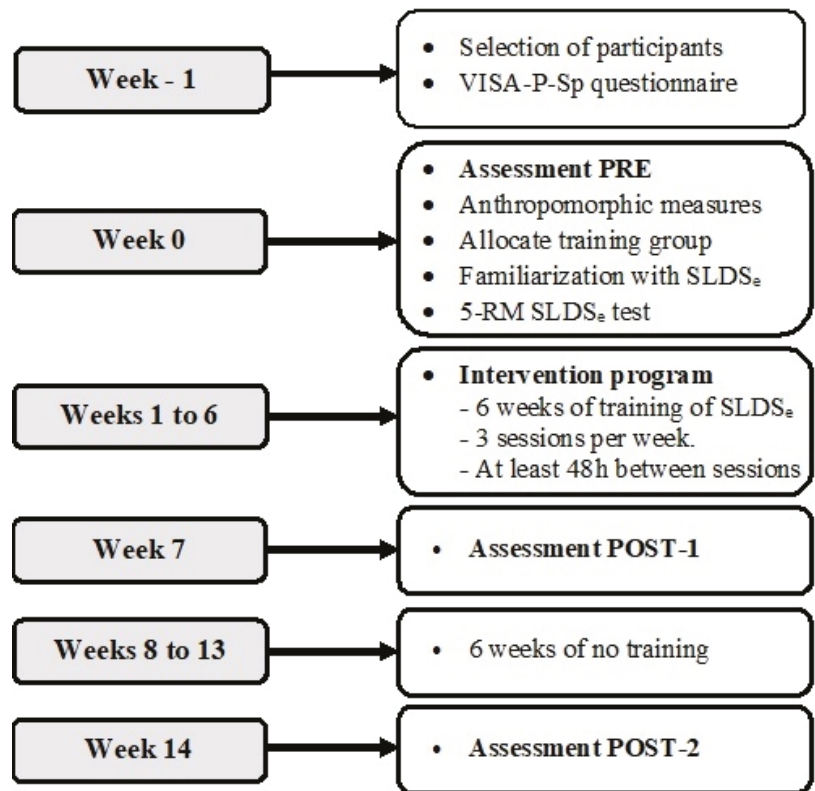


Figure 2. Design of the investigation.

### 2.3. Intervention Program

The intervention program of the SLDS<sub>e</sub> training for both groups (EG6s and EG3s) lasted 6 weeks. Three training sessions were carried out every week at least 48 h apart. Three sets of 8 repetitions were performed in each training session (the rest duration between repetitions was 6 s and between sets was 2 min) of the SLDS<sub>e</sub> exercise previously described [20] with the dominant lower limb at 80% of the 1 RM calculated previously for each group in their specific working execution time. A 25° decline board was used to perform the SLDS<sub>e</sub> in both experimental groups [20].

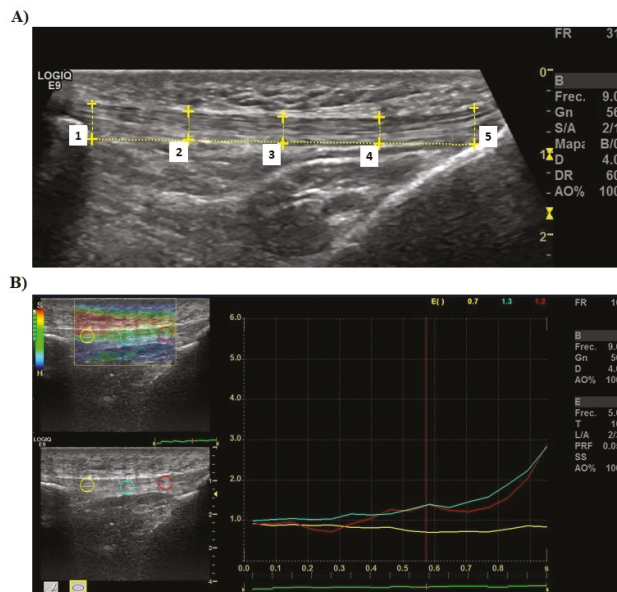
The eccentric phase of movement of each repetition lasted 6 s in EG6s and 3 s in EG3s. The 1 RM of the SLDS<sub>e</sub> for each experimental group was calculated before the start of the training program (in week 0) and just after the training in weeks two and four to recalculate the training load. Participants were trained to complete the back-squat exercises with the trunk in a vertical position in a multipower machine (Technogym, Gambettola, Italy). Both experimental groups were trained to perform the exercise by slowly bending the knee to 90° of flexion and, in order to focus only the eccentric loading of the quadriceps and to return to the initial position, the participants were trained to use the non-dominant lower limb in addition to the dominant one and two assistants lifted the weights to prepare them for the next repetition. Thereby, we decreased the influence of the concentric phase of the anterior thigh muscles. A metronome ([www.webmetronome.com](http://www.webmetronome.com); accessed on 5 February 2019) was employed to establish the start and the end of the eccentric phase in each group.

#### 2.4. 5-RM SLDSe Test

The 5 RM SLDSe test was used to calculate 1 RM. This test was performed with the dominant lower limb in a multipower system (Technogym, Gambettola, Italy), with two parallel straight bars that only allowed vertical movements. The participants were trained to position themselves under the shoulder bar of the multipower rack machine. A specific warm-up routine was carried out using the weight of the bar (~18 kg), the participants performed two sets of twelve eccentric repetitions, and one minute rest. Each of the eccentric repetitions was carried out by the two experimental groups in a time of 6 s (EG6s) or 3 s (EG3s), reaching up to 90° of knee flexion, which was assessed with a goniometer placed on the knee joint, and resting for 6 s between each repetition. After 2 min of rest, the test was carried out with the participants performing a series of 5 eccentric repetitions with increasing intensity, starting with the weight registered in the pre-testing. The load was increased until the 5 RM was determined with a 2 min rest between sets. If the subject did not keep the execution speed of the movement or did not reach 90° of knee flexion, the repetition was considered invalid. In the event that the participants could not perform the repetition due to fatigue or inability to endure the weight, the weight lifted in the last series of 5 RM was recorded. Afterwards, the 5 RM was converted to 1 RM [11,21].

#### 2.5. Thickness of PT Assessment

The thickness of the PT was measured by B-mode ultrasonography (LOGIQ E9, GE Healthcare, Milwaukee, WI, USA) with a 5.5 cm, high frequency 6–15 MHz linear probe. The participants lay supine on a stretcher, with the knee flexed to 15° [16,22]. The transducer was placed parallel to the direction of the PT fibers. The length of the PT was measured from the lower pole of the patella to the anterior tuberosity of the tibia (deep insertions of the PT). The tendon thickness was measured in five regions of interest (ROIs) (lower pole of the patella, at 25%, 50% and 75% of the total PT length, and anterior tuberosity of the tibia) with the specific software for the US machine (Figure 3). These measurements have been used in previous research and have shown acceptable reliability [23].



**Figure 3.** (A) B-mode ultrasound image of the Achilles tendon in a longitudinal plane. (B) Example of elastography measurement of the Achilles tendon.



### 2.6. Elastic Properties of PT Assessment

The elastographic index (EI) of the PT was evaluated in three ROIs (at 25%, at 50% and at 75% of the total length PT; Figure 3) and in the same position as in the previous measurement by real-time elastography (LOGIQ E9, GE Healthcare, Milwaukee, WI, USA). This measurement was made following the methodology of previous research [24–28]. The average of the three measurements was stated. The B-mode screen was set as translucent and color-coded, real-time images were used to record the sonograms. The color code showed the strain of the tissues within the ROI, in which blue indicated hard elasticity, green and yellow corresponded to medium elasticity, and red indicated soft elasticity. A lower value of EI is related to a lower stiffness level. The ultrasound evaluation was carried out by one of the authors, F.J., who has proven experience with this type of measurement.

### 2.7. Statistical Analysis

The data were analyzed with the statistical package IBM SPSS Statistics 23.0 (SPSS, Chicago, IL, USA). The normality of each variable was initially tested with the Shapiro-Wilk test. All the variables presented normal distributions ( $p > 0.05$ ). Then, the main effects of the two training interventions were determined by a two-factor ( $3 \times 3$ ) mixed-model ANOVA. The first factor was the group (CG, EG6s and EG3s) and the second factor was the timeline (PRE-, POST-1 and POST-2). Effect size (ES) statistics were calculated according to the formula proposed by Cohen [29] to quantify the magnitude of the difference in pairwise comparisons. The magnitude of Cohen's effect size was interpreted using the following scale: an ES lower than 0.2 was considered as small, an ES around 0.5 was considered as medium and an ES over 0.8 was considered as large. All data were presented as mean  $\pm$  standard deviation. The significance level was set at  $p < 0.05$  for all statistical analyses.

## 3. Results

In the PRE-tests, no outcome measures differed statistically among groups (CG, EG6s and EG3s) except between CG and EG3s in the thickness of PT at 50%. Additionally, in the CG, no significant differences were found between the measurements obtained at PRE, POST-1 and POST-2 tests in any of the dependent variables. No significant differences were found in the length of the PT between PRE, POST-1 and POST-2 evaluations in any of the groups.

A significant increase was found after 6 weeks of eccentric training in the 1 RM in both experimental groups (EG6s: difference =  $89.7 \pm 29.9$  kg; confidence interval (CI) 95%: from 73.2 to 106.2 kg,  $p < 0.001$ , ES = 3.8 and EG3s: difference =  $100.5 \pm 33.7$  kg; CI 95%: from 84.5 to 116.5 kg,  $p < 0.001$ , ES = 3.9) and a decrease after 6 weeks of detraining was found in EG6s (difference =  $-30.8 \pm 24.2$  kg; confidence interval (CI) 95%: from  $-43.0$  to  $-18.6$  kg,  $p < 0.001$ , ES = 0.8) and EG3s (difference =  $-41.9 \pm 21.0$  kg; CI 95%: from  $-53.8$  to  $-30.1$  kg,  $p < 0.001$ , ES = 1.0). No significant changes in the 1 RM were found in the control group.

The values obtained for the length and the thickness of the PT in the different zones analyzed are shown in Table 1. The thickness of the PT in the lower pole of the patella was  $0.03 \pm 0.03$  cm (CI 95%, from 0.01 to 0.06 cm,  $p = 0.015$ , ES = 0.4) greater in EG3s after 6 weeks of eccentric training and a decrease after a 6 week follow-up was found in EG3s ( $p = 0.005$ ). A significant increase was found after 6 weeks of eccentric training in the thickness of the PT at 25% in both experimental groups (EG6s: difference =  $0.05 \pm 0.02$  cm; CI 95%: from 0.03 to 0.06 cm,  $p < 0.001$ , ES = 0.6 and EG3s: difference =  $0.03 \pm 0.03$  cm; CI 95%: from 0.01 to 0.04 cm,  $p = 0.001$ , ES = 0.4) and a decrease after 6 weeks of detraining was found in EG6s ( $p < 0.001$ ) and EG3s ( $p < 0.001$ ). The values in the thickness of the PT at 50% and 75% were greater in EG6s ( $p < 0.001$ ) and EG3s ( $p = 0.013$  and  $p = 0.001$ , respectively) after 6 weeks of eccentric training. Significant decreases ( $p < 0.05$ ) were found after the six weeks of non-training compared to POST-1 in the thickness of the PT at 50% (EG6s:  $p < 0.001$ ) and 75% (EG6s:  $p < 0.001$ ; and EG3s:  $p = 0.024$ ).

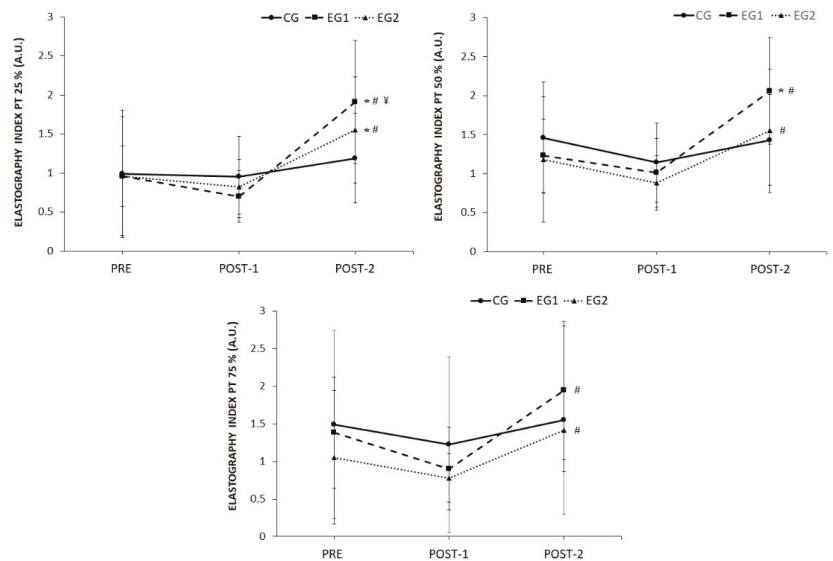
**Table 1.** Morphological properties of the PT in response to six weeks of eccentric training (POST-1) with two execution runtimes (EG6s = 6 s and EG3s = 3 s) and a 6 week follow up of detraining (POST-2).

	PRE	POST-1	POST-2	ANOVA Main Effects	F	p-Value
Length PT (cm)						
CG	4.53 ± 0.42	4.45 ± 0.47	4.44 ± 0.46	Time	0.37	0.694
EG6s	4.49 ± 0.64	4.45 ± 0.58	4.46 ± 0.55	Group	0.36	0.702
EG3s	4.47 ± 0.43	4.38 ± 0.39	4.47 ± 0.38	Time × group	0.60	0.667
Thickness PT in patella (cm)						
CG	0.48 ± 0.05	0.48 ± 0.06	0.47 ± 0.05	Time	12.35	<0.001
EG6s	0.45 ± 0.10	0.47 ± 0.09	0.43 ± 0.08 #	Group	0.48	0.621
EG3s	0.45 ± 0.08	0.48 ± 0.09 *	0.44 ± 0.07 #	Time × group	2.69	0.037
Thickness PT 25% (cm)						
CG	0.41 ± 0.06	0.40 ± 0.06	0.39 ± 0.06	Time	27.67	<0.001
EG6s	0.38 ± 0.07	0.42 ± 0.07 *	0.36 ± 0.07 #	Group	0.26	0.773
EG3s	0.39 ± 0.07	0.41 ± 0.08 *	0.35 ± 0.05 #	Time × group	8.10	<0.001
Thickness PT 50% (cm)						
CG	0.39 ± 0.06	0.37 ± 0.05	0.37 ± 0.04	Time	13.17	<0.001
EG6s	0.35 ± 0.05	0.42 ± 0.07 *	0.34 ± 0.06 #	Group	3.58	0.037
EG3s	0.31 ± 0.07 †	0.34 ± 0.07 *,†	0.33 ± 0.07	Time × group	7.80	<0.001
Thickness PT 75% (cm)						
CG	0.40 ± 0.05	0.39 ± 0.04	0.40 ± 0.04	Time	11.89	<0.001
EG6s	0.38 ± 0.06	0.44 ± 0.07 *	0.37 ± 0.07 #	Group	1.51	0.232
EG3s	0.36 ± 0.07	0.39 ± 0.07 *	0.35 ± 0.07 #	Time × group	3.91	0.006
Thickness PT in tibia(cm)						
CG	0.51 ± 0.07	0.49 ± 0.08	0.50 ± 0.07	Time	6.23	0.004
EG6s	0.51 ± 0.09	0.56 ± 0.10 *	0.48 ± 0.08 #	Group	1.20	0.310
EG3s	0.46 ± 0.10	0.48 ± 0.09	0.47 ± 0.07	Time × group	7.14	<0.001

‡ =  $p < 0.05$  from CG; # =  $p < 0.05$  from POST-1 evaluation; \* =  $p < 0.05$  from pre-evaluation; † =  $p < 0.05$  from EG6s; PT = Patellar Tendon.

The thickness of the PT in the anterior tuberosity of the tibia in EG6s was  $0.05 \pm 0.04$  cm (CI 95%, from 0.02 to 0.07 cm,  $p < 0.001$ , ES = 0.5) greater after the 6 weeks of eccentric training compared to the baseline and a decrease from POST-1 was found after 6 weeks of detraining (EG6s: difference =  $0.08 \pm 0.07$  cm; CI 95%: from 0.04 to 0.12 cm,  $p < 0.001$ , ES = 0.9).

Significant increases ( $p < 0.05$ ) in the EI at 25% of the PT were found after the six weeks of non-training compared to PRE evaluation (EG6s:  $p = 0.002$ ; and EG3s:  $p = 0.039$ ) and compared to POST-1 (EG6s: difference =  $1.21 \pm 0.77$  A.U.; CI 95%: from 0.72 to 1.69 A.U.,  $p < 0.001$ , ES = 2.2; and EG3s: difference =  $0.72 \pm 0.81$  A.U.; CI 95%: from 0.30 to 1.15 A.U.,  $p < 0.001$ , ES = 1.4). A significant increase ( $p < 0.05$ ) was found after 6 weeks of detraining compared to POST-1 in the EI at 50% of the PT (EG6s:  $p < 0.001$ ; and EG3s:  $p = 0.001$ ) and in the EI at 75% of the PT (EG6s: difference =  $1.04 \pm 1.08$  A.U.; CI 95%: from 0.50 to 1.57 A.U.,  $p < 0.001$ , ES = 1.4; and EG3s: difference =  $0.64 \pm 0.52$  A.U.; CI 95%: from 0.17 to 1.10 A.U.,  $p < 0.001$ , ES = 1.5) in both experimental groups. In the same way, EG6s showed an increase ( $p < 0.05$ ) in POST-2 from PRE evaluation in the EI at 50% of the PT (Figure 4).



**Figure 4.** Elastic properties of the patellar tendon (PT) at 25%, 50% and 75% of the length of the PT after six weeks of eccentric training (POST-1) with two execution times (EG6s = 6 s and EG3s = 3 s) and after a 6 week follow-up of detraining (POST-2). ¥ =  $p < 0.05$  from CG; # =  $p < 0.05$  from POST-1 evaluation; \* =  $p < 0.05$  from pre-evaluation.

#### 4. Discussion

The main purpose of this study was to compare the effects of the execution time of eccentric contraction (3 s vs. 6 s) on the morphological and elastic properties of the PT after a 6 week SLDSe training program. Although changes in the thickness of the PT were evidenced in both experimental groups after the completion of the SLDSe program, it is worth noting an increase of ~7% at the level of the lower pole of the patella in EG3s, and an increase of ~10% at the level of the anterior tuberosity of the tibia in EG6s. This is an important finding to be taken into account by physical therapists and sports adapters in the recovery of patellar tendinopathy, since depending on the level of the injury in the PT (proximal or distal), one execution time of the eccentric contraction or the other should be used in the SLDSe exercise.

Although the tendon is considered a poorly vascularized structure [30], it has been shown to respond to external mechanical loads by altering its biomechanical (Young's modulus) [31,32] and/or morphological (CSA) [10,33] properties. These responses depend fundamentally on the activity and intensity of the stimulus to which it is subjected [34]. To our knowledge, this is the first study that has compared the execution time effects of an eccentric contraction in SLDSe exercise on the morphological and elastic properties in different regions (lower pole of the patella, at 25%, 50% and 75% of the total PT length, and anterior tuberosity of the tibia) of healthy PT. Previous research shows that the areas of greatest increase in PT CSA are located at the proximal and distal ends [10,33,35]. This may be due to the fact that they are the areas in which the tendon is most vascularized [10]. In this respect, the differences found between both experimental groups in the ROIs analyzed may be due to the time during which the PT was subjected to stress [36]. Although participants were urged at all times to maintain a constant speed in the eccentric contraction, it is possible that longer execution times would cause the participant to spend more time with the degree of knee flexion close to 90°, thus transferring more tension to the anterior tuberosity of the tibia zone. Conversely, shorter execution times could have produced

the opposite effect, and put the PT under stress for a longer time in degrees closer to full extension. A solution to this limitation of the study could be in the placement of a digital goniometer on the knee joint that controls the instantaneous angular speed in the path of the execution of the eccentric contraction to test this hypothesis.

Previous studies in animal models have shown that eccentric exercise modifies tendon gene expression producing an increase in collagen synthesis [37,38]. In the case of human tendons, it has been shown that high loads can increase collagen synthesis 48–72 h after exercise [39], but in the bibliographic review carried out to date, studies were not found that specifically evaluate the effect of execution time of the eccentric contraction on collagen expression, its growth factors and the ability to produce hypertrophy in human tendons.

The results obtained in tendon thickness agree with previous investigations that have analyzed the thicknesses of healthy and pathological tendons [16,40]. On the other hand, although most of the investigations that have achieved hypertrophic increases in the PT have used at least twelve weeks of intervention by means of eccentric exercise [10,11,33,35], the present study, according with Abian et al. [16], shows that six weeks of intervention using the SLDSe exercise focused on its eccentric phase are sufficient to increase the thickness of the PT, independently of the execution time of the eccentric contraction performed. This may be due to the high load (80% of the eccentric 1 RM) and the type of exercise (SLDSe) used in our intervention program.

No research has been found in the literature that has evaluated the residual effects on the morphological properties of the PT after the cessation of a training program using eccentric contractions. In the present investigation, it was observed that after hypertrophy in the tendon at the end of the intervention, the tendons of both experimental groups returned to their previous values recorded at the beginning of the study six weeks after cessation of activity. This situation may be due to the fact that, in addition to an increase in the synthesis of the collagen process, there is an increase in the concentrations of internal water and additional material that are more easily lost after the cessation of activity [41].

In 2001, an increase in PT stiffness was reported for the first time after twelve weeks of training using isometric contractions [42]. Moreover, sonoelastography has proven to be a reliable method in the exploration of the EI of healthy PTs [43]. In the present investigation, no differences were observed at the end of the intervention, and increases were found in both experimental groups (EG3s and EG6s) after the detraining period. Few studies were found in the literature review showing a decrease in tendon stiffness after an intervention program using eccentric contractions [12,13]. The results in these studies found a decrease of 6–15%. It should be noted that none of the previous investigations used sonoelastography for evaluating the PT stiffness index or the detraining period, which makes it difficult to compare with the data recorded in this study.

On the other hand, transient reductions in the hydration of Achilles [44] and PTs [45] have been found after undergoing physical exercise. Further research is required to ascertain if the reduction in EI observed in EG6s after the end of the intervention period is associated with a change in the hydration status of the PT or with the realignment of the newly formed collagen fibers. In our research it was observed that six weeks after the end of the intervention, both experimental groups increased the EI of the PT in the ROIs evaluated. Other previous investigations found a reduction in the stiffness index of Achilles [46] and PTs [47] one month and two months, respectively, after completing a training program using isometric contractions. These discrepancies in the results obtained may be due to the different evaluation methodologies used and the characteristics of the intervention program, with eccentric exercise being able to cause adaptations in the stiffness index over a longer term than isometric exercise. Furthermore, the increase in EI coupled with the reduction in PT thickness found after the detraining process may be due to a change in the mechanical properties of the components that make up the tendon structure. These properties may have been modified through mechanisms, such as increased packaging of collagen fibers or through alterations in the angle projected by them [48].

The PT is considered to be a short and thick tendon whose main function is to transmit the forces generated in the quadriceps to the tibia. In addition, the PT has other important functions, such as energy storage/release in the articular loading and unloading phases and protection against muscle injuries [49]. Therefore, the increase in the stiffness of the PT found in our research could be adequate to transmit the forces quickly and efficiently, but it could affect its mechanical damping function and its elastic energy-saving capacity for economy of movement [40].

## 5. Conclusions

In conclusion, 6 weeks of SLDS<sub>e</sub> produced significant changes in the morphological properties of the PT. These adaptations were lost after 6 weeks of detraining. It was observed that longer times of execution of the eccentric contraction were associated with increases in the thickness of the PT at the distal level, and shorter times with increases at the proximal level. On the other hand, longer times of execution of the eccentric contraction induced a reduction in the tendon EI, and that regardless of the time of execution of the eccentric contraction, after 6 weeks of detraining there was an increase in the stiffness of the PT.

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Article

# Effects of 8-Week In-Season Contrast Strength Training Program on Measures of Athletic Performance and Lower-Limb Asymmetry in Male Youth Volleyball Players

Abdeltif Mesfar <sup>1</sup>, Raouf Hammami <sup>1,2</sup>, Walid Selmi <sup>1,3</sup>, Sabri Gaied-Chortane <sup>1,3</sup>, Michael Duncan <sup>4</sup>, Thomas G. Bowman <sup>5,\*</sup>, Hadi Nobari <sup>6,7,8,\*</sup> and Roland van den Tillaar <sup>9,\*</sup> †

- <sup>1</sup> Higher Institute of Sport and Physical Education of Ksar Said, Manouba University, Tunis 2010, Tunisia; mesfarabdellatif.ksarsaid@gmail.com (A.M.); raouf.cnmss@gmail.com (R.H.); selmiwalid13@yahoo.fr (W.S.); sabrigaied1@gmail.com (S.G.-C.)
- <sup>2</sup> Research Laboratory: Education, Motor Skills, Sports and Health (EM2S, UR15JS01), Higher Institute of Sport and Physical Education of Sfax, University of Sfax, Sfax 3029, Tunisia
- <sup>3</sup> Research Unit (UR17JS01) "Sport Performance, Health & Society", Higher Institute of Sport and Physical Education of Ksar Said, Manouba University, Tunis 2010, Tunisia
- <sup>4</sup> Centre for Sport, Exercise and Life Sciences, Coventry University, Coventry CV1 5FB, UK; aa8396@coventry.ac.uk
- <sup>5</sup> Department of Athletic Training, College of Health Sciences, University of Lynchburg, Lynchburg, VA 24501, USA
- <sup>6</sup> Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain
- <sup>7</sup> Department of Exercise Physiology, Faculty of Educational Sciences and Psychology, University of Mohaghegh Ardabili, Ardabil 56199-11367, Iran
- <sup>8</sup> Department of Motor Performance, Faculty of Physical Education and Mountain Sports, Transilvania University of Braşov, 500068 Braşov, Romania
- <sup>9</sup> Department of Sports Science, Nord University, 7600 Levanger, Norway
- \* Correspondence: bowman.t@lynchburg.edu (T.G.B.); nobari.hadi@unitbv.ro or hadi.nobari1@gmail.com (H.N.); roland.v.tillaar@nord.no (R.v.d.T.)
- † These authors contributed equally to this work.

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**Abstract:** Strength training using high and lower load such as contrast training (CST) seems to be beneficial as it addresses larger adaptive reserves in youth athletes. Therefore, the aim of this study was to investigate the effects of CST on dynamic balance (composite score during dynamic balance test (CS-YBT)), one repetition maximum lower-limb back squat (1RM), jumping performance (single-leg hop (SLH) or countermovement jump height (CMJ)), lower-limb asymmetry (predicted from the single-leg jump performance between two legs [ILA]) in elite youth male volleyball players. Thirty-one male youth volleyball players aged 14 years were randomly assigned to a CST group ( $n = 16$ ) or a control group ( $n = 15$ ). The tests were performed before and after 8 weeks of training. Significant group  $\times$  time interactions was observed for CS-YBT [ $p < 0.001$ ,  $\eta_p^2 = 0.70$ ], 1RM [ $p < 0.001$ ,  $\eta_p^2 = 0.95$ ], SLH with right and left leg [ $p < 0.001$ ,  $\eta_p^2 = 0.69$  and  $0.51$ ], CMJ [ $p < 0.001$ ,  $\eta_p^2 = 0.47$ ], whilst it was not notable in ILA [ $p < 0.294$ ]. Post hoc tests showed that CST group demonstrated greater improvement in all of the dependent variables from medium to large effect size (for all  $p < 0.001$ ). As a result, 8 weeks of CST twice a week can be an effective and efficient training along with volleyball training to improve skill-related fitness measures, except for lower-limb asymmetry in young volleyball players.

**Keywords:** resistance training; power exercise; team sport; conditioning capabilities; lower extremity; dynamic balance

## 1. Introduction

The performance of volleyball players is related to their ability to produce explosive actions in attack and block situations, as the goal of the game is to make the ball cross



the 2.43 m net (for males) and hit the ground, while maintaining control of balance [1,2]. For instance, in highly dynamic situations in volleyball, rapid vertical jumping, proper dynamic alignment of the center of pressure relative to the base of support is essential for successful performance [3,4]. Furthermore, due to the importance of balance and muscular strength, for necessary power development while jumping during sport participation and volleyball training [5]. In addition, volleyball players' jump performance might be hampered by strength imbalances in the lower limbs [1]. Therefore, identifying optimal resistance training methods to increase balance, muscle strength and power performance, and reduce asymmetry may be crucial to volleyball training and, indeed, performance.

Contrast training is commonly utilized by volleyball players during resistance training. Contrast strength training (CST) is characterized by the use of high and low loads in the same training session [6,7] and has previously been shown as an effective modality in improving both muscle strength and power, with adaptations in both neuromuscular function and muscle morphology [8]. For example, Smilios et al. [7] showed that contrast training program led to increases in vertical jump, sprint and agility levels in prepubertal children who exhibit high muscular strength trainability. Furthermore, Hammami et al. [6] demonstrated that both contrast strength and plyometric training programs enhanced muscle power and sprint performance at 5 and 40 m distances and change-of-direction test scores relative to controls in male youth soccer players. The authors concluded that the improvement of physical performance was greater following eight weeks of contrast strength training than with plyometric training. Whilst current research has endeavored to verify the effectiveness of contrast training on enhancing athletic performance with youth, there is a shortage of research exploring the dose–response relationship of such modality in volleyball.

In addition to performance, lower-limb asymmetries (LLA) have frequently been studied to quantify performance differences between limbs [9–12] and a bilateral asymmetry in youth [1]. Madruga-Parera et al. [13] demonstrated that contrast resistance training programs reduced interlimb asymmetries score (−0.70 moderate vs. −0.32, small), more than isoinertial resistance training in youth male handball players. While some level of asymmetry is to be expected in youth populations using both isoinertial and contrast resistance training programs, further examination of the effects contrast training between-limb differences using practically viable screening tasks is warranted. Our starting point was the fact that contrast training provides broader neuromuscular adaptations, which result in greater transfer to a wide variety of performance variables [1,6,7,13]. Cumulatively, there is a dearth of available evidence to compare the effects of CST, followed by a detraining period, on measures of dynamic balance control and lower-limb asymmetry during field-based tests with youth volleyball players. Therefore, the aim of this study was to examine the effect of eight-week CST on measures of dynamic balance performance, muscle strength and power performance and lower-limb asymmetry in youth male volleyball players.

Based on earlier longitudinal studies, we hypothesized that the CST program would result in athletic performance improvements [6–8] and reduce the LLA scores [13], compared to a control group, in young volleyball players.

## 2. Materials and Methods

### 2.1. Study Design

To assess the effects of an 8-week, in-season CST program on dynamic balance, muscle strength and power performance, and lower-limb asymmetry in male youth volleyball players, a randomized pre- and post-test group design with a CST and a regular volleyball training (control) group was used. In the present study, participants were randomly allocated to one experimental group and a control. Group allocation was realized by adjusting for, age, maturation and their performance in the CMJ and 1RM of the study sample. In addition, the order of each trial was changed randomly between participants, in

order to avoid learning effects and fatigue. The dependent variables were dynamic balance, muscle strength (1RM) and power (CMJ, SLH), and LLA.

## 2.2. Participants

Thirty-one male youth volleyball players, belonging to a first division Tunisian volleyball club (Club Sfaxien, Sfax, Tunisia), were recruited in this study. All players were randomly assigned to a CST program ( $n = 16$ ; age:  $14.4 \pm 0.6$  years; height:  $181.8 \pm 6.6$  cm; body mass:  $68.5 \pm 11.1$  kg; maturity offset:  $+1.49 \pm 0.63$  years) or a control group ( $n = 15$ ; age:  $14.5 \pm 0.5$  years; height:  $180.1 \pm 2.9$  cm; body mass:  $67 \pm 5.7$  kg; maturity offset:  $+1.36 \pm 0.43$  years). All participants had the same daily school and volleyball team-training schedules. They all had been playing volleyball on a regular basis three–four times a week (i.e., ~90 min per session), with a match played during the weekend, for more than 3 years. Maturity offset of participants was calculated according to Moore et al. [14].

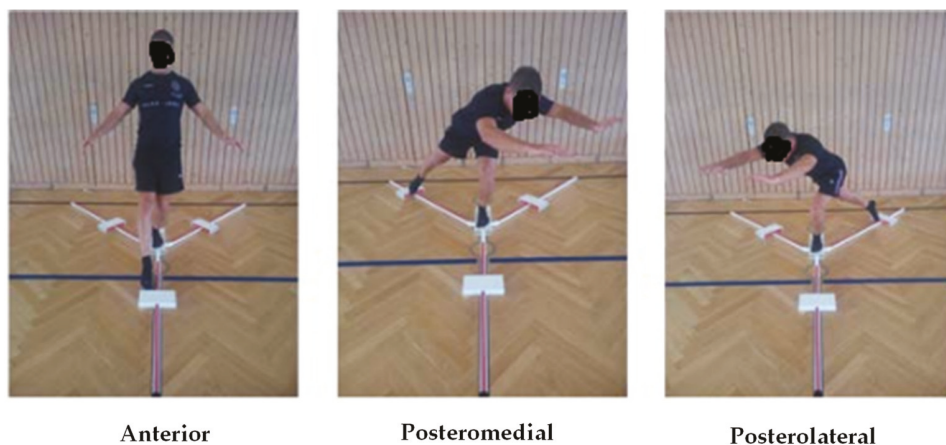
Before experimental testing, the study was conducted according to the Declaration of Helsinki, and the protocol was fully approved by the Ethics Committee of the National Centre of Medicine and Science of Sports of Tunis (CNMSS-LR09SEP01) before the commencement of the assessments. Written informed consent was obtained from parents/legal representatives of all participants before the commencement of the study.

## 2.3. Procedure

All procedures were performed during the second half of the competitive volleyball season (February and March 2020). Before any data were collected, all athletes participated in two orientation sessions to familiarize themselves with the experimental procedures to minimize any learning effect of testing. Participants were assessed for balance (CS-YBT), muscle strength (1RM half squat) and power (single-leg hop and counter movement jump) and lower-limb asymmetry, respectively, before and after eight-week CST program. Procedures were undertaken after a general warm-up that consisted of running, calisthenics, and stretching.

### 2.3.1. Dynamic Balance

Dynamic balance performance was evaluated by the Y-Balance Test [14]. All trials were conducted barefoot. Participants stood on the dominant leg, with the most distal aspect of their big toe on the center of the footplate from the YBT Kit. The participants were then asked to push the reach-indicator block with the free limb in the anterior, posterior medial, and posterior lateral directions in relation to the stance foot on the central footplate, while maintaining their single-limb stance. [14]. Data collection followed the protocol of Kang et al. [15], where participants were not allowed to lift their heel during the Y-Balance test. The allowance of heel lift could reduce the importance of ankle dorsiflexion, while requiring the heel to maintain contact with the ground would emphasize ankle range of motion [16]. Maximal reach distances were recorded to the nearest 0.5 cm marker on the Y-balance test kit (Figure 1). The trial was repeated if the participant failed to maintain a unipedal stance, or failed to return the reaching foot to the starting position. A composite score [CS-YBT (%)] was calculated using the formula by summing the three distances covered divided by three times the length of the leg and multiplied by 100. Leg length was measured from the anterior superior iliac spine to the most distal part of the medial malleolus by using a tape measure while the subject laid in supine position [14,17]. In the present study, an excellent reliability score was reported for the CS-Y balance test with the intra class correlation coefficient (ICC) value of 0.93.



**Figure 1.** The Y-Balance test.

### 2.3.2. Dynamic Strength

Muscle strength was assessed with a 1RM squat as reported by Keiner et al. [18]. Before attempting the 1RM, participants performed three sub-maximal sets of 1–6 repetitions with a light-to-moderate load (40 to 50% 1RM). Participants then performed a series of repetitions with an increased load. The increments in resistance were dependent on the effort required for the lift and became progressively smaller as the players approached their 1RM. Failure was defined as a lift falling short of the full range of motion on at least two attempts, spaced at least two minutes apart. The 1RM was then determined within 5 to 6 trials. Test–retest reliability was excellent for 1RM with an ICC of 0.98 with the present study.

### 2.3.3. Single-Leg Hop Test

Single-leg hop tests have been shown to evaluate lower-limb power performance requiring slow stretch-shortening-cycle action in accordance to the described protocol by Ramirez-Campillo et al. [19]. The test was executed using a 5 m fiberglass metric tape affixed to a wooden floor. Players were instructed to use their arms to aid in the jump phase, using a one-foot stand (right and left), and perform a fast movement (approximately 120° knee flexion angle) followed by a jump for maximal distance. All players were instructed to land in an upright position during the jumps and to flex their knees after landing. The test was executed three times for each leg, with the starting order of the right or the left leg randomly assigned, with a 1 min rest period, and the best value was recorded for analysis. In the present study, test–retest reliability scores have been shown to be good with an ICC of 0.81.

Bilateral asymmetry was calculated from the performance measure during the single-leg hop test. A negative sign (–) was arbitrarily assigned when the left leg was the stronger one, and a positive sign (+) was used when the right leg was the stronger one. In the literature [20], relative inter-limb asymmetry for the lower limbs was determined the formula:  $(\text{Right leg} - \text{Left leg}) / (\text{Right leg} + \text{Left leg}) \times 100$ . With this formula, it seems that index number 10 is more suitable for calculating the LLA between the two legs during the single-leg hop test. Test–retest reliability scores for LLA measures from the present results have been shown good with our study (ICC = 0.73).

### 2.3.4. Vertical Jump Test

Subjects were instructed to keep an upright standing position until an angle of 90° knee flexion and perform a vertical jump. Players were performed to perform the jump with a quick manner in order to maximize their performance. During the test, two trials were

carried out, with two minutes of passive recovery. The highest jump height performance was used for analysis. Test–retest reliability has been reported good for the CMJ test with an ICC value of 0.89 with the present study.

### 2.3.5. Training Program

After the pretest, participants were randomly assigned to a perform CST or to a control group. Groups were matched for anthropometrics and physical characteristics. The groups did not differ in measures of the pre-test. The CST group performed the eight-week in-season training program consisting of strength exercises, including bench press, pull over, half squat and forward lunge (Table 1). These exercises were included in the study based on the muscle groups solicited in volleyball game and training. Sessions were performed twice weekly on non-consecutive days (Tuesday and Thursday).

**Table 1.** Design of the 8-week contrast strength training program.

Weeks	CST
	(% 1RM) for All Exercises
1–2	3 sets × (2 reps at 70% 1RM + 4 reps at 40%)
3–4	4 sets × (2 reps at 80% 1RM + 4 reps at 50%)
5–6	3 sets × (2 reps at 70% 1RM + 4 reps at 40%)
7–8	4 sets × (2 reps at 80% 1RM + 4 reps at 50%)

Notes: CST = contrast strength training.

The CST program used free weight resistance training exercises at 40 to 80% of 1-repetition maximum with 3–4 sets of 2–4 repetitions. The training was based on performing a heavy load lift followed by a light load lift, in the same series and the same exercise. Participants of the control group followed their standard volleyball practice over the same duration with no strength training design. It is important to note that all participants from the two groups had regularly performed strength/power training exercises (i.e., bench press, pull over, squat, forward lunge snatch, and plyometric) during competitions and training for a minimum of 2 years before the start of the study. The volume of training remained constant for all exercises. During this period, the control group was exposed to a 15 min period of passing drills. Of note, training volume (i.e., total time of training exposure) was similar between the two groups. Qualified coaches and experienced sports scientists supervised both groups. Throughout all training exercises, the instructor-to-player ratio of 1:1 was maintained. All subjects received treatment conditions as allocated. A standardized 10 min warm-up containing jogging, dynamic stretching exercises, calisthenics, and preparatory exercises (e.g., fundamental weightlifting exercises specific to the training program) was provided for all experimental groups before the beginning of each training session. The training session lasted ~35 min and ended with 5 min of cool down activities including dynamic stretching. All groups performed regular volleyball practice during 5 to 6 sessions throughout the study and no injuries were obtained over the training program.

### 2.4. Statistical Analysis

Data are presented as means and standard deviations (SD) and normality was assessed and confirmed using the Shapiro–Wilk test. The data were then analyzed using a 2 (groups: CST and control group) by 2 (time: pre, post) analysis of variance (ANOVA) for repeated measures. Where the assumption of sphericity was violated, Greenhouse–Geisser correction was used to interpret the results. If group × time interactions reached the level of significance, post hoc tests, using Bonferroni corrections, were computed to identify the comparisons that were statistically significant. Partial eta-squared ( $\eta_p^2$ ) was used as a measure of effect size. ES can be classified as small ( $\eta_p^2 = 0.01$ ), medium ( $\eta_p^2 = 0.06$ ), or large ( $\eta_p^2 = 0.14$ ) [21]. Test–retest reliability was assessed using the ICC and the standard

error of measurement (SEM) expressed as coefficient of variation [21]. For the interpretation of ICC values, a value greater than 0.80 reflects an excellent reliability, whereas ICCs from 0.70 to 0.79 reflect a good reliability [22]. The alpha level of significance was set at  $p < 0.05$ . All data analyses were performed using SPSS 26.0 (SPSS, Inc., 288 Chicago, IL, USA).

### 3. Results

All 31 young volleyball players from CST and control group completed the study according to the study design and methodology. Participants attended all training sessions, and none reported any training- or test-related injury. There were no statistically significant between-group baseline differences found for any of the analyzed parameters (Table 2).

**Table 2.** Effects of contrast strength training on measures of athletic performance in youth.

Variables	Groups	Pre-Intervention	Post-Intervention	$\Delta$	Time	ANOVA $p$ -Value Group	ANOVA $p$ -Value Group $\times$ Time
Composite score Y-balance test (%)	CST	82.10 (5.19)	93.90 (3.36)	11.8	0.01	0.01	0.01
	Control	76.80 (5.51)	78.85 (5.37)	2.0			
One repetition maximum (kg)	CST	61.71 (2.28)	80.57 (2.26)	18.8	0.01	0.01	0.01
	Control	56.22 (2.92)	58.48 (2.59)	2.2			
Single-leg hop test right leg (cm)	CST	124.39 (19.23)	145.00 (19.66)	20.6	0.01	0.01	0.01
	Control	104.69 (5.09)	107.52 (5.09)	2.8			
Single-leg hop test left leg (cm)	CST	112.50 (4.65)	124.27 (4.11)	11.8	0.01	0.01	0.01
	Control	104.36 (5.85)	108.789 (5.91)	2.8			
Countermovement jump height (cm)	CST	32.69 (3.52)	38.94 (3.96)	6.2	0.01	0.03	0.01
	Control	32.50 (3.65)	33.50 (3.91)	1.0			
Inter limb asymmetry (%)	CST	7.83 (12.55)	12.94 (10.94)	5.1	0.01	0.19	0.29
	Control	4.69 (5.09)	7.52 (5.09)	2.8			

CST = contrast strength training.

#### 3.1. Dynamic Balance

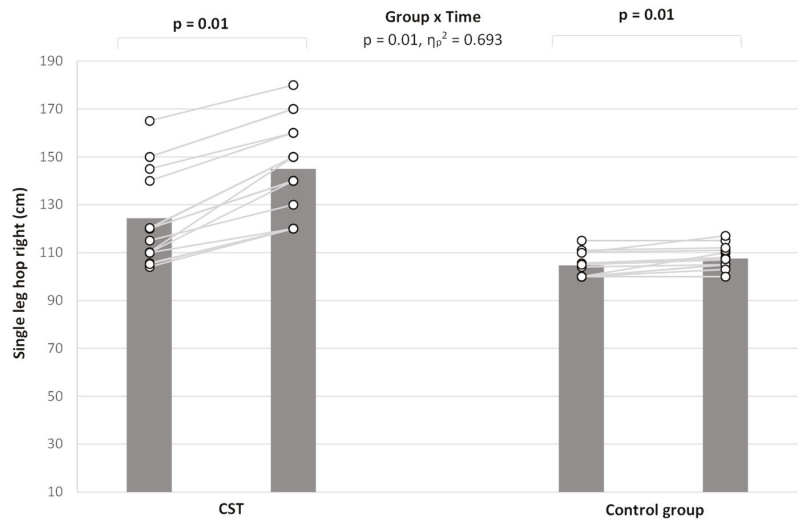
Statistical calculations revealed a significant group  $\times$  time interaction ( $p < 0.01$ ,  $\eta_p^2 = 0.703$ ) for CS-YBT. Bonferroni post hoc comparisons indicated significant increase from pre- and post-test were significant for both groups (both  $p = 0.001$ ). Percent changes in ages ( $\Delta$ ) in CS-YBT between pre- and post-test were significantly greater in CST group ( $\Delta = 11.7$ ) than control group ( $\Delta = 2.0$ ).

#### 3.2. RM Test

In view of the 1RM test, a significant group by time interaction was observed ( $p = 0.02$ ,  $\eta_p^2 = 0.945$ ). Bonferroni post hoc pairwise comparisons indicated significant differences at pre- and post-test between groups ( $p = 0.001$ ). The increase pre- and post-test was significant for both intervention ( $p = 0.001$ ) and control groups ( $p = 0.003$ ) but the magnitude of change was greater for the intervention group ( $\Delta = 18.8$ ) compared to the control group ( $\Delta = 2.2$ ).

#### 3.3. Single-Leg Hop Test

A significant group  $\times$  time interactions were also noted ( $p < 0.01$ ,  $\eta_p^2 = 0.693$ ) (Figure 2). Bonferroni post hoc pairwise comparisons indicated significant differences at pre and post between groups ( $p = 0.001$ ). The increase pre and post was significant for both intervention and control groups (both  $p = 0.001$ ), but the magnitude of change was greater for the intervention group ( $\Delta = 20.6$ ) compared to the control group ( $\Delta = 2.8$ ).



**Figure 2.** Participants performance before and after intervention period of single-leg hop right performance by the two groups. CST: contrast strength training group.

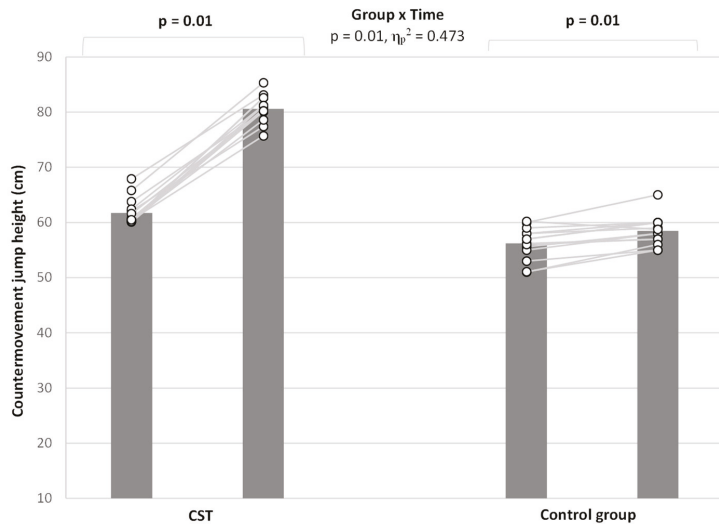
Similarly, a significant group  $\times$  time interactions was found for the SLHL ( $p < 0.02$ ,  $\eta_p^2 = 0.511$ ). Bonferroni post hoc pairwise comparisons indicated significant differences at pre and post between groups ( $p = 0.001$ ). The increase pre and post-test was significant for both intervention and control groups (both  $p = 0.001$ ), but the magnitude of change was greater for the intervention group ( $\Delta = 11.7$ ) compared to the control group ( $\Delta = 2.8$ )

### 3.4. Countermovement Jump Height

A group-by-time interaction was observed significant for the CMJ height ( $p < 0.02$ ,  $\eta_p^2 = 0.473$ ) (Figure 3). Analyses demonstrated a medium improvement in CMJ height before and after training in favor of the CST group ( $p < 0.01$ ,  $\eta_p^2 = 0.158$ ). Bonferroni post hoc pairwise comparisons indicated no significant differences at pre-test between groups ( $p = 0.88$ ) but a significant difference post between groups ( $p = 0.01$ ). The increase at pre- and post-test was significant for both intervention and control groups (both  $p = 0.01$ ), but the magnitude of change was greater for the intervention group ( $\Delta = 6.2$ ) compared to the control group ( $\Delta = 1.0$ ).

### 3.5. Interlimb Asymmetry

There was no significant group  $\times$  time interaction ( $p = 0.29$ ) but there was a significant main effect for ILA ( $p = 0.01$ ,  $\eta_p^2 = 0.331$ ). Bonferroni post hoc pairwise comparisons indicated a significant increase in ILA from pre- to post-test ( $6.2 \pm 1.7$  vs.  $10.2 \pm 1.6$ , mean diff = 3.968).



**Figure 3.** Participants performance before and after intervention of CMJ performance by the two groups.

#### 4. Discussion

The aim of this study was to compare the effect of 8-week in-season CST on dynamic balance, muscle strength and power performance and lower-limb asymmetry scores in youth male volleyball players. The main findings were that the CST group improved in all physical fitness parameters, compared to the control group, with the exception of lower-limb asymmetry.

The results demonstrated an improvement of dynamic balance performance in the CST group. Resistance training is an effective and safe mode of training for children and adolescents with positive effects on balance and stability [2,23,24]. Although balance and coordination performance are not yet mature in pediatric populations [25], it is possible that greater enhancement of dynamic balance performance with contrast strength exercises could lead to a greater muscle power output in this population. Particularly in the CST group, the coordinated postural control to mobilize a load with an important range of motion and with a force production necessitates higher level of balance and muscle strength and power capabilities. In the current study, CST was performed under less stable conditions with high-speed dynamic contractions performed within a more limited base of support or with the center of gravity being moved outside the base of support, which would be affected to a much greater extent by balance and strength/power output. Thus, the participants in this study positively responded to these balance stressors with demonstrable enhancement in dynamic balance performance, to a greater extent with contrast training.

The present results demonstrate that 8 week of contrast training resulted in an improvement in 1RM half squat and muscle power performance (i.e., CMJ) and SLH) in the CST group to a greater extent than the control group. The results of the present study are in agreement with prior investigations which have found an enhancement of 1RM half-squat and power performance after contrast strength training [1,6,26,27]. Vissing et al. [27] suggested that power training-induced improvements in all 3 tests of maximal strength (leg extension; knee extension; and hamstring curl), and demonstrated these improvements during shorter training periods or higher initial training status of participants. Because youth athletes rely less on their glycolytic metabolism [2], have a weak hypertrophic response [24] and lower type 2 fiber composition [2] in comparison with adults, it has been suggested that

high-speed strength and power training programs, such as contrast training, may benefit youth athletes to enhance muscle strength and power performance. Accordingly, the effects of CST thus seem primarily due to neuromuscular adaptations, such as more effective motor unit recruitment, rate coding (frequency or rate of action potentials), synchronization, and intermuscular coordination [27,28].

Our results demonstrate that 8 week of CST did not reduce LLA scores in youth volleyball players. However, the range of LLA in the present study were low, and lower than the previously reported figures for runners of  $\geq 10\%$  [29,30]. The training process for volleyball in youth players emphasizes movement in a symmetrical manner. Coupled with the age of the youth participants involved, it is less likely that asymmetrical movement patterns had been developed, and therefore, we would not necessarily expect to observe a difference in LLA for this group because of the training stimulus that was applied. Given that balance and coordination are not fully developed in youth athletes [25], the implementation of 8 weeks of CST in youth resistance training, as implemented in the current study, should not be considered effective in reducing LLA in youth volleyball players.

Although the results of the present study should be considered a novel addition to the literature, this study is not without limitations, which should be mentioned accordingly. First, the results obtained should only be generalized to similar samples of participants. Furthermore, for a better understanding of underlying physiological mechanisms of adaptations associated with CST, a longer intervention duration, along with the evaluation of neuromuscular and muscle adaptations using electromyography, ultrasound, or other imaging technology, should be employed. Another limitation is that this study did not compare or contrast with plyometric and traditional strength training. Such a comparison would be useful in future research because previous investigations have indicated that differences in the adaptive responses between these methods may exist. Second, the sample size did not allow for participants to be grouped according to maturity status. This should be controlled for in future studies. Finally, the sample size of each groups was small. Therefore, this study is preliminary. However, it is difficult and almost impossible to recruit large sample sizes in elite sport, especially in a highly professionalized elite sport such as volleyball.

## 5. Conclusions

The current study shows that in youth volleyball players, after 8-week, the CST group had larger improvements in dynamic balance and muscle strength and power performances, to a greater extent than the control group. However, CST was not effective in enhancing LLA. Combining a specific stimulus, such as CST, into training sessions seems to be a safe training modality in this age cohort and facilitates continued progressive neuromuscular adaptation. Practitioners should include specific strength exercises, such as combining strength and power exercises with a progressive overload using CST to optimally enhance athletic performance in youth volleyball players. On that basis, coaches and key stakeholders of youth athletes are advised to add CST to their training routines with a view to maintain, and enhance, athletic performance in an optimal fashion to reduce LLA. This suggests that there could be interdependent positive transfer effects, from training that is singularly focused on strength and conditioning. Despite this, the present findings are based on longitudinal data which does conclusively allow for cause and effect to be determined. Furthermore, the findings suggest that when designing training programs aimed to improve athletic performance in volleyball players, coaches should pay attention to this specific adaptation, which can necessitate an individualized approach to program design.

### *Practical Applications*

- The result of the CST intervention shows that youth volleyball players had larger improvements in dynamic balance and muscle strength, power performances, but not in LLA after 8 weeks.



- CST seems to be a safe training modality in youth age cohorts, and facilitates continued progressive neuromuscular adaptation.
- Coaches and key stakeholders of youth athletes may add CST to their training routines with a view to maintaining, and enhancing, athletic performance in an optimal fashion to reduce LLA.

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**Data Availability Statement:** The data presented in this study are available on reasonable request from Abdelatif Mesfar. Requests for access to data should be sent to mesfarabdellatif.ksarsaid@gmail.com.

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Article

# Executive Function Level in Cadets' Shooting Performance

Dariusz Jamro <sup>1</sup>, Grzegorz Zurek <sup>2,\*</sup>, Malgorzata Dulnik <sup>2</sup>, Maciej Lachowicz <sup>2</sup> and Dariusz Lenart <sup>1</sup>

<sup>1</sup> Department of Physical Education and Sport, General Tadeusz Kosciuszko Military University of Land Forces, 51-147 Wrocław, Poland; [dariusz.jamro@awl.edu.pl](mailto:dariusz.jamro@awl.edu.pl) (D.J.); [dariusz.lenart@awl.edu.pl](mailto:dariusz.lenart@awl.edu.pl) (D.L.)

<sup>2</sup> Department of Biostructure, Wrocław University of Health and Sport Sciences, 51-612 Wrocław, Poland; [mal.gosia009@gmail.com](mailto:mal.gosia009@gmail.com) (M.D.); [maciej.lach93@gmail.com](mailto:maciej.lach93@gmail.com) (M.L.)

\* Correspondence: [grzegorz.zurek@awf.wroc.pl](mailto:grzegorz.zurek@awf.wroc.pl)

**Abstract:** Executive functions (EF) are crucial to a person's unique abilities, enabling one to achieve goals, adapt to new situations and manage social interactions. EF are also very important for the effective performance of military tasks including the shooting performance (SP) of soldiers. The aim of this study was to investigate the association of EF with SP and gender differences in the level of these traits among cadets of the General Tadeusz Kosciuszko Military University of Land Forces in Wrocław i.e., 156 persons (19 females and 137 males). The level of EF and processes related to attention was measured with usage of the Color Trails Test (CTT-1 and CTT-2). SP was assessed on the basis of scores from four different small arms and rifle shootings at a fixed target and at emerging targets. The relations between explained and explanatory variables were assessed using Spearman correlation. The variation in the mean values of CTT scores and SP of men and women was compared using the Mann–Whitney U test for independent samples. The results of the present study did not reveal any significant differences between women and men in the level of EF and SP. The key finding of the present study is that the higher SP of males in all shooting events of the study and of females in pistol shooting were significantly correlated with higher executive functions.

**Keywords:** executive functions; shooting performance; gender differences; cadets

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## 1. Introduction

Executive functions (EF) are processes that allow the control of complex, conscious as well as intentional tasks. Thanks to them, humans are able to monitor and regulate their behaviour and perform so-called intentional activities [1]. These activities are particularly identified with the frontal lobe of the cortex and the cortical–subcortical neuronal network, but other areas are also involved in their regulation, i.e., the dorsolateral prefrontal cortex, anterior cingulate cortex or insula [2,3]. EF include cognitive abilities that enable a person to store information in working memory, inhibit automatic responses to stimuli (inhibitory control) and shift attention between related (attentional shift flexibility) but distinct aspects of a task or problem. Executive abilities, or cognitive control abilities, allow individuals to inhibit behaviors, focus attention and organize thoughts in the face of distraction, task complexity and stress [4].

The level of EF is positively influenced by sports participation. Studies of athletes' brains show that enhanced neuronal networks and plastic changes are induced by the acquisition and execution of complex motor skills during daily intensive physical training. This training often requires rapid stimulus discrimination, specific attention and decision making. It is likely that the mode of neural modulation varies depending on the sport practised. Studies also confirm that open sports (e.g., basketball) can partially compensate for impaired executive control in people with limb impairments by supporting stability of motor responses and fostering flexibility of responses [5,6].

EF are also very important for the effective performance in military tasks performed by soldiers, including shooting abilities [7]. Military personnel are often required to engage

in complex higher order cognitive tasks (working memory, inhibitory control, cognitive flexibility, planning, reasoning and problem solving) [8]. This occurs during physically demanding and stressful military exercises and especially in combat tasks under time pressure and threats to life and health. These tasks may include, but are not limited to, complex topographical orientation, decision making, memorising operational layouts and procedures or performing effective shooting tasks. The ability to maintain a high level of EF is certainly an important element of success among soldiers [9,10].

However, it is still unclear how and whether there are gender differences in the level of EF. Due to methodological variability and the involvement of multiple neuronal networks, it is not possible to make a simple, clear statement regarding differences between men and women in this area [11]. The literature provides divergent information regarding gender differences in CTT performance. Results from a study using the above instrument among 163 healthy participants aged 19–75 years showed a significant influence of age and education level on time to complete both parts of the CTT (higher age and lower education level contributed to slower time to complete both parts), while gender had no effect on time to complete Part B [12]. Data obtained from a U.S. standardized sample (1528 subjects, including 182 African-Americans and 292 Hispanics, ranging in age from 18 to 89 years) also confirm that the influence of gender on CTT scores is not significant; however, increasing age and lower education have been shown to adversely affect performance on this neuropsychological test [13]. Statistically significant differences between men and women were also not found by Konstantopoulos et al. and Hsieh and Tori [14,15]. Contrastingly, in other studies normalizing the Brazilian population, the data on CTT-1 and CTT-2 performance time by men and women were significantly different. Women presented higher mean scores (completion time), which corresponded to poorer performance [16]. Overall, gender does not seem to have a significant effect on CTT results.

Shooting efficiency, together with psychophysical efficiency, makes up the overall combat training of soldiers [17]. It involves accurate, i.e., effective shooting from individual weapons, and is a highly important skill, necessary to be mastered by every soldier. This activity, in connection with weapon handling, requires well-developed small motor skills [18,19]. Shooting is also a closed motor skill [20,21], which requires from a shooter highly developed anti-interference abilities, i.e., focus of attention and high mental intensity. Correct shooting actions (mainly pulling down the trigger tongue and maintaining a stable stance) are particularly associated with high demands on executive and inhibitory functions. In addition, the shooter updates his current behaviour with previous experiences in order to achieve a high score in the shooting task [22].

Often when shooting, shooters are exposed to stressful situations, e.g., during competitions, military exercises, difficult weather conditions or, finally, in warfare. In such situations, executive processes must occur at the highest level; this is necessary for the effective performance of the task [7,23,24]. During the firing of a shot it is also important to focus on the target, with simultaneous control of the body posture and such a positioning of the fingers to keep control over the trigger of the pistol. For this reason, both cognitive abilities and vigilance as well as appropriate motor skills are necessary to make an accurate shot [25]. For this to happen, alternating attention, which is one of the components of attention as a cognitive function, is also of considerable importance. Thanks to it, the shooter can control and coordinate all the activities mentioned above [26].

Various determinants of SP are sought in research; one of them is gender. Reports from the literature on gender differences in SP are divergent on this issue [27,28]. In a study by Kemnitz et al., female and male soldiers did not differ in their SP, although the men in the study sample had significantly less body fat, slimmer arms and were generally physically stronger than the female participants [29]. In the authors' subsequent study, no significant differences were again found between male and female soldiers in accuracy or shooting precision, although it was initially suspected that this difference might be due to the size of the weapon (carbine weight and barrel length), which might be worse for women with less

arm strength. As it has been proved, the above parameters proved to be significant for SP, however, irrespective of the gender [27].

Research on gender differences in SP has also been conducted in groups of athletes, where a good opportunity is a championship competition such as the European Championships. Mon-López et al. similarly showed no significant differences in SP in both rifles and air pistols. The lack of significant differences, according to the researchers, confirms that physical strength is an insignificant factor influencing performance in sport shooting, so the determinants of SP should be sought in other factors, perhaps in the shooter's cognitive performance zone [30]. SP was also analysed after intensive exercise and caffeine consumption among reservists, where again no gender differences were found [31].

Gender differences in SP are extremely important in the police community where effective skill in the use of a personal weapon can often be decisive for one's own and others' safety. In fact, police officers may find themselves in a situation of direct danger on any duty day. Contrasting with the above literature reports, the results of studies in these populations indicate that male police officers shoot more effectively with handguns compared to females. The main factor attributed to the above differences is grip strength, which is potentially important to pistol shooting accuracy [28,32].

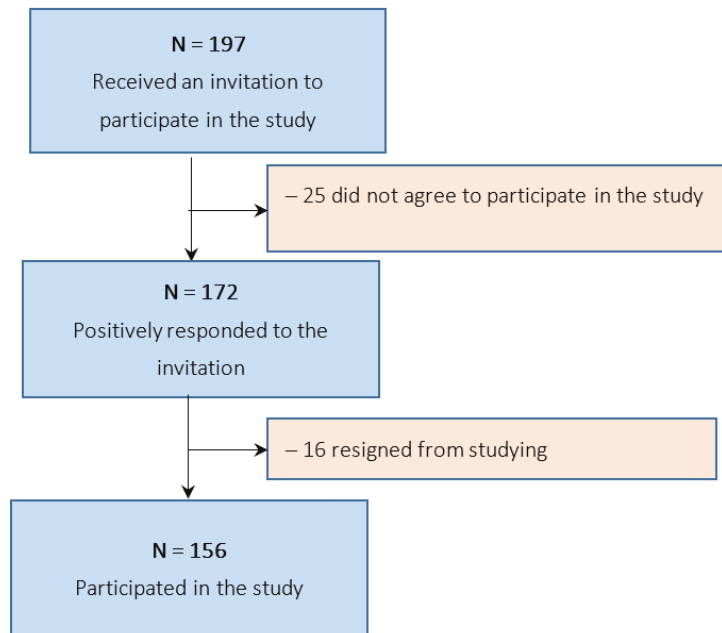
In another study, Johnson and Merullo showed that while men maintained the same accuracy during 3-h sentry sessions, accuracy in women deteriorated after 1.5 h. The shooting sessions consisted of detecting and shooting at targets that appeared infrequently [33]. In a recent study by Mon-López et al. comparing the performance of 704 shooters who participated in the recent World Shooting Championships, it was shown that men's performance in pistol shooting was better than the performance of women. However, men and women performed the same in the overall analysis, while their performance differed by category and competition [34]. Researchers have also mainly focused their attention on rather obvious differences in physical factors such as upper body strength, grip strength, balance and coordination, important for high SP, neglecting the shooter's cognitive abilities, which are potentially important [32].

To our current knowledge, the available literature does not provide sufficient information on the link between EF and SP. The issue of gender differences in the level of EF as well as differences in soldiers' SP also requires supplementation and continuous analysis. The present study may therefore complement the literature with research in military populations. At the same time, the importance of conducting scientific research in the group of soldiers is emphasized, as the results may be particularly important from the point of view of benefits for national defence. The aim of this study was to investigate the association of EF with SP and gender differences in the level of these traits among cadets of the General Tadeusz Kosciuszko Military University of Land Forces in Wrocław (MULF). We hypothesized that regardless of gender, cadets' SP level would be related to their EF level.

## **2. Materials and Methods**

The study group consisted of cadets—first-year students of the MULF. Cadets were in the course of their candidate military service and after 5 years of training they will start their professional military service. The main effect of education at MULF is the possession of appropriate knowledge and competences by graduates required to take up their first command positions in the officer corps.

The criterium for inclusion in the study was obtaining promotion to the third semester of study. The respondents gave written, informed consent to participate in the study and were fully informed about the purpose of the study. Initially, 172 cadets fulfilling the study inclusion criteria were enrolled in the study. During the course of the study, 16 cadets dropped out of the study as they opted out of further military service. Ultimately, 156 individuals (19 females and 137 males), i.e., all cadets meeting the inclusion criteria, were included in the study. The study was conducted at the MULF in the period May–July 2021 (Figure 1).



**Figure 1.** Flowchart of participant enrolment.

### 3. Executive Functions

Measures of the level of EF and attention-related processes were the results of the Color Trails Test (CTT-1 and CTT-2). The Color Trails Test contains numbered coloured circles and language symbols with wide cross-cultural applicability (no language influence). The colours used in the CTT are universal. The visual stimuli in the CTT are circles with the numbers 1 to 25 written in the middle. Each circle is coloured yellow or pink. These colours are also seen by people with colour blindness. In the first part of the test (CTT-1), all odd numbers are in the pink circles and all even numbers are in the yellow circles. In the second part of the test (CTT-2), each number is printed twice, once in a yellow circle and once in a pink circle. The CTT sheets are printed on white paper measuring  $21.59 \times 27.94$  cm. The universality of the CTT is due to the use of numbers and colours as symbols, with little involvement of speech and knowledge. The CTT is intended for adults (18+) and is designed to avoid making correct performance dependent on the knowledge of any alphabet and, to the greatest extent possible, on the influence of language [13,35].

The CTT-1 asks the participant to connect the circles in order from 1 to 25 with a rope as quickly as possible without taking the pencil off the paper. Before proceeding to the main task, the subject performs a trial task as fast as they can. The measure of the CTT performance is the time (in seconds) to correctly complete the task of connecting all circles from 1 to 25. The time is measured from the moment the test subject brings the pencil close to the first circle (starts the test). The stopwatch switches off as soon as the test subject touches the outer edge of the last circle with the pencil. In CTT-2, the tested person is asked to line up the numbered circles as fast as possible, taking into account the condition of colour alternation (pink circle 1, yellow circle 2, pink circle 3, etc.). As in CTT-1, a test task is performed before the main task.

The study was carried out among healthy individuals with no diagnosed clinical problems. Temporal indices of CTT performance are used to measure functions related to frontal lobe brain function. In interpreting the CTT results, a variety of processes related to attention and EF were examined, and, in particular, intentional search for material,

sustained and metastable attention, sequential processing of information and monitoring of own behaviour were assessed [26].

The CTT was performed by a psychologist, took place in a lecture room, always under the same conditions (psychologist–subject) and at the same time of day. The time of task performance was measured with an accuracy of 1 s. The correctness and evaluation of the CTT performance was checked twice.

#### 4. Shooting Performance

SP was assessed on the basis of the results of four different small arms and rifle shootings at fixed and emerging targets:

1. Rifle Shooting (RS)—consisted of shooting from a rifle in a lying position with the use of a stand on a stationary target 100 m away. The shooter had 5 cartridges and his task was to shoot with single fire. Accuracy and focus were important in this test. The score was determined by a number of points ranging from 0 to 50.
2. Shooting with a military pistol (PS)—this consisted of shooting from a military pistol in a standing stance at a stationary target 15 m away. The shooter had 5 cartridges; his/her task was to shoot with single fire; the shooting was performed with accuracy and focus. The score was determined by a number of points from 0 to 50.
3. Machine Pistol Shooting (MPS)—on the command “forward” the shooter marched or ran 10 m from the starting line to the firing line, assumed a shooting stance, prepared to fire and then began shooting. The first target was sighted 30 s after the command “forward” was given. The target appeared 5 times at a distance of 75 m. The time for each target to appear was 30 s. The interval between target appearances was 10 s. The score was determined by the number of hits on the target (score 5—4 hits, score 4—3 hits, score 3—2 hits).
4. Shooting from a rifle in a gas mask (RSG-M)—on the command “forward” the shooter marched or ran 10 m from the starting line to the firing line, assumed a shooting stance, prepared for shooting and then, after the first target appeared, started shooting. The first target appeared 30 s after the command “forward” and shooting was in short bursts while lying down with support. The target appeared 5 times at a distance of 100 m. The time taken for each target to appear was 30 s. Intervals between target appearances lasted 10 s. The shooter performed all actions in a gas mask. The score was determined by the number of hits on the target (score 5—4 hits, score 4—3 hits, score 3—2 hits).

All subjects had the same shooting experience resulting from the same military uniform training program. Every effort was made to ensure that shooting always took place in similar weather conditions and at the same time of day. The basis for conducting the tests was the consent of the Rector—Commandant of the university (No. 271 of 18 January 2021) and the consent of the Senate Committee on Research Ethics of the University of Health and Sport Sciences in Wrocław (No. 2/2021 of 12 February 2021). All procedures performed in this study involving human participants were in accordance with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Written informed consent was obtained from all participants included.

#### 5. Statistical Analysis

The collected results were subjected to statistical analysis. The normality of the distribution of individual variables was assessed using the Kolmogorov–Smirnov test. Of the variables analysed, the normal distribution was held by the shooting scores from all the shootings from the study in the women’s group and the scores from all the shootings and the time achieved in the CTT-1 in the men’s group. Non-normal distribution had CTT-1 and CTT-2 test scores in the female group and CTT-2 test scores in the male group.

The relationships between the explained and explanatory variables were evaluated using the Spearman correlation because not all variables analysed in the study had normal



distributions. The correlation of each individual shooting score with CTT test scores was evaluated separately due to the different shooting conditions of the individual shooters and the different types of weapons. Different battlefield situations force a soldier to perform different shooting tasks, from shooting on full rest without additional stressors to shooting under conditions of very high physical effort, limited visibility or under time and danger regime. The different results of the correlation analysis are, in a way, a confirmation of the validity of this choice. This is because the authors wanted not only to see if there were relationships between EF and SP, but also to try to determine precisely whether, if such relationships existed, EF equally affected each shooting modality of the different weapons. The variation in mean values of CTT scores and SP of men and women was compared using the Mann–Whitney U test for independent samples.

Statistical significance was assumed at the level of  $p < 0.05$  for all the applied tests. Calculations were performed using Statistica v. 13.1 software by StatSoft (Wroclaw, Poland) in the Biostructure Research Laboratory of the University of Health and Sport Sciences in Wroclaw, certified according to ISO 9001.

## 6. Results

The variation in the mean values of the tested variables by Mann–Whitney U test is presented in Table 1. As a result of the analysis, no significant differences in the mean CTT scores as well as in the SP of individual shooters were found between men and women. However, when comparing the mean scores of males and females, a lower arithmetic mean of the times achieved in CTT-1 and CTT-2 as well as higher SP in three out of four shooting events among females was observed. However, as mentioned above, these differences were statistically insignificant, so they can only be treated as a trend requiring possible further confirmation in subsequent studies.

**Table 1.** Variation in mean CTT scores and shooting performance by Mann–Whitney U test for independent samples between men and women. Variation coefficients in bold are significant with  $p < 0.05$ .

Variable	Men (N = 137)			Women (N = 19)			U Mann–Whitney Test
	$\bar{x}$	sd	v	$\bar{x}$	sd	v	<i>p</i>
Age (years)	21.03	1.31	6.22	21.15	1.08	5.11	0.3683
Body height (cm)	179.17	5.58	3.11	167.27	4.40	2.63	<b>0.0000</b>
Body mass (kg)	76.82	7.69	10.01	61.83	3.41	5.52	<b>0.0000</b>
CTT-1 (s)	31.12	7.99	25.66	28.58	10.68	37.37	0.2105
CTT-2 (s)	59.84	12.54	20.95	58.74	11.11	18.92	0.8550
RS (score)	36.64	5.90	16.10	36.16	5.74	37.37	0.4255
PS (score)	34.53	8.63	24.99	35.53	9.32	18.92	0.5576
MPS (grade)	4.46	0.84	18.84	4.89	1.13	15.87	0.0651
RSG-M (grade)	4.39	0.93	21.08	4.79	0.32	26.24	0.0745

RS—rifle shooting, PS—pistol shooting, MPS—machine pistol shooting, RSG-M—shooting from a rifle in a gas mask.

The analysis of simple correlations between the level of EF and attention-related processes and the SP of male and female cadets is presented in Table 2. Of the four shooting events, only military PS was found to be statistically significantly negatively correlated with CTT-2 scores. Higher military PS scores were significantly associated with shorter CTT-2 performance in the female group. Such a high correlation coefficient ( $r = -0.76$ ), indicates a very strong interdependence of the studied variables. The remaining correlations in the female group were statistically insignificant.

**Table 2.** Spearman correlation results between the study variables. Correlation coefficients in bold are significant with  $p < 0.05$ .

Variable	Men		Women	
	CTT-1	CTT-2	CTT-1	CTT-2
RS	<b>−0.23</b>	<b>−0.47</b>	−0.20	−0.35
PS	−0.10	<b>−0.51</b>	−0.21	<b>−0.76</b>
MPS	−0.13	<b>−0.39</b>	−0.16	0.28
RSG-M	−0.03	<b>−0.18</b>	−0.11	−0.28

RS—rifle shooting, PS—pistol shooting, MPS—machine pistol shooting, RSG-M—shooting from a rifle in a gas mask.

In the men's group, higher SP in RS was significantly correlated with shorter CTT-1 time. This is evidenced by the negative correlation coefficient, but the strength of these correlations was weak ( $r = -0.23$ ). The results from the other shootings in the male group were not significantly correlated with the time achieved in the CTT-1 attention test. The correlation results, however, revealed mutual correlations between all the shooting results from our study and the time achieved in CTT-2. SP appeared to be significantly negatively correlated with CTT-2 performance, so in the male group, higher SP was significantly correlated with shorter CTT-2 performance time. Strong correlations occurred between RS ( $r = -0.47$ ) and PS ( $r = -0.51$ ) and CTT-2 performance. The correlation between MPS performance and CTT-2 time was at the average level ( $r = -0.39$ ). A weak but also significant correlation occurred between RSG-M scores and time in the CTT-2 ( $r = -0.18$ ).

## 7. Discussion

EF help to resist strong internal tendencies and external stimuli, including controlling attention, behaviour, emotions and thinking, and focusing on ongoing action to make appropriate behavioural decisions [36]. The results of our study did not reveal significant differences between women and men in the level of EF. The literature provides divergent information regarding gender differences in CTT performance. Statistically significant differences were not found by Konstantopoulos et al. or Hsieh and Tori [14,15]. Different data were presented by Rabelo et al., who obtained a statistically significant difference in favour of men [16]. In another study, Gaillard et al. conducted a systematic review of the literature aimed at summarising the current evidence on sex differences in three domains of EF: performance monitoring, response inhibition and cognitive set-shifting using functional neuroimaging tools (fMRI, PET, EEG and NIRS). A meta-analysis of 21 studies, involving a total of 677 women and 686 men, indicated that due to methodological variability and the involvement of multiple neuronal networks, it was not possible to provide a simple, binding statement on the differences between men and women in levels of EF. However, there is now evidence of sex differences in the neural networks underlying all EF considered in this review, suggesting that men and women use different strategies depending on the demands of the task. However, functional neuroimaging, although a highly detailed and valuable study, may not be sufficient to identify sex differences in the level of EF, so work using neuropsychological tests such as the CTT may complement the above studies [11].

Comparative value with the results of our own research in the context of gender differences in the level of EF is provided particularly by data from standardisation trials for the CTT. The results of the American standardisation trial among 1531 individuals (male = 1345, female = 183) confirmed that, regardless of age group, no gender factor influence on CTT scores was found. It should be noted that women in this sample constituted 12% of the total study population, as in our study [13].

On the other hand, in other studies normalizing the Brazilian population, the data on CTT-1 and CTT-2 performance time by men and women were significantly different. Women presented higher mean scores (completion time), which corresponded to poorer performance [16]. A subsequent study among a Greek population of healthy subjects (men = 79, women = 84) showed little effect of gender on CTT-1 performance time (women

performed relatively worse compared to men). This contradictory finding compared to the results of our own study may be attributed to the fact that women in the above studies had lower levels of education compared to men [14]. The literature confirms that education has a significant effect on the level of EF tested by the CTT test [16,37]. In our study, men and women represented the same level of education resulting from the same educational program. It should additionally be noted, as is usually the case with normative research in studies, the participants were from a wide age range, while in our study the study group was a first-year military community in the age range (20–26 years).

Studies in military settings confirm that men perform better in military tasks, especially those requiring prolonged use of strength and endurance [38,39]. However, it is still not entirely clear whether gender differences are equally obvious in such specific tasks as shooting. Shooting is one of the most important skills indicative of a soldier's preparedness, which translates into the combat capabilities of a military unit. Since women began to join the ranks of armies in various countries, a discussion has begun about their role in combat operations. Researchers began to focus their attention mainly on rather obvious physiological differences, neglecting cognitive abilities and various elements of combat training including shooting efficiency. According to our current knowledge, the literature on the subject is quite poor in cross-gender comparative analysis of soldiers in terms of SP. In our study, no significant differences in shooting efficiency were observed between men and women. Despite the fact that in three out of four shootings women achieved slightly better results, these results were not statistically significant. Similar results were observed in their study by Kemnitz et al., who evaluated the effect of gender on shooting accuracy in a group of 15 male and 13 female soldiers. The Noptel simulator was used to assess accuracy (distance of shots from the centre of the target) and precision (distance of shots from each other regardless of distance from the centre of the target). As in our own study, no significant differences were found in any of the measures of SP according to gender. Although the above results confirm the reports from our own study, it should be noted that it was conducted under different shooting conditions. The main difference consisted in shooting from a simulator, whereas the shooting in our study took place on an open range; additionally, different weapons were used and different targets were shot at different distances. However, it can be concluded that women and men do not differ in SP regardless of the different shooting conditions [27].

In another study involving 292 shooters who competed in the 2016 and 2018 European Championships, men and women shot equally well with rifles, and although men's average pistol scores were higher than women's, the difference was not statistically significant. It was concluded that in sports where physical strength is a less important factor, as in the case of sport shooting, the rules should be revised for greater gender equality [30]. The above results are consistent with those of our own study, but the fundamentally different shooting conditions and the different type of weapons should be noted. In the study cited above, participants dressed in a special shooting suit shot with an air rifle, whereas in our study soldiers shot with firearms in tactical gear. In contrast, a study by Goldschmied et al. found no differences in the performance of men and women in shooting either an Olympic air rifle or a 22 caliber rifle in shooting competitions. The authors justify this on the grounds that "in shooting, the physical demands on athletes are relatively low". The study by Goldschmied et al. corresponds with the results of our own study confirming the lack of significant differences in SP among both shooters with low shooting experience and at the highest competitive level [40].

Interesting results were presented by Vučković et al., who, in a group of male and female police officers, determined the effectiveness of a basic training program in the use of small arms. During the three stages of the study, i.e., at the beginning, in the middle and at the end of the shooting training, significant differences in SP between men and women emerged only at the beginning of the training. Moreover, the same shooting training program increased the final SP by 136.43% among women, while the increase was 45.69%

among men. Thus, the above results confirm that women do not differ from men in SP in both long and short arms [41].

However, some studies report gender differences in small arms SP. Anderson et al. found that male police officers performed better with pistols than female police officers [32]. Similar results were obtained by Copay et al., who observed that males performed better than females in shooting 9 mm, 0.40 inch (.40 Smith & Wesson) and 0.45 inch (.45 Automatic Colt Pistol) pistols [28]. The different shooting effects are attributed to the difference in grip strength, although its effect on SP was small. The results of our own study also do not agree with other studies that found that men performed better than women in shooting with 22 caliber rifles at a distance of 50 metres [42] and in military conditions with rifles [33]. The military study, unlike our own work, was conducted on a Weapon simulator. The results of this study showed that during the first 1.5 h of guard duty, women shot as accurately as men. It was only after 1.5 h that their rifle shooting accuracy deteriorated and they did less well with accurate shots on target, with no deterioration in reaction time in the form of detecting the target and firing the shot. According to the authors, this difference may be due to gender differences in hand stability or to possible weaker upper body strength (causing greater fatigue after a longer duration of the combat task). However, both hypotheses are speculative and require further research.

It seems crucial to note in our study that the higher shooting scores achieved by males in all shooting tasks of the study and by females in PS were strongly significantly associated with higher EF. The strongest significant correlations occurred in carbine and pistol shooting in the male group, which was probably caused by the shooting conditions consisting in firing at a fixed target with accuracy and focus without a time regime and on full rest. A weaker correlation occurred in the machine pistol shooting, in which there was another stress factor of moving quickly to the shooting position and the target appearing in a limited time, which was undoubtedly a big limiting factor for the shooter [7]. These factors revealed that in more dynamic shooting, further variables related to SP, such as physical fitness, are likely to emerge alongside EF.

The weakest significant correlations occurred in shooting after rapid movement to the shooting position, in shooting under the regime of target appearance time and probably due to the gas mask worn during all activities. The gas mask makes it very difficult to fire effectively; it sometimes fogs up during physical exertion and shooting, which impedes the visibility of the target and aiming devices, has a limited field of vision and significantly impedes the acquisition of a comfortable and appropriate shooting stance (head position in relation to the weapon) [43].

The strongest significant effect of EF on SP was revealed in military pistol shooting in both the male and female groups. This shooting was performed on accuracy and focus in a standing stance. It was by far the easiest or one of the easiest shootings of the test, as the distance to the target was only 15 m and the target at which the fire was conducted was the same as in the carbine shooting at a distance of 100 m. Of course, small arms were fired, but the very short distance to the target made it more “forgiving” of possible shooter errors [30]. The lack of significant correlation between CTT-1 performance time and SP, except for carbine shooting in the group of males where the strength of correlation was weak anyway ( $r = -0.23$ ), clearly indicates that the second part of CTT, i.e., CTT-2, should be used to study complex performance functions with a potential relationship to the SP of soldiers [13].

Since males and females did not differ in their level of SP, and among males all shootings were positively associated with shorter CTT-2 performance, the lack of significant correlations in three out of four shootings with CTT-2 performance time in the female group can be explained by their small numbers. It can also be assumed that the remaining correlations would probably have been revealed if the number of women had been similar to the number of men. However, the study included all women who studied at MULF.

The literature recognizes that military performance depends on high levels of cognitive, EF, especially during heavy physical exercise [7]. Associations of some EF with SP

among soldiers were found, among others, by Hillman et al. They examined EEG activity during the preparation period between executed and rejected shots to better understand the attentional processes associated with the pre-shooting state. As in our study, they recognised the large role of attention in the complex process of firing a shot. Additionally, it was found that the decision to reject a shot appears to be characterised by a misallocation of neural resources associated with task performance. The study, unlike our own research, was conducted on a group of skilled sharpshooters who performed shooting at much greater distances than cadets, so comparisons should be made with caution [25].

Additionally, the results of our own research confirm the recent study by Shao et al. in which it was reported that self-control (as one of the elements of EF) during the performance of closed motor tasks in the environment determines that shooters have a higher anti-interference ability. This ability, in turn, boils down to the deliberate and active selection of specific data from the environment in shooting, i.e., focusing attention only on selected important information while ignoring other distracting stimuli from the environment [22]. Furthermore, the work of Sattlecker et al. focused on athletes at World and European Cup level in biathlon showed that postural balance and rifle stability play a key role in this sport [44]. Studies on such a skilled shooting group prove that shooters must be strongly engaged with the target while aiming and working on the weapon (attentional control) and constantly monitoring their behaviour and controlling their emotions to minimise the risk of making a mistake. On the other hand, when a mistake is made, they should make sure it has as little impact on the result as possible. For example, if a mistake occurs in the form of a “missed shot”, i.e., a trigger pull that is too fast, the shooter should quickly and accurately identify the problem, which is related to the control of emotions, and then make the appropriate correction and prevent a similar mistake from being repeated in the future (behavioural control). Comparing the above study with the results of our own research, it should be noted that the shooting was performed indoors at a distance of 50 m with a specialised air rifle fitted individually to each athlete without any additional physical load. Military shooting, on the other hand, usually involves an additional mental and physical load, so the greater use of EF in the form of behavioural control, emotions, focus of attention and the ability to eliminate external stimuli may be key determinants in SP.

Our results also show that the strength of the correlation between SP and the level of EF decreased with the more difficult level of shooting (shooting after a run-in and in a gas mask). This was probably related to a decrease in the level of EF under the influence of a greater external stressor such as a short and quick change in shooting stance, a gas mask and targets appearing in a limited time. The above results correspond with those of previous studies on EF at specific exercise intensities. Labelle et al. observed a significant decrease in EF at both 60% and 80% of peak power output [45]. Furthermore, Lo Bue-Estesa et al. confirmed that EF decreased during post-exercise assessment [43]. Additionally, the results of our own study are consistent with the results of a study among Reserved Officer Training Corps (ROTC) cadets, in which it was proven that high-intensity exercise decreases EF [7].

The results of our study confirm the hypothesis that gender is not a significant factor that affects the level of EF and the SP of soldiers. A higher level of EF has a significant effect on higher SP of male soldiers in all types of shooting from the present study with both carbines and pistols. However, among females, higher EF has a significant effect on higher pistol SP.

Summarizing the results of our own research and from the literature reports on gender differences in SP, it can be noted that there are still many unexplained issues. Noticeable discrepancies may be a result of differences in the way SP is measured in individual studies, the degree of shooting difficulty and natural somatic and motor differences. Therefore, it seems reasonable to systematically examine the SP of soldiers in a way that most accurately reflects the requirements of the population of interest, in particular, the study of soldiers' actions in real combat and training ground conditions. The level of shooting training of an individual soldier and his cognitive efficiency (especially of a commander-leader) is

of great importance for the combat potential of the armed forces and should be a key aspiration of each military unit. The results of the conducted research as well as many other works confirm the necessity of placing great emphasis on the cognitive preparation of a modern soldier, in particular aimed at the development of EF skills. They have a significant impact on the level of a soldier's shooting efficiency.

### 8. Limitations, Strengths and Future Research

The upper or lower values for some variables are due to hazard. This is the meaning of statistics when  $p < 0.05$ . Significant relationships and differences were shown on a sample with small numbers of women relative to men. We saw significant relationships between the variables studied, or lack thereof, but our evidence is weak because we showed them on a small number of subjects relative to men. Thus, we are not entirely sure that what we see is not a coincidence (which is always easier the smaller the sample), and that we will see a strong relationship again when we repeat this experiment; so more research is needed. In contrast, we saw strong evidence in statistically significant correlations in a large group of men.

Due to the simplicity of application in a military setting, the level of EF and processes related to attention were assessed using the Color Trails Test (CTT-1 and CTT-2). Despite the great popularity of neuropsychological tests for the assessment of EF in research (CTT, Wisconsin Card Sorting Test, Stroop Test) [46,47], it is now also possible to assess EF using neurophysiological methods such as visual or auditory evoked potentials and gamma oscillation. Increasingly, high-tech neuroimaging methods are also being used for neurocognitive measurements.

There is a lack of research in the literature on the relationship between EF and cadet SP, so this opens up useful new areas of research, particularly for researchers from the military community. Further research would be worthwhile to consider other factors influencing SP (e.g., type of shooting training, kinematic analysis of shooting stance and weapon stability, physical fitness) potentially important for the level of shooting training. Moreover, a lot of valuable information may be provided by the results of future research on dynamic SP under conditions of higher physical and mental fatigue in both male and female groups, as they will be more relevant to the realities of combat operations.

### 9. Conclusions

EF are crucial to unique human abilities including, as it turns out, soldiers' SP. A more thorough analysis of the components of EF may help to develop targeted interventions to improve them. This knowledge should be of particular interest to researchers in the uniformed services community and those professional groups where weapons are the primary tool.

The results of our own research indicate that female cadets represent a similar level of EF as well as SP; therefore, the load and evaluation methods in training should not be differentiated by gender. However, the problem of gender differences in specific professions such as uniformed services remains open and requires further detailed analysis.

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Article

# Sport Activity Load and Skeletomuscular Robustness in Elite Youth Athletes

Irina Kalabiska <sup>1</sup>, Annamaria Zsakai <sup>2,\*</sup>, Dorina Annar <sup>2</sup>, Robert M. Malina <sup>3</sup> and Tamas Szabo <sup>1,4</sup>

<sup>1</sup> Center for Sport Physiology, University of Physical Education, Research, Alkotás u. 44, 1123 Budapest, Hungary; kalabiskai@gmail.com (I.K.); szabo.tamas@mksz.hu (T.S.)

<sup>2</sup> Department of Biological Anthropology, Eotvos Lorand University, Pazmany p. s. 1/c., 1117 Budapest, Hungary; annar.dorina@gmail.com

<sup>3</sup> Department of Kinesiology and Health Education, University of Texas at Austin, Austin, TX 78712, USA; rmalina@lskyconnect.net

<sup>4</sup> Sport Sciences and Diagnostic Research Centre, Hungarian Handball Federation, Konyves K. Krt. 76, 1087 Budapest, Hungary

\* Correspondence: annamaria.zsakai@ttk.elte.hu

**Abstract:** In an earlier report, bone mineral reference values for young athletes were developed. This study addressed variations in bone mineral parameters of young athletes participating in sports with different mechanical loads. The bone mineral status of 1793 male and female athletes, 11 to 20 years of age, in several sports was measured with DEXA. Specific bone mineral parameters were converted to z-scores relative to age- and sex-specific reference values specified by the DEXA software. Z-score profiles and principal components analyses were used to identify body structural components in the young athletes and to evaluate the associations between the identified component and type of sport defined by mechanical load. A unique skeletomuscular robusticity of male wrestlers, pentathletes, and cyclists was noted: wrestlers had significantly more developed skeletomuscular robusticity and bone mineral density compared to the age-group average among elite athletes, while pentathletes and cyclists had lower bone mineral parameters than the age-group references among elite athletes. Among female athletes, bone mineral parameters of both the trunk and extremities of rhythmic gymnasts and pentathletes were significantly lower compared to the age-group means for elite athletes. The bone mineral development of elite young athletes varies with the impact forces associated with their respective sports. The skeletal development of cyclists, pentathletes, and rhythmic gymnasts should be monitored regularly as their bone development lags behind that of their athlete peers and the reference for the general population.

**Keywords:** bone mineral; skeletomuscular robusticity; elite athletes; DEXA

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## 1. Introduction

Athletes are not a homogenous population as both intrinsic (genetic and biological) and extrinsic (environmental, nutritional, and training) factors affect performance [1]. Both regular training for sport [2] and regular participation in physical activity [3] generally have a beneficial effect on mineral accrual and, correspondingly, bone mass and bone mineral density (BMD). The skeleton in childhood and adolescence is sensitive to the mechanical stimulation elicited by physical activity so that regular physical activity during childhood and adolescence can optimize skeletal health that persists through adulthood. Factors other than mechanical stimulation also influence bone development and include genotype and adequate levels of vitamin D and calcium [4–6]. Although BMD among athletes systematically training in different sports has received considerable attention [7–9], variations in total body BMD and BMD of body segments among athletes training in different sports merits attention.

Longitudinal training studies indicate that strength training and high-impact endurance training (anaerobic exercises) are associated with an increase in BMD [10]. On the other hand, negative effects of training (i.e., reduced BMD) in distance runners have also been reported [11,12]. Evidence for beneficial effects of non-weight-bearing activities (water sports) on bone mineral accrual and maintenance is somewhat controversial [13,14], and it has been questioned whether the effects are similar to those observed in athletes participating in weight-bearing sports [15,16]. There is also some indication that weight-bearing aerobic exercise may be more beneficial for bone health than non-weight-bearing activities [17]. Nevertheless, the most effective training protocol for attaining and maintaining a high bone mass and BMD has not been firmly established. Maintaining high bone mass and strength has an important role in the prevention of stress fractures and other skeletal injuries.

The growth status of children and adolescents, including youth athletes, is routinely evaluated relative to an established growth reference appropriate for a geographical region and, perhaps for the time of study, a given population variability and secular trends in growth [18,19]. Allowing for the selectivity of many sports (e.g., size and physique) and systematic effects of training on body composition and specific components of body composition, it has been suggested that reference values for athletes in specific sports may be more appropriate [20]. Although systematic training for sport influences the body composition of youth athletes, training does not influence linear growth nor growth in stature [21].

Reference values for several bone mineral parameters among youth athletes that are 10–20 years old have been developed [20] in the context of the need for reference data for national and athlete-specific samples. The initial results, though of interest, also suggested a potential need for bone mineral reference data for youth athletes training in specific sports. In this context, the purpose of the present study was to compare variations in bone mineral parameters of young athletes participating in sports with different mechanical loads.

The asymmetries of skeletomuscular development between the upper and lower body segments (e.g., rowers versus cyclers) or between the dominant and non-dominant arms (e.g., tennis players versus swimmers) associated with training load are reasonably well-established. Skeletomuscular development is usually significantly asymmetric in favor of the dominant side or region of the body, although an increased level of physical activity may help to prevent incorrect body posture, while asymmetric training loads on skeletal muscles may also enhance incorrect posture [22–25]. It is not clear, however, if this developmental asymmetry is also manifest in bone mineral parameters of body segments with different training loads. Thus, a secondary objective of this study was to compare bone mineral parameters of the upper and lower extremities and of the trunk among young athletes participating in different types of sports.

## 2. Materials and Methods

### 2.1. Study Design

This project was approved by the Research Ethics Committee of the University of Physical Education in Budapest, Hungary (ID of approval: TE-KEB/No42/2019). The Research Center for Sport Physiology (University of Physical Education, Hungary) has a cooperation agreement with numerous sports federations, associations, and clubs that focus on a variety of sports. The governing bodies of the respective sports organizations also approved the ethical codes established by Research Ethics Committee of the university. The research was carried out following the rules of the Declaration of Helsinki of 1975 (<https://www.wma.net/what-we-do/medical-ethics/declaration-of-helsinki/>, accessed on 21 December 2021) as revised in 2013. Parents of athletes <18 years old and the athletes were informed of the details of the project; both provided written informed consent. Details of the project were also provided for older athletes who also provided informed written consent.

## 2.2. Subjects

Subjects included a cross-sectional sample of 1734 athletes, 1299 males (11–20 years old) and 435 females (13–20 years old), who voluntarily agreed to participate in the study (Tables 1 and 2) [17]. The athletes represented several sports academies—primarily basketball, football, and handball, with smaller numbers for several individual sports such as pentathlon, rhythmic gymnastics, kayak, canoe, rowing, and wrestling. The athletes trained regularly, several times per day in many instances, and also regularly participated in competitions and tournaments on weekends. The athletes, as a group, had been training in their respective sports since 6–7 years of age and were considered elite.

**Table 1.** Distribution of young athletes by gender within age groups and sports.

Age (Years)	Males	Females	Sports	Males	Females
11	25	-	Wrestling	19	-
12	36	-	Rowing	16	-
13	89	32	Football	531	-
14	138	97	Kayak/canoe	19	-
15	240	103	Bicycling	33	-
16	308	57	Handball	225	251
17	226	71	Basketball	428	139
18	154	31	Pentathlon	28	22
19	50	25	Rhythmic gymnastics	-	23
20	33	19			
Total	1299	435		1299	435

**Table 2.** Medians for body size, fat and muscle mass, BMC, and BMD in the young male and female athletes by age group.

Age (Years)	Height (cm)	Weight (kg)	Fat Mass (kg)	Muscle Mass (kg)	BMC (g)	BMD (g/cm <sup>2</sup> )
Males						
11	155.8	44.5	3.3	31.6	1689.0	0.962
12	163.2	43.7	3.4	33.0	1878.0	1.007
13	169.8	50.9	3.3	38.1	2123.0	1.055
14	176.4	61.2	3.7	48.4	2573.0	1.159
15	179.3	67.9	4.1	53.4	2901.0	1.246
16	179.8	70.0	4.1	55.8	3082.5	1.298
17	182.6	73.5	4.4	59.3	3230.0	1.349
18	183.2	75.0	4.4	60.3	3272.0	1.367
19	180.6	73.8	4.2	60.5	3377.5	1.378
20	178.0	74.2	4.3	60.3	3455.5	1.443
Females						
13	168.5	54.9	5.2	39.0	2193.5	1.111
14	172.5	63.4	6.7	44.1	2573.0	1.241
15	171.4	62.5	6.6	44.4	2584.0	1.226
16	172.3	64.8	6.6	46.3	2699.0	1.284
17	172.6	67.7	7.0	47.1	2753.0	1.295
18	172.7	65.9	7.2	47.6	2840.0	1.307
19	174.4	68.2	6.1	48.6	2949.5	1.356
20	174.1	71.6	6.4	51.7	2983.0	1.376

## 2.3. Data Collection

The cross-sectional research was conducted between September 2015 and February 2020. Whole-body bone mineral density (BMD), bone mineral content (BMC), and body mass components (fat mass, muscle mass) were measured with a GE Lunar Prodigy dual-energy X-ray scanner (GE Medical Systems, Madison, WI, USA).

Subjects were asked to avoid eating and drinking for at least 60 min prior to examination and to follow their habitual training regime during the week of the examination. All examinations took place between 9:00 and 12:00 in the morning.

Several athletes were excluded from the study if they:

1. Had severe degenerative lesions or fractures/deformations in the measurement area;
2. Were unable to reach the correct position and/or remain immobile during measurement;
3. Had a very high or low body mass index (i.e., a BMI that could adversely affect the accuracy of measurement process);
4. Were exposed to an enhanced X-ray/CT scan several days prior to the study;
5. Were pregnant.

#### 2.4. Statistical Analyses

The body mass components and bone mineral densities were expressed relative to height (m) to reduce the influence of body size on the specific parameters. The structural and bone mineral parameters of each athlete were converted to z-scores relative to age- and sex-specific mean and standard deviation values estimated for the total sample ( $z = (\text{individual value} - \text{age-group mean}) / \text{age-group standard deviation}$ ). Bone mineral densities of the upper extremities, lower extremities, and trunk were also expressed as a percentage of total BMD to address asymmetries in BMD by type of sport [26].

Wilcoxon signed-rank test was used to compare the z-scores of components of body mass and of bone parameters in the athletes by sport (an alpha of 0.05 was used as the cut-off for significance in all analyses). Linear regression analysis was used to evaluate the relationship between the L1–L4 BMD z-scores estimated by the GE Prodigy Lunar reference and the youth athlete reference series.

Principal components analysis (PCA) was used to reduce the body parameters to a smaller set of components that accounted for most of the variance in the variables considered. Sex-specific PCAs were initially conducted; as the analyses showed similar components and loadings, the analysis was also conducted for the total sample. The original variables were log-transformed to approximate the normality assumption of PCA. Based on eigenvalues  $>1.0$ , loadings  $>0.90$  were used to identify the variables characteristic of the respective components. Cronbach's alpha was used to estimate the reliability of the analysis. Principal component scores were also calculated for each athlete.

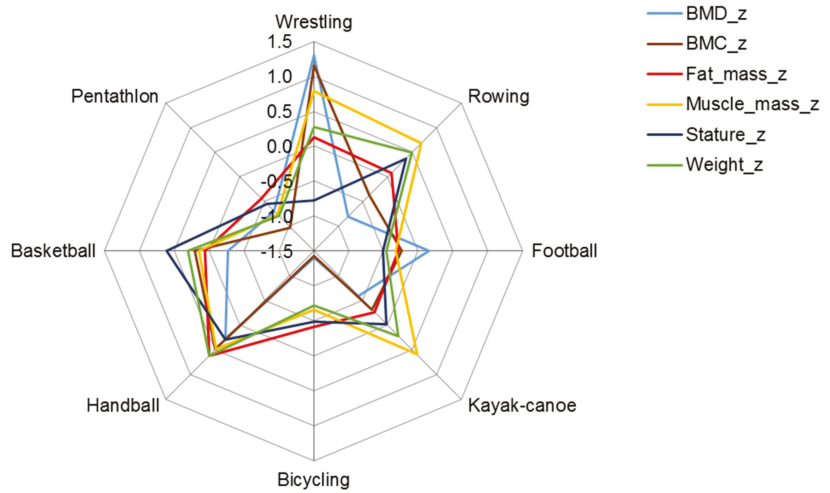
### 3. Results

The z-score profiles of body mass components and bone parameters indicated greater skeletal and muscular robustness of wrestlers compared with male participants in other sports, especially pentathletes, whose skeletomuscular robustness was the least developed among the sports represented in the sample. The skeletomuscular robustness of cyclists was also less developed compared to the age-group reference, but their muscular development was similar to the age-group mean value for males (Figure 1, Table 3). Muscle mass was also highly developed among rowers, kayaker–canoeists, and handball players.

The skeletomuscular development of female basketball and handball players was greater than the age-group average, although the bone mineral density of basketball players was slightly less compared to handball players (Figure 2, Table 3). The bodily structure of rhythmic gymnasts and pentathletes, on the other hand, differed significantly from athletes in the two team sports; their average skeletomuscular development and fatness were also below the age-group means (Figure 2).

Results of the principal components analysis are summarized in Table 4. Two components were indicated: PC 1 described skeletomuscular robustness identified by body mass, total body BMC, and muscle mass, while PC 2 described body composition in the context of BMD (positive) and body fatness (negative). PC 1 accounted for more than 60% of the total variance, while PC 2 explained an additional 20%. Overall, the results suggested that

skeletomuscular robustness is the main source of variance in the overall body structure and bone development of youth athletes.



**Figure 1.** Z profile (mean of z-scores) of structural parameters in male athletes; BMC in grams and fat and muscle mass in kilograms were expressed as a percentage of height in meters.

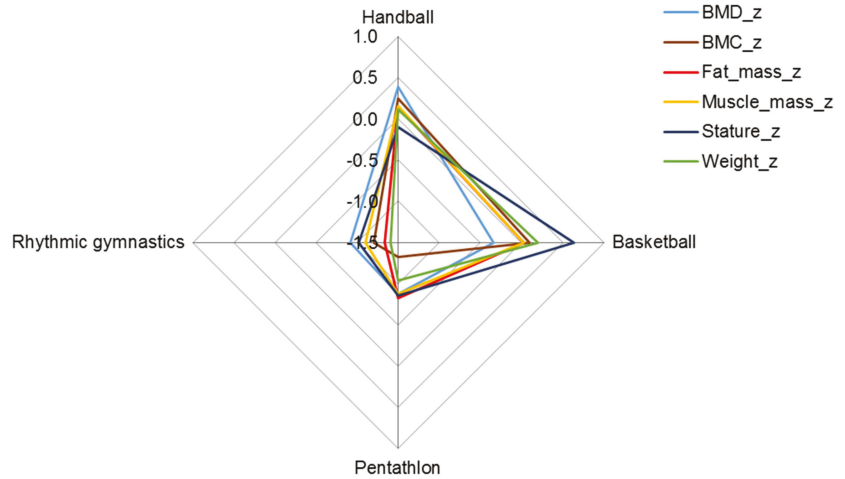
**Table 3.** Significance level of the Wilcoxon signed-rank tests (significant values in italics) of z-scores for components of body mass and for bone parameters in young athletes by sport.

Males	Wr	Ro	Fo	KC	Bi	Hb	Bb	Pe
Weight	0.629	0.003	<i>p &lt; 0.001</i>	0.184	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Height	0.001	0.007	<i>p &lt; 0.001</i>	0.629	0.002	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Muscle mass	0.006	0.002	<i>p &lt; 0.001</i>	0.001	0.003	<i>p &lt; 0.001</i>	0.009	<i>p &lt; 0.001</i>
Fat mass	0.444	0.836	<i>p &lt; 0.001</i>	0.049	0.001	<i>p &lt; 0.001</i>	0.116	<i>p &lt; 0.001</i>
BMC	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMD total	<i>p &lt; 0.001</i>	0.006	<i>p &lt; 0.001</i>	0.003	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMD UE	<i>p &lt; 0.001</i>	0.379	<i>p &lt; 0.001</i>	0.004	0.001	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMD Tr	0.004	0.002	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	0.049	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMD Le	<i>p &lt; 0.001</i>	0.003	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Females	Hb	Bb	Pe	RG				
Weight	0.245	0.008	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
Height	0.044	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
Muscle mass	0.075	0.908	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
Fat mass	0.069	0.735	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
BMC	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
BMD total	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
BMD UE	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	0.017				
BMD Tr	<i>p &lt; 0.001</i>	0.315	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				
BMD Le	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>				0.002

Wr, wrestling; Ro, rowing; Fo, football; KC, kayak-canoe; Bi, bicycling; Hb, handball; Bb, basketball; Pe, pentathlon; RG, rhythmic gymnastics.

The distribution of PC scores for each component (PC 1 x-axis, PC 2 y-axis) by sport is illustrated in Figure 3 for both males and females. The skeletomuscular robustness of wrestlers (upper right of the plot) stood out relative to male athletes in other sports. The position of the male cyclists and pentathletes (lower left) highlighted their lower level of overall skeletomuscular development and lower BMD compared to athletes in the other sports. Nevertheless, the overlap of the distribution of PC scores should be noted as it highlights the variability among individual athletes in the respective sports.

Among female athletes, the position of rhythmic gymnasts and pentathletes relative to handball and basketball players stood out (Figure 3). In contrast to the overlap among male athletes in different sports, the overlap of rhythmic gymnasts and pentathletes compared to handball and basketball players was minimal.



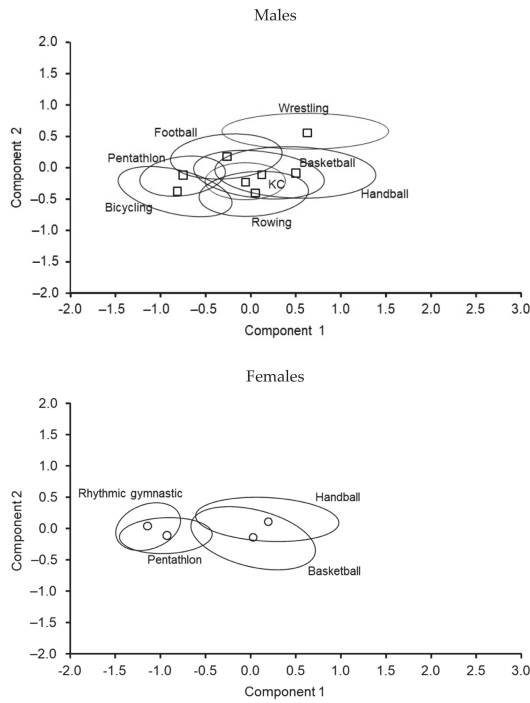
**Figure 2.** Z profile (mean of z-scores) of structural parameters in female athletes; BMC in grams and fat and muscle mass in kilograms were expressed as a percentage of height in meters.

**Table 4.** Results of the principal components analysis (Cronbach’s alpha: 0.946, bold and italic represent the absolute amount of the component loadings).

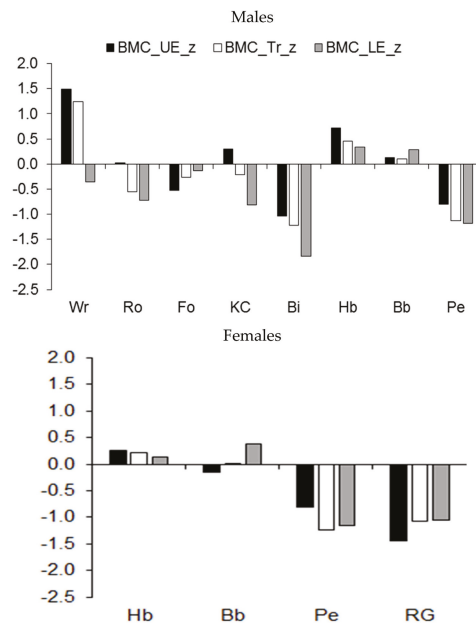
	Component 1	Component 2
Eigenvalue	3.172	1.074
% of variance	63.43	19.87
BMD_z	0.742	<b>1.693</b>
BMC_z	<b>1.101</b>	0.708
Fat mass_z	0.865	<b>−0.980</b>
Muscle mass_z	<b>1.090</b>	−0.272
Weight_z	<b>1.141</b>	−0.797

Median z-scores for BMC and for muscle mass of the upper and lower extremities and of the trunk are illustrated in Figure 4, while results of the comparisons of athletes by sport are summarized in Table 5. Among male athletes, median z-scores for BMC in the upper extremities and the trunk of wrestlers and handball players were above the age-group average, while the BMC of the lower extremities was above the age-group average in both handball and basketball players (Figure 4, upper part). In contrast, the BMC of the lower extremities and trunk of rowers and kayaker–canoeists and the BMC of the upper extremities in football players lagged behind the age-group average. The BMC of male cyclists and pentathletes was smaller in each region of the body compared to the age-group average. Among female athletes, rhythmic gymnasts and pentathletes had a lower BMC than similar-aged peers in each region of the body (Figure 4, lower part).

The regional development of muscle mass showed a similar pattern (Figure 5). Major differences were apparent in the increased muscular robustness of the upper extremities and the trunk relative to the age-group reference among male wrestlers, rowers, and kayakers/canoists, and to a lesser extent, among handball players (Figure 5, upper part). On the other hand, the development of muscle mass in the three regions tended to be lower among pentathletes and cyclists and, to a lesser extent, among football players. Among female athletes, rhythmic gymnasts had reduced development of muscle mass in the three regions relative to the age-group reference, while pentathletes had reduced development of muscle mass in the lower extremities compared to the upper extremities and trunk (Figure 5, lower part).



**Figure 3.** Distribution of PC scores by component (PC 1 on the x-axis and PC 2 on the y-axis) among youth athletes by sport (□ and ○: sport type mean and 95% confidence ellipse around the mean).



**Figure 4.** Median z-scores of BMC (expressed as a percentage of stature) of the upper extremities (UE), trunk (Tr), and lower extremities (LE) in youth athletes by sport.

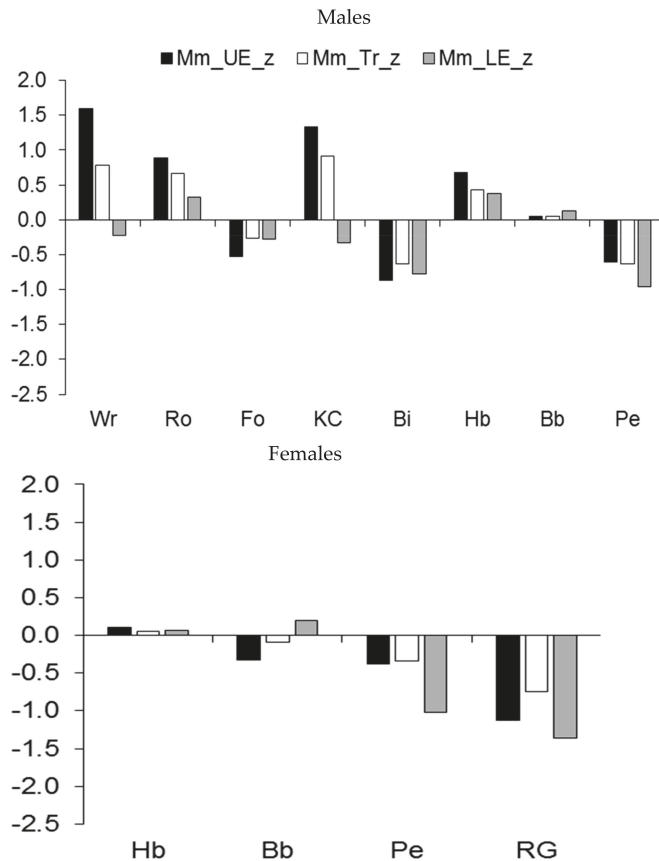


**Table 5.** Significance level in Wilcoxon signed-rank test (significant values in italics) of body mass components in the studied body regions (z-values) in young athletes by the type of sport.

Males	Wr	Ro	Fo	KC	Bi	Hb	Bb	Pe
BMC-UE	<i>p &lt; 0.001</i>	0.569	<i>p &lt; 0.001</i>	0.107	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	0.070	<i>p &lt; 0.001</i>
BMC-Tr	<i>p &lt; 0.001</i>	0.045	<i>p &lt; 0.001</i>	0.107	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	0.002	<i>p &lt; 0.001</i>
BMC-LE	0.398	0.034	0.002	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Muscle mass-UE	<i>p &lt; 0.001</i>	0.001	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	0.001	<i>p &lt; 0.001</i>	0.034	0.002
Muscle mass-Tr	0.022	0.002	<i>p &lt; 0.001</i>	0.001	0.003	<i>p &lt; 0.001</i>	0.042	0.001
Muscle mass-LE	0.872	0.023	<i>p &lt; 0.001</i>	0.070	0.003	<i>p &lt; 0.001</i>	0.001	<i>p &lt; 0.001</i>
Fat mass-UE	0.354	0.234	<i>p &lt; 0.001</i>	0.295	0.002	<i>p &lt; 0.001</i>	0.112	0.001
Fat mass-Tr	0.351	0.877	<i>p &lt; 0.001</i>	0.004	0.002	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Fat mass-LE	0.184	0.569	<i>p &lt; 0.001</i>	0.334	0.001	<i>p &lt; 0.001</i>	0.345	0.010

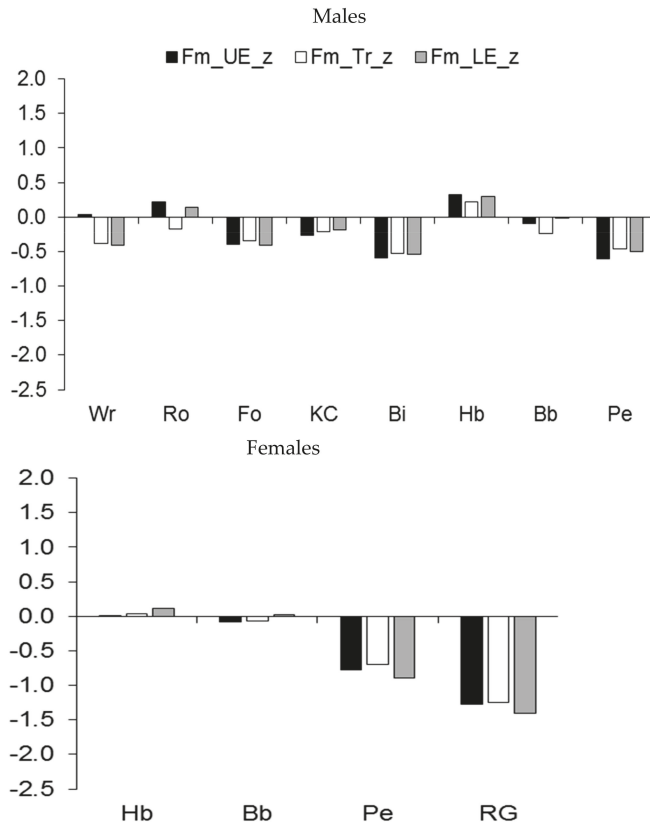
  

Females	Hb	Bb	Pe	RG
BMC-UE	<i>p &lt; 0.001</i>	0.027	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMC-Tr	<i>p &lt; 0.001</i>	0.683	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
BMC-LE	0.049	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Muscle mass-UE	0.008	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Muscle mass-Tr	0.115	0.740	0.008	0.001
Muscle mass-LE	0.204	0.027	0.026	<i>p &lt; 0.001</i>
Fat mass-UE	0.073	0.240	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Fat mass-Tr	0.210	0.445	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>
Fat mass-LE	0.208	0.449	<i>p &lt; 0.001</i>	<i>p &lt; 0.001</i>



**Figure 5.** Median z-scores for muscle mass (expressed as a percentage of stature) of the upper extremities (UE), trunk (Tr), and lower extremities (LE) in youth athletes by sport.

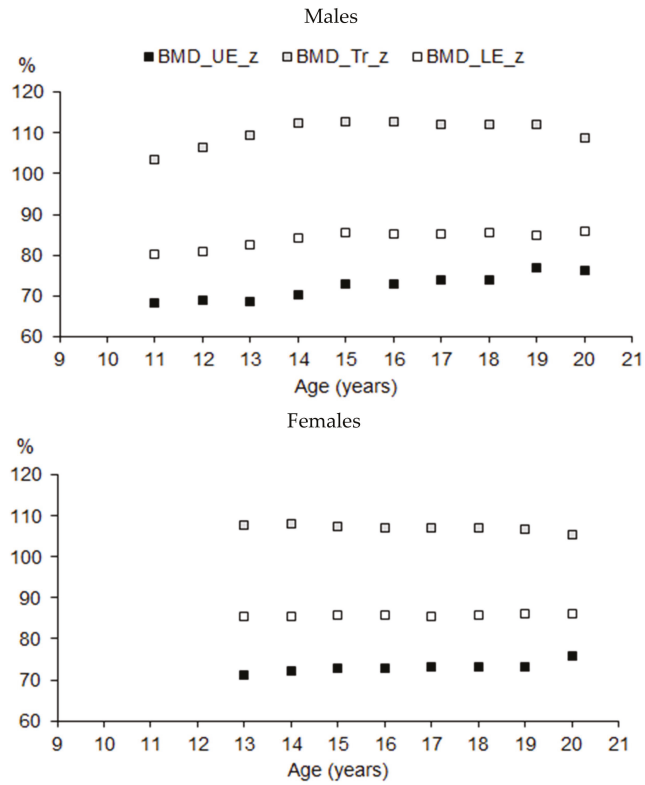
In contrast to BMC and muscle development, the fat mass of the youth athletes showed a similar pattern among the male athletes in different sports (Figure 6, upper part). Cyclists and pentathletes, however, had somewhat less fatness in the different regions compared to similar-aged peers in other sports. Among females, pentathletes and rhythmic gymnasts had considerably less fatness in the three regions than similar-aged peers in handball and basketball (Figure 6, lower part).



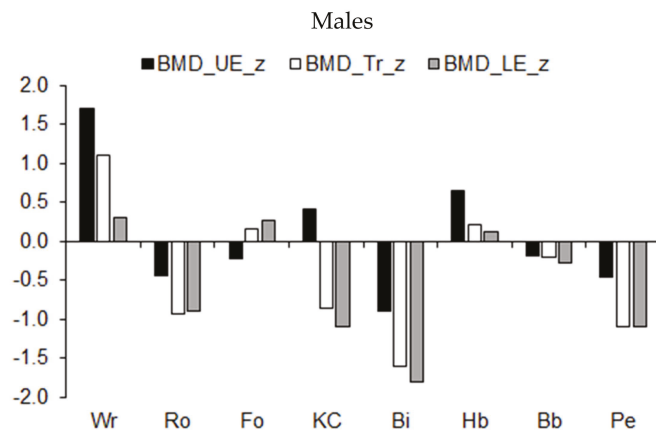
**Figure 6.** Median z-scores for fat mass (expressed as a percentage of stature) of the upper extremities (UE), trunk (Tr), and lower extremities (LE) in youth athletes by sport.

The pattern of BMD in the extremities and the trunk expressed as a percentage of total BMD (BMD%) in the total samples of male and female athletes suggested that the mechanical load on the human body varied by the position of the regions relative to posture (bipedal standing or bipedal movement). The higher the position of the region in the human body, the lower the BMD% (Figure 7).

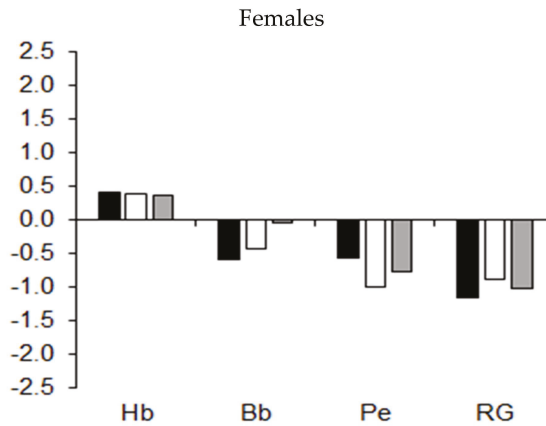
Mean z-scores for the BMD (expressed as a percentage of stature) of the extremities and trunk among athletes in the different sports are illustrated in Figure 8. Overall, the trend indicated asymmetric BMD development by sport. The BMD of the upper extremities and trunk of wrestlers and of the upper extremities of handball players was higher, while the BMD in the lower extremities and trunk of rowers, kayaker–canoeists, and pentathletes was lower relative to the age-group average for male athletes (Figure 8, upper part). Compared to the age-group average, BMD in the three regions was reduced among male cyclists. BMD in the three bodily regions was also reduced among female pentathletes and rhythmic gymnasts (Figure 8, lower part).



**Figure 7.** BMD of upper extremities (UE), trunk (Tr), and lower extremities (LE) expressed as a percentage of total BMD by age in youth male and female athletes (Mann–Whitney test: UE— $p < 0.05$  in 13, 14, and 19 years of age; Tr— $p < 0.05$  in 13, 14, and 16 years of age; Le— $p < 0.05$  in the entire interval).

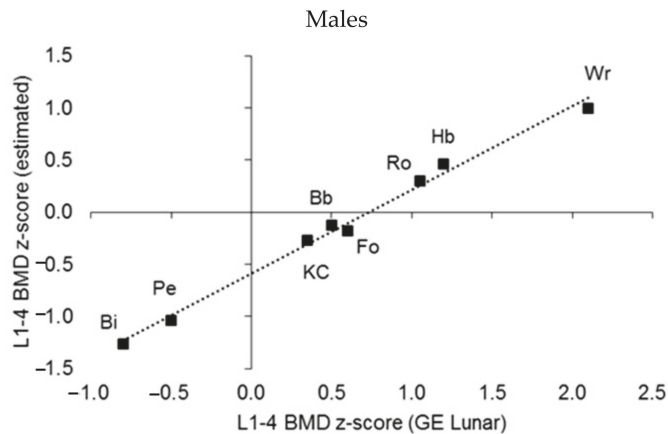


**Figure 8.** Cont.

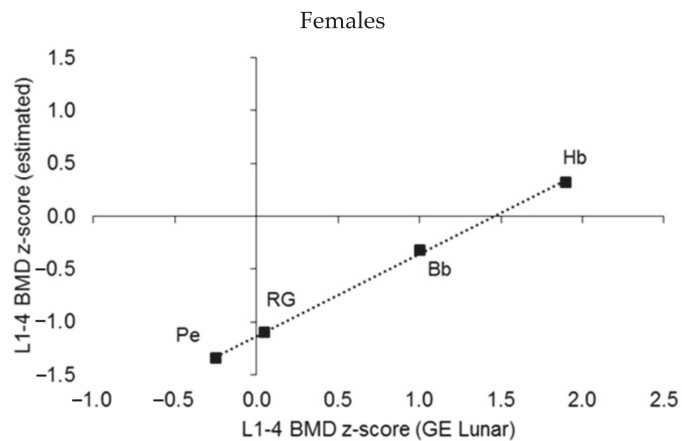


**Figure 8.** Median z-scores for BMD (expressed as a percentage of stature) of the upper extremities (UE), trunk (Tr), and lower extremities (LE) in youth athletes by sport.

A DEXA scan usually includes the BMD of the lumbar spine and proximal femur region; both regions are commonly used to estimate the risk of osteopenia, osteoporosis, and fractures. Z-scores for the BMD of the lumbar spine (Figure 9) were estimated with both the GE Lunar Prodigy DEXA scanner software and the respective BMD reference for the total sample of male and female athletes [20]. The correlation between the z-scores in both sexes was  $>0.99$  ( $p < 0.001$ ), but z-scores based on the athlete reference underestimated those based on the GE Lunar unit by 0.59 z-score units in males and 1.1 z-score units in females. Since the BMD of athletes is usually greater than the 90th percentile of the non-athlete/general population reference [20], the enhanced BMD development in athletes would seem to justify the use of a reference based on athletes, specifically in the context of follow-up examinations after training programs. Nevertheless, the use of a non-athlete reference with athletes is also justified as potential underdevelopment of BMD can be diagnosed with this screening reference.



**Figure 9.** Cont.



**Figure 9.** Median L1–L4 BMD z-scores estimated by sport by using the GE Prodigy Lunar reference and the youth athlete reference (Kalabiska et al. 2020); linear regressions: males  $p < 0.001$ ,  $R^2 = 0.991$ , intercept =  $-0.59$ , females  $p < 0.001$ ,  $R^2 = 0.999$ , intercept =  $-1.14$ .

The median z-score of the BMD of lumbar vertebrae L1 to L4 estimated by the GE Lunar Prodigy DEXA software approached the low BMD category (osteopenia, cut-off value, BMD z-score  $< -1.0$ ) in male cyclists (BMD z-score =  $-0.82$ , see Figure 9). Median z-scores of L1–L4 BMD in athletes in other sports showed a similar pattern to that noted for the total BMD of the youth athletes (Figure 9): pentathletes of both sexes and female rhythmic gymnasts had very low BMD in the lumbar spine region, while male wrestlers and basketball and handball players of both sexes had the highest median BMD z-scores in the sample of athletes considered in the present study (Figure 9).

#### 4. Discussion

Higher estimates of BMD and a larger bone mass have been described in athletes in several sports compared to non-athletes and the general population [20,27,28]. The results are generally interpreted in the context of positive benefits of systematic physical activity on bone. However, several studies have shown that not all sports have the same bone-related benefits, especially among those where training starts in childhood and adolescence [29–32].

Overall, the observations of male athletes highlighted the increased skeletal and muscular robustness of wrestlers and increased muscular development of rowers, kayaker–canoeists, and handball players, but they also highlighted the reduced skeletal and muscular robustness of pentathletes and reduced skeletal development of cyclists in male athletes. The observations were generally consistent with those in studies of male athletes in sports with prevailing speed and strength loadings, e.g., wrestling and rowing [33,34]. However, one study was focused on the proximal femur [33], while the other focused on the mesomorphic component of somatotype estimated from anthropometry [34]. Results for female athletes in the present analysis highlighted the increased skeletomuscular development of basketball and handball players and reduced skeletomuscular development among rhythmic gymnasts and pentathletes. The observations were consistent with those of other studies evaluating body structure among young female athletes. Results from a study of rhythmic gymnasts noted that poor energy balance was associated with a lower lean body mass and reduced skeletomuscular development [35], while other studies noted higher estimates of BMD in a number of skeletal sites among female handball and basketball players [36–38].

Observations in the three bodily regions considered in the study indicated reduced BMD among male cyclists, female pentathletes, and rhythmic gymnasts, while the BMD of

the lower extremities and trunk was reduced among male rowers, kayaker–canoeists, and pentathletes. On the other hand, BMD was increased in the upper extremities and trunk of wrestlers and in the upper extremities of male handball players. The differences in BMD and variations among bodily regions reflect the frequency patterns of use of the bodily regions in the activities associated with training in the respective sports. The lower the position of the region in the body, the greater the weight loading on the region and, in turn, the higher the BMD in both sexes in the specific sports was observed. Morphofunctional asymmetry has been noted in several studies of youth athletes. For example, skeletomuscular asymmetry was noted between the right and left sides of the body among field hockey players [39] and between the dominant and non-dominant arms of tennis players [40]. Nevertheless, asymmetry in skeletomuscular development among bodily regions in youth athletes participating in different types of sports has not been systematically addressed.

Regular physical activity is an important factor in bone health. Physical activity interventions aimed at augmenting bone mineral were consistent with the preceding observations based on comparisons of active and less active youth. Physical activity interventions, e.g., two or three times per week of moderate-to-high-intensity activities, weight-bearing activities of a longer duration (45–60 min), and/or high-impact activities over a shorter duration (10 min), are associated with enhanced BMC in children and adolescents [41–43]. Consensus regarding the type and amount of physical activity required for enhanced development of BMD needs more attention [44–46]. Both systematic weight training, which forces the body to work against gravity, and isometric training are associated with increased BMD in athletes due to the greater forces acting on muscle and bone tissues [47].

It is important to study bone mineral characteristics of youth athletes in general and of elite athletes in sports characterized by variations in impact forces. The skeletal development of cyclists, pentathletes, and rhythmic gymnasts should be monitored more frequently since bone development among athletes in these sports tends to lag relative to elite athletes of the same chronological age and relative to population-based reference values. There is also a need to systematically consider the cortical and trabecular architecture of bone among youth athletes in different sports and how it relates to variations in training load.

## 5. Limitations

Given the limited number of female athletes, the analysis of sex differences in total body and regional BMD should be interpreted with caution. The lack of an indicator of maturity status in the younger athletes was also a limitation.

## 6. Conclusions

Results of this study confirmed that youth athletes in sports without systematic weight-bearing tend to have reduced BMD compared to peers of the same chronological age in sports with systematic weight-bearing. Decreased skeletomuscular development and lower bone mineral parameters were noted among male pentathletes and cyclists and among female rhythmic gymnasts and pentathletes. In contrast, increased skeletomuscular development and enhanced bone mineral parameters were noted among male wrestlers, rowers, kayaker–canoeists, and handball players and among female basketball and handball players.

Different loading patterns for specific bodily regions associated with the different sports were reflected in the skeletomuscular development of the respective bodily regions in young athletes. Asymmetry of skeletomuscular development was evident not only from the gradient of weight-bearing but also from the pattern of localization of activities associated with training in specific sports, i.e., the differences in the skeletomuscular development of the upper and lower extremities and the trunk reflected the different types of activities associated with the specific sports considered in the study. Of note, cyclists, pentathletes, and rhythmic gymnasts were at a higher risk for the development of inferior bone structure.

Age- and sex-dependent critical cut-off values for the structural parameters of bone density, architecture, and mass should be specified by the type of sport to facilitate the development of training protocols for elite youth athletes and to reduce the risk of bone stress-related injuries. In addition to bone mineral parameters, indicators of bone geometry indicators should be considered in the screening of stress-related bone injuries since bone geometry is also a determinant of the mechanical resistance of bones [48].

Reference values for indicators of body composition and bone mineral are potentially useful in examinations of athletes in specific sports. The observed differences among athletes in different sport types suggest a need for sport-specific reference values, but such references are not currently available.

**Author Contributions:** I.K. participated in data collection, bone development estimation, the statistical analysis and interpretation of the results, and manuscript construction. A.Z. participated in the statistical analysis and interpretation of the results and manuscript construction. D.A. aided in the preparation of the manuscript. R.M.M. participated in the interpretation of the results and preparation of the manuscript. T.S. participated in the statistical analysis and interpretation of the results and manuscript construction. All authors have read and agreed to the published version of the manuscript.

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**Institutional Review Board Statement:** The study was conducted in accordance with the Declaration of Helsinki, and approved by the Institutional Ethics Committee of University of Physical Education in Budapest, Hungary (ID of approval: TE-KEB/No42/2019, 3 January 2019).

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

**Data Availability Statement:** Tables with descriptive statistics (means, standard deviations) for age, height, weight, body composition, and bone mineral parameters by age groups for football, handball, and basketball in males and handball and basketball in females and for the total samples for wrestling, rowing, kayak/canoe, bicycling, and pentathlon in males and for pentathlon and rhythmic gymnastics in females are available upon request.

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Article

# Oxygen Consumption ( $VO_2$ ) and Surface Electromyography (sEMG) during Moderate-Strength Training Exercises

Muhammad Adeel <sup>1,2</sup>, Hung-Chou Chen <sup>3,4</sup>, Bor-Shing Lin <sup>5</sup>, Chien-Hung Lai <sup>3,6</sup>, Chun-Wei Wu <sup>2</sup>, Jiunn-Horng Kang <sup>3,6</sup>, Jian-Chiun Liou <sup>2</sup> and Chih-Wei Peng <sup>1,2,7,\*</sup>

- <sup>1</sup> International Ph.D. Program in Biomedical Engineering, College of Biomedical Engineering, Taipei Medical University, Taipei 110, Taiwan; d845108004@tmu.edu.tw
  - <sup>2</sup> School of Biomedical Engineering, College of Biomedical Engineering, Taipei Medical University, Taipei 110, Taiwan; george.jasonbiolab@gmail.com (C.-W.W.); jcliou@tmu.edu.tw (J.-C.L.)
  - <sup>3</sup> Department of Physical Medicine and Rehabilitation, School of Medicine, College of Medicine, Taipei Medical University, Taipei 110, Taiwan; 10462@s.tmu.edu.tw (H.-C.C.); chlai@tmu.edu.tw (C.-H.L.); jhk@tmu.edu.tw (J.-H.K.)
  - <sup>4</sup> Department of Physical Medicine and Rehabilitation, Shuang Ho Hospital, Taipei Medical University, New Taipei City 235, Taiwan
  - <sup>5</sup> Department of Computer Science and Information Engineering, National Taipei University, New Taipei City 237, Taiwan; bsline@mail.ntpu.edu.tw
  - <sup>6</sup> Department of Physical Medicine and Rehabilitation, Taipei Medical University Hospital, Taipei 110, Taiwan
  - <sup>7</sup> School of Gerontology Health Management, College of Nursing, Taipei Medical University, Taipei 110, Taiwan
- \* Correspondence: cwpeng@tmu.edu.tw

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**Abstract:** Oxygen consumption ( $VO_2$ ) during strength training can be predicted through surface electromyography (sEMG) of local muscles. This research aimed to determine relations between  $VO_2$  and sEMG of upper and lower body muscles to predict  $VO_2$  from sEMG during moderate-intensity strength training exercises. Of the 12 participants recruited, 11 were divided into two groups: untrained ( $n = 5$ ; with no training experience) and trained ( $n = 6$ ; with 2 months of training experience). On different days, each individual completed six training sessions. Each participant performed training sessions consisting of three types of dumbbell exercises: shoulder press, deadlift, and squat, while wearing a mask for indirect calorimetric measurements of  $VO_2$  using the Cortex Metalyzer 3B. sEMG measurements of the bilateral middle deltoid, lumbar erector spinae, quadriceps (rectus femoris), and hamstring (biceps femoris) muscles were recorded. The  $VO_2$  was predicted from sEMG root mean square (RMS) values of the investigated muscles during the exercise period using generalized estimating equation (GEE) modeling. The predicted models for the three types of exercises for the untrained vs. trained groups were shoulder press [QIC = 102, \*  $p = 0.000$  vs. QIC = 82, \*  $p = 0.000$ ], deadlift [QIC = 172, \*  $p = 0.000$  vs. QIC = 320, \*  $p = 0.026$ ], and squat [QIC = 76, \*  $p = 0.000$  vs. QIC = 348, \*  $p = 0.001$ ], respectively. It was observed that untrained vs. trained groups predicted GEE models [quasi-likelihood under an independence model criterion (QIC) = 368,  $p = 0.330$  vs. QIC = 837,  $p = 0.058$ ], respectively. The study obtained significant  $VO_2$  prediction models during shoulder press, deadlift, and squat exercises using the right and left middle deltoid, right and left lumbar erector spinae, left rectus femoris, and right and left biceps femoris sEMG RMS for the untrained and trained groups during moderate-intensity strength training exercises.

**Keywords:** GEE modeling; oxygen consumption; strength training; surface electromyography

## 1. Introduction

According to the American College of Sports Medicine (ACSM) [1] and the American Heart Association (AHA), strength training is beneficial to one's health [2]. It has several advantages, including increased strength and beneficial changes in body composition [1]. According to the ACSM, gaining health and fitness benefits from resistance training requires

at least one set of eight to twelve repetitions of each of eight to ten exercises involving the major muscle groups [3] on two or more days per week [4].

Previous research has explored the acute metabolic demands during strength exercise. Variables such as the muscle mass [5], exercise speed [6–8], number of sets [9,10], number of repetitions [11,12], workload [13,14], training volume [15], and rest intervals [11,16,17] resulted in substantially greater increases in oxygen consumption ( $VO_2$ ) and energy expenditure (EE). Currently, oxygen consumption is commonly measured through indirect calorimetry, which has a stated accuracy of  $-2\%$  to  $4\%$  [16]. However, acute physiological responses to skeletal muscle activation during moderate-strength exercises have not been thoroughly investigated.

Surface electromyography (EMG; sEMG), one of the most common methods of measuring muscle activation, is an electrophysiological recording technique for detecting the electric potential across muscle fiber membranes [17]. One recent study reported a relation between  $VO_2$  and muscle activity for squats and heel raises with 80% of one repetition maximum (1RM) in healthy male participants and observed an increase in oxygen uptake after 6 weeks of resistance exercises [18]. Another study investigated the mean correlation between the surface EMG amplitude and oxygen uptake for lower extremity muscles as 0.69–0.87 during treadmill walking in young males [19].

To the best of our knowledge, no single study has found that oxygen consumption ( $VO_2$ ) is related to the sEMG of the various upper limb and lower limb muscles during strength exercises at 60% of 1RM. It is an unexplored field of research to find which muscle sEMG has a significant association in computing oxygen consumption in healthy populations. By determining this kind of relationship, we can better explain which muscle activation can more or less predict  $VO_2$ . Some muscles are highly active during the shoulder press, deadlift, and squat, but we also observed other muscles which are not highly active during these strength workouts.

The purpose of this study was to model oxygen consumption with the sEMG of the bilateral middle deltoid, lumbar erector spinae, quadriceps, and hamstring muscles during three dumbbell exercises (shoulder press, deadlift, and squat). The main objective of this research was to calculate the multilinear relationship between  $VO_2$  as a dependent variable with sEMG measurements of the bilateral middle deltoid, lumbar erector spinae, quadriceps (rectus femoris), and hamstring (biceps femoris) muscles as independent outcome measures using generalized estimating equations (GEEs).

## 2. Materials and Methods

### 2.1. Subject Recruitment

A clinical controlled trial was undertaken at Taipei Medical University Hospital, and the protocol was accepted by the TMU-Joint Institutional Review Board (IRB no.: N202004023). [ClinicalTrials.gov](https://clinicaltrials.gov) was used to register the study (NCT04532905). Between December 2020 and May 2021, convenience sampling was used to enroll 12 young male and female volunteers into two groups. One of them was left out, and the untrained group (with no strength training experience) contained five participants and the trained group (with 2 months of strength training experience) six participants. Prior to the start of the trial, each participant signed a written consent. The research's goals and associated risks were explained to the participants.

The following criteria were used to include participants for the study: (1) a healthy male or female between the ages of 20 and 40; (2) without recent metabolic, systematic, or musculoskeletal disease or injury from the previous six months; (3) no recent surgical procedure that might impair workouts; (4) no medication use, particularly for sleep, depression, blood pressure control, etc.; and (5) physically fit according to the Physical Activity Readiness Questionnaire (PAR-Q) [20] (Figure 1). Experts in the field of research who were not engaged in the intervention performed the randomization, functional outcome

measures, and data analysis. In this study, the order of the exercises was concealed from the participants. (Table 1).

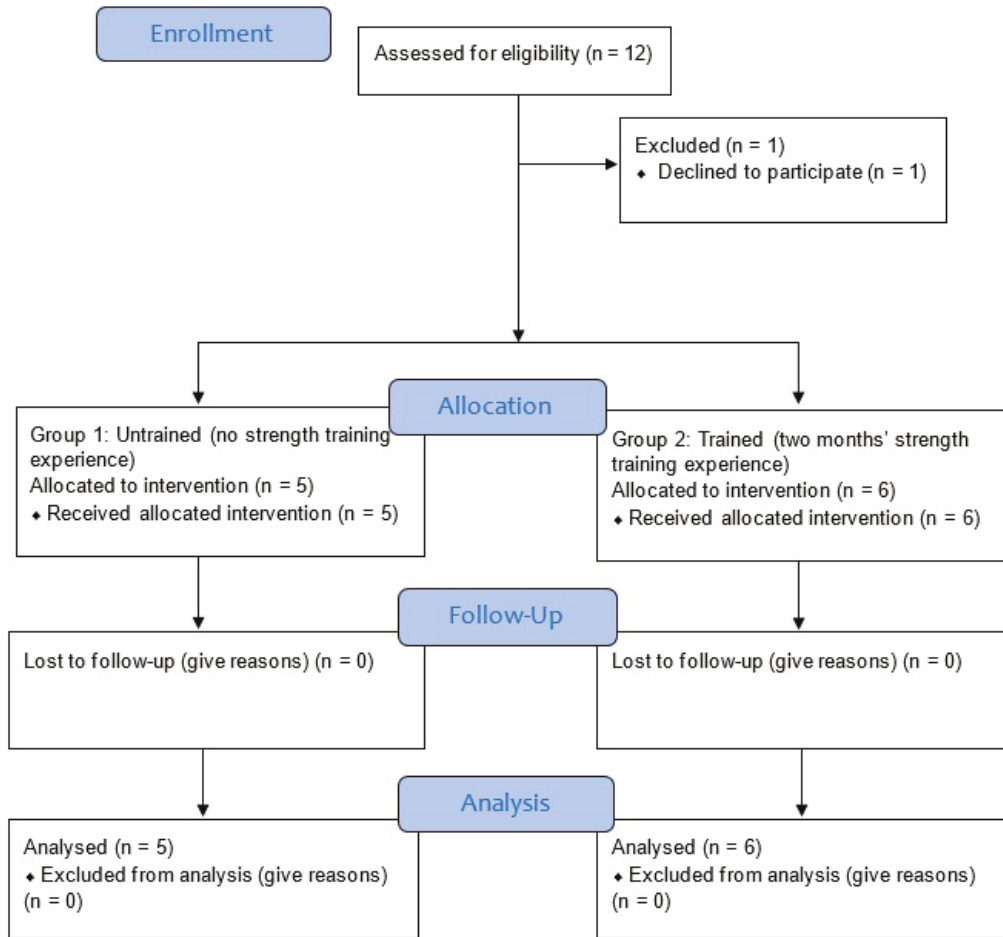


Figure 1. Study flow diagram.

Table 1. Exercise order (interval strength training).

Exercise Order	Training Session	Exercises		
		1	2	3
Sequence 1	Training 1	Shoulder press	Deadlift	Squat
	Training 2	Shoulder press	Deadlift	Squat
Sequence 2	Training 3	Deadlift	Shoulder press	Squat
	Training 4	Deadlift	Shoulder press	Squat
Sequence 3	Training 5	Squat	Shoulder press	Deadlift
	Training 6	Squat	Shoulder press	Deadlift

Each training was conducted on a different day for a total of six workouts.

## 2.2. Experimental Procedure

Every individual went to the exercise facility for eight separate sessions, during which they were tested, and data was collected [21]. All individuals were informed to have a meal 2~4 h before the test, to avoid alcohol and caffeine for 24 h before the test, and to avoid vigorous activity for 24~48 h before the test [22].

### 2.2.1. Session 1

Using a Karada scan-371 body scale, the initial body weight (kg), height (cm), and body-mass index (BMI; kg/m<sup>2</sup>) were assessed on the first appointment (Omron, Kyoto, Japan). The PAR-Q assessed each participant's physical fitness, and an expert researcher described the testing and training techniques to them. In this session, participants used the Cortex Metalyzer 3B (Cortex, Leipzig, Germany) to undertake an incremental cycling test while wearing a mask to measure their cardiorespiratory fitness and familiarize themselves with using a mask for workouts. Before each test, the flow and gas sensors were calibrated. The room's temperature and humidity were set to 22~27 °C and 52~64%, respectively. During the VO<sub>2</sub>max testing, the individual pedaled the bicycle at 60 revolutions per minute (rpm) against 25 W of resistance. At the start of each 2 min stage, the resistance was raised by 25 W [23]. The test was terminated when two of three requirements were met: a respiratory exchange ratio (RER) of  $\geq 1.1$  occurred, a heart rate (HR) within 10 beats or over their theoretical aged-predicted maximal HR (220—age) was reached, or an expression of Borg rate of perceived exertion (RPE) of  $\geq 16/20$  was achieved [24].

### 2.2.2. Session 2

Using an audible metronome, each participant did three to five sets of shoulder press, deadlift, and squat exercises using dumbbells at a tempo of 1.5 s up and 1.5 s down to attain their maximal 1RM. To account for any changes in the lifting cadence of participants, an audible metronome was utilized. Participants warmed up by performing eight to ten times with a light weight, three to five times with a moderate weight, and one to three times with a heavy weight. Following the initial sets, 1RM load was assessed by gradually rising the weight on consecutive tries until an individual was failing to carry out an attempt using appropriate technique and through the full range of motion [25]. A 2~4 min rest time was granted between each set for each participant. Between the 1RM shoulder press, deadlift, and squat, participants were given a 10~15 min rest interval during which they may walk, do minor stretches, and drink small quantities of water [26]. Participants completed three familiarization sessions on consecutive days after measuring 60% RM before commencing the regular trainings.

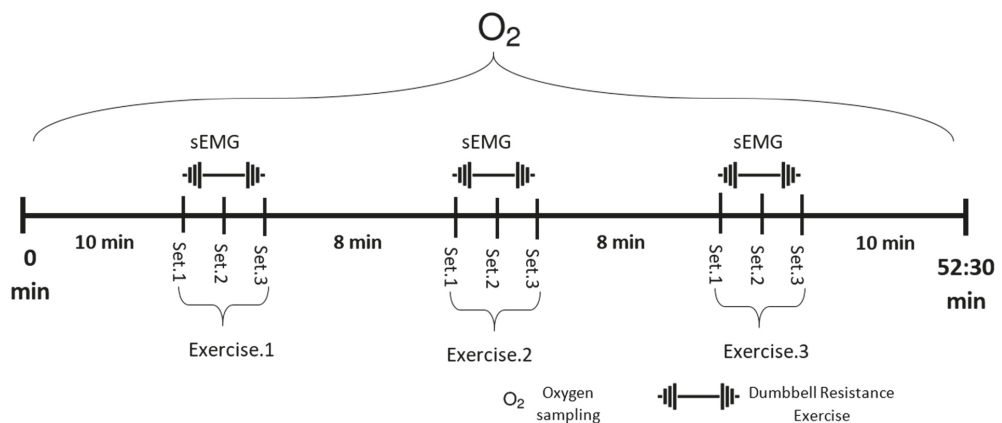
### 2.2.3. Sessions 3–8

Prior to the commencement of the training session, every individual stretched and warmed up for about 10 min. All individuals did six training sessions in an alternate order (Table 1), each at the same time of day, consisting of shoulder press, deadlift, and squat exercises at 60% of 1RM, three sets of 10 repetitions at a tempo of 1.5 s concentric and 1.5 s eccentric way using a metronome to control for possible changes in the lifting speed of individuals. There was a 2 min rest break between each set and an 8 min rest period between each type of exercise. Each training session was separated by 24~48 h of rest periods.

During each training session, oxygen consumption (VO<sub>2</sub>) was measured using a breath-by-breath analysis on a Cortex Metalyzer 3B. Individuals of the study were given details of the Borg rate of perceived exertion (RPE) scale (6~20) before they began training. For each training session, the VO<sub>2</sub>, RER, and HR were measured for a total of 52:30 min, which included the resting (10 min), exercise (30 s  $\times$  9 sets), rest after exercise (2~8 min), and recovery (10 min). A portable Omron sphygmomanometer (Omron Healthcare, Lake Forest, IL, USA) and an RPE scale were used to measure blood pressure (systolic (SBP) and

diastolic blood pressure (DBP)) and RPE before the beginning of a workout session and shortly after each exercise. The strength of the bilateral middle deltoid, lumbar erector spinae, quadriceps (rectus femoris), and hamstring (biceps femoris) muscles was measured two times before the first training and after the six sessions of training with a microFET3 dynamometer in newtons (N) (Hoggan Scientific, Salt Lake, UT, USA).

sEMG readings were recorded for training sessions 1, 3, and 6 with Noraxon wireless sensors (Noraxon USA, Scottsdale, AZ, USA) using bipolar AgCl<sub>2</sub> surface electrodes applied over eight muscles: RMD, right middle deltoid; LMD, left middle deltoid; RLES, right lumbar erector spinae; LLES, left lumbar erector spinae; RRF, right rectus femoris; LRF, left rectus femoris; RBF, right biceps femoris; and LBF, left biceps femoris. A 2 cm gap was provided between the two sensors of an electrode pair [18]. The sampling rate of the sEMG device was 1500 Hz, and raw signals during the training session were collected for each exercise set for 30 s (Figure 2).



**Figure 2.** Experiment training protocol. Each exercise took 30 s with a 2 min rest interval between each set and an 8 min rest interval between each type of exercise. VO<sub>2</sub> was measured during the entire session, while sEMG from eight different muscles was measured during the exercise periods.

Raw sEMG signals were treated in MATLAB R2021a (MathWorks, Natick, MA, USA) for signal analysis and processing. The raw signal for each set was filtered using a fourth-order Butterworth bandpass filter [27] (high pass with 50-Hz cutoff and low pass with 450-Hz cutoff) and smoothed, and the root mean square (RMS) was calculated. The RMS of each set of exercises was normalized by dividing the RMS value by the total training weight. The total training weight is a sum of bodyweight plus training load of every exercise [18]. The same researcher performed electrode placement throughout the training sessions.

### 2.3. Outcome Measures

The variables that were measured were: VO<sub>2</sub> (ml/kg/min), sEMG (microvolts (μV)), and muscle strength (N). For each exercise of training sessions 1, 3, and 6, VO<sub>2</sub> was recorded during the exercise, rest, and recovery periods, and results are presented as average values, while the normalized sEMG<sub>rms</sub> (μV) for each exercise set was calculated.

### 2.4. Statistical Analysis

Excel software was utilized to handle raw data from the Cortex analyzer and normalized sEMG rms (μV) from MATLAB. SPSS software (IBM SPSS Statistics vers. 19, Armonk, NY, USA) was used for statistical analysis. To confirm that the data was normal, a bell-shaped histogram was utilized. Results shows the baseline characteristics of the subjects. Study data are presented in the form of the mean ± standard deviation (SD),

and the significance level was set to  $p < 0.050$ . The data of this research were continuous repeated measurements and generalized estimating equations (GEEs) [28]; a backward deletion approach was utilized in SPSS to model the  $\text{VO}_2$  of various factors [21].  $\text{VO}_2$  (ml/kg/min) and normalized sEMG rms ( $\mu\text{V}$ ) of eight muscles throughout the workout session were used in the GEE analysis. GEE models were calculated in two categories:

- (1) **Group models:** Data from the exercise session, including  $\text{VO}_2$  (ml/kg/min) and normalized sEMG rms ( $\mu\text{V}$ ), were used to construct group models. The exercise type (shoulder press, deadlift, and squat) was analyzed as a factor, as well as the dependent variable ( $\text{VO}_2$  in ml/kg/min) and covariates or independent variables (normalized sEMG\_rms in  $\mu\text{V}$ ) of eight muscles for the untrained and trained groups across each set of data.
- (2) **Exercise models:** Data from the exercise phase, including  $\text{VO}_2$ , was used to estimate exercise models. Using the same variables as described before, three types of exercise models were established. For the untrained and trained groups, the factors considered were training session (sessions 1, 3, and 6), dependent variable ( $\text{VO}_2$  in ml/kg/min), and covariates (normalized sEMG rms in  $\mu\text{V}$ ) of eight muscles across each set of data.

For the GEE models, the estimate ( $\beta$ ), standard error (SE), 95% confidence interval (CI), and  $p$ -value were derived [29]. A more accurate estimate has a narrower 95% CI, whereas a less accurate estimate has a wider 95% CI [30]. The quasi-likelihood under an independent model criterion (QIC) was availed to calculate the fit of the GEE models for the group and exercise models, with a smaller QIC indicating a better model fit [31].

The difference within and between subjects for  $\text{VO}_2$  and normalized sEMG rms ( $\mu\text{V}$ ) of eight muscles was presented by using a two-way repeated measure ANOVA. Pre- vs. post-exercise muscle strengths were also compared using a two-way repeated measure ANOVA.

### 3. Results

#### 3.1. Baseline Characteristics of Participants

A total of 12 individuals were recruited for this study, one of which was eliminated. 11 of the 12 individuals were divided into two groups based on their strength training experience: untrained ( $n = 5$ ) and trained ( $n = 6$ ). In Table 2, all participants' baseline characteristics are reported, including age (years), gender (male/female), height (cm), body weight (kg), BMI ( $\text{kg}/\text{m}^2$ ), and 60% 1RM training weight of shoulder press, deadlift, and squat. The two groups differed in age, male/female ratio, height, body weight, BMI, and 60% 1RM training loads for the three exercises. Neither group had any side effects during or after the six workout sessions.

**Table 2.** Individual physical characteristics and training loads ( $n = 11$ ).

Participant	Age (Years)	Gender	Height (cm)	Body Weight (kg)	Body-Mass Index ( $\text{kg}/\text{m}^2$ )	Shoulder Press (60% RM)	Deadlift (60% RM)	Squat (60% RM)
<b>Untrained (<math>n = 5</math>)</b>								
S1	23	F	151	50	22	11.5	24	19
S2	21	F	158	59	23.7	9	16.5	14
S3	20	F	159	53	21	7	16.5	14
S4	21	F	165	53	19.5	9	16.5	14
S5	25	F	160	51	20.1	9	24	19
Mean $\pm$ SD	22.00 $\pm$ 1.79	–	158.60 $\pm$ 4.50	53.20 $\pm$ 3.12	21.26 $\pm$ 1.48	9.10 $\pm$ 1.43	19.50 $\pm$ 3.67	16.00 $\pm$ 2.45
<b>Trained (<math>n = 6</math>)</b>								
S1	26	M	175	92	30	16.5	29	26.5
S2	23	M	186	100	28.8	19	34	34
S3	29	F	160	55	21.6	9	34	29
S4	20	M	184	90	26.7	16.5	34	34

Table 2. Cont.

Participant	Age (Years)	Gender	Height (cm)	Body Weight (kg)	Body-Mass Index (kg/m <sup>2</sup> )	Shoulder Press (60% RM)	Deadlift (60% RM)	Squat (60% RM)
S5	29	F	165	58	21.5	16.5	39	34
S6	28	M	170	94	32.5	19	36.5	36.5
Mean ± SD	25.83 ± 3.34	–	173.33 ± 9.45	81.50 ± 17.96	26.85 ± 4.12	16.08 ± 3.36	34.42 ± 3.03	32.33 ± 3.44

Untrained; without strength training experience, Trained; two month’s strength training experience, SD, standard deviation; RM, repetition maximum of training weights in kilogram (kg) for both right and left sides; S, denotes participant number (untrained group *n* = 5 and trained group *n* = 6); F, female; M, male.

3.2. VO<sub>2</sub> Models of Three Training Sessions (Group Models)

VO<sub>2</sub> was predicted by the GEE model over the course of three training sessions but did not reach the level of significance. For the untrained group, right biceps femoris (RBFsEMG\_rms) [*p* = 0.330; 95% CI = −0.532~1.586] predicted VO<sub>2</sub>, and for the trained group, left middle deltoid (LMDsEMG\_rms) [*p* = 0.058; 95% CI = −0.010~0.607] predicted VO<sub>2</sub> without attaining the level of significance. QIC values for the group models were 368 and 867 for the untrained and trained groups, respectively (Table 3).

Table 3. Generalized estimating equations for oxygen consumption (VO<sub>2</sub>; ml/min/kg) predictions for three training sessions (*n* = 11).

Group	Model	Parameter	Estimate (β)	SE	95% CI (Lower~Upper)	<i>p</i>
UTr	Model 1 (QIC 368)	Intercept	9.068	0.530	8.030~10.107	0.000
		RBFsEMG-rms	0.527	0.541	−0.532~1.586	0.330
Tr	Model 2 (QIC 837)	Intercept	11.134	0.703	9.757~12.511	0.000
		LMDsEMG-rms	0.298	0.157	−0.010~0.607	0.058

UTr, untrained (*n* = 5); Tr, trained (*n* = 6). SE, standard error; 95% CI, confidence interval; QIC, quasi-likelihood under an independence model criterion; RBFsEMG-rms, right biceps femoris muscle root mean square surface electromyography (sEMG); LMDsEMG-rms, left middle deltoid.

3.3. VO<sub>2</sub> Models of Three Training Sessions (Exercise Models)

For the untrained group during the shoulder press, the left biceps femoris (LBFsEMG\_rms) [*p* = 0.000; 95% CI = −27.967~−19.721], right middle deltoid (RMDsEMG\_rms) [*p* = 0.000; 95% CI = 0.543~1.341], left middle deltoid (LMDsEMG\_rms) [*p* = 0.000; 95% CI = −1.016~−0.457], right biceps femoris (RBFsEMG\_rms) [*p* = 0.000; 95% CI = 9.890~24.286], and left rectus femoris (LRFsEMG\_rms) [*p* = 0.001; 95% CI = −1.646~−0.454] significantly predicted VO<sub>2</sub>. For the trained group, only the left rectus femoris (LRFsEMG\_rms) [*p* = 0.000; 95% CI = 4.131~11.240] attained a significant level in predicting VO<sub>2</sub>. QIC values for the shoulder press models were 102 and 82 for the untrained and trained groups, respectively (Table 4a).

Table 4. Generalized estimating equations for oxygen consumption (VO<sub>2</sub>; mL/min/kg) estimation for three training sessions (*n* = 11). (a) Shoulder Press. (b) Deadlift. (c) Squat.

(a)							
Exercise	Group	Model	Parameter	Estimate (β)	SE	95% CI (Lower~Upper)	<i>p</i>
Shoulder press	UTr	Model 1 (QIC 102)	Intercept	3.489	1.009	1.512~5.466	0.001
			LBFsEMG-rms	−23.844	2.104	−27.967~−19.721	0.000 *
			RMDsEMG-rms	0.942	0.204	0.543~1.341	0.000 *
			RBFsEMG-rms	17.088	3.673	9.890~24.286	0.000 *
			LMDsEMG-rms	−0.737	0.143	−1.016~−0.457	0.000 *



Table 4. Cont.

			LRF <sub>sEMG-rms</sub>	−1.050	0.304	−1.646~−0.454	0.001 *
	Tr	Model 2 (QIC 82)	Intercept	5.727	0.271	5.195~6.259	0.000
				LRF <sub>sEMG-rms</sub>	7.685	1.814	4.131~11.240
(b)							
Exercise	Group	Model	Parameter	Estimate (β)	SE	95% CI (Lower~Upper)	p
Deadlift	UTr	Model 3 (QIC 172)	Intercept	11.701	1.065	9.613~13.789	0.000
			RLES <sub>sEMG-rms</sub>	9.366	1.425	6.573~12.159	0.000 *
			LLES <sub>sEMG-rms</sub>	−10.428	2.030	−14.407~−6.448	0.000 *
			RBF <sub>sEMG-rms</sub>	−2.086	0.459	−2.985~−1.186	0.000 *
	Tr	Model 4 (QIC 320)	Intercept	9.314	1.339	6.689~11.939	0.000
			LLES <sub>sEMG-rms</sub>	3.362	1.506	0.411~6.313	0.026 *
(c)							
Exercise	Group	Model	Parameter	Estimate (β)	SE	95% CI (Lower~Upper)	p
Squat	UTr	Model 5 (QIC 76)	Intercept	10.328	0.875	8.612~12.043	0.000
			LBF <sub>sEMG-rms</sub>	−11.262	0.538	−12.318~−10.207	0.000 *
			RLES <sub>sEMG-rms</sub>	−3.318	0.514	−4.325~−2.312	0.000 *
			LLES <sub>sEMG-rms</sub>	6.891	1.420	4.108~9.675	0.000 *
			LMD <sub>sEMG-rms</sub>	0.653	0.126	0.406~0.901	0.000 *
			RBF <sub>sEMG-rms</sub>	1.758	0.391	0.992~2.524	0.000 *
	Tr	Model 6 (QIC 348)	Intercept	10.781	0.758	9.295~12.266	0.000
			LLES <sub>sEMG-rms</sub>	0.494	0.155	0.191~0.797	0.001 *

UTr, untrained (*n* = 5); Tr, trained (*n* = 6). \* Shows a significant difference *p* < 0.050. SE, standard error; 95% CI, confidence interval; QIC, quasi-likelihood under an independence model criterion; Root mean square surface electromyography (sEMG) of RMD<sub>sEMG-rms</sub>, right middle deltoid; LMD<sub>sEMG-rms</sub>, left middle deltoid; RLES<sub>sEMG-rms</sub>, right lumbar erector spinae; LLES<sub>sEMG-rms</sub>, left lumbar erector spinae; RBF<sub>sEMG-rms</sub>, right biceps femoris; LBF<sub>sEMG-rms</sub>, left biceps femoris; RRF<sub>sEMG-rms</sub>, right rectus femoris; LRF<sub>sEMG-rms</sub>, left rectus femoris.

For the untrained group during the deadlift, the right lumbar erector spinae (RLES<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = 6.573~12.159], left lumbar erector spinae (LLES<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = −14.407~−6.448], and right biceps femoris (RBF<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = −2.985~−1.186] significantly predicted VO<sub>2</sub>. For the trained group, the left lumbar erector spinae (LLES<sub>sEMG\_rms</sub>) [*p* = 0.026; 95% CI = 0.411~6.313] attained a significant level in predicting VO<sub>2</sub>. QIC values for deadlift models were 172 and 320 for the untrained and trained groups, respectively (Table 4b).

For the untrained group during squat, the left biceps femoris (LBF<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = −12.318~−10.207], right lumbar erector spinae (RLES<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = −4.325~−2.312], left lumbar erector spinae (LLES<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = 4.108~9.675], left middle deltoid (LMD<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = 0.406~0.901], and right biceps femoris (RBF<sub>sEMG\_rms</sub>) [*p* = 0.000; 95% CI = 0.992~2.524] significantly computed VO<sub>2</sub>. For the trained group, the left lumbar erector spinae (LLES<sub>sEMG\_rms</sub>) [*p* = 0.001; 95% CI = 0.191~0.797] attained a significant level in predicting VO<sub>2</sub>. Values of the QIC for the squat models were 76 and 348 for the untrained and trained groups, respectively (Table 4c).

### 3.4. Comparison between VO<sub>2</sub> and Normalized sEMGrms

Table 5 presents the within and between-subject comparison of different variables using repeated ANOVA. For the within-subject comparison, *p*-values were significantly different for only right lumbar erector spinae during shoulder press (SP) *p* = 0.016 \* and squat (SQ) *p* = 0.023 \* exercises. Meanwhile, for the between-subject comparison, oxygen consumption during shoulder press *p* = 0.005 \*, right and lumbar erector spinae during deadlift (DL) *p* = 0.048 \* and *p* = 0.033 \*, right and left rectus femoris during squat *p* = 0.014 \* and *p* = 0.032 \*, and left biceps femoris during deadlift *p* = 0.045 \* attained the significant level.

**Table 5.** Two-way repeated measures ANOVA three exercises (*n* = 11).

Parameters		Untrained ( <i>n</i> = 5)	Trained ( <i>n</i> = 6)	<i>p</i> -Value				
				Within Subject		Mauchly's Sphericity		Between Groups
				Rep	Tra	Rep	Tra	
Oxygen Consumption	SP	4.61 ± 0.48	7.04 ± 0.44	0.372	0.909	0.724	0.369	0.005 *
	DL	9.17 ± 0.80	10.81 ± 0.73	0.687	0.120	0.870	0.104	0.165
	SQ	9.35 ± 0.76	11.60 ± 0.70	0.254	0.058	0.909	0.478	0.057
Right Middle Deltoid	SP	3.45 ± 0.68	3.23 ± 0.62	0.860	0.570	0.832	0.009 <sup>a</sup>	0.817
	DL	1.06 ± 0.41	0.80 ± 0.37	0.555	0.214	0.016 <sup>b</sup>	0.012 <sup>c</sup>	0.653
	SQ	2.18 ± 0.56	1.65 ± 0.51	0.493	0.826	0.108	0.043 <sup>d</sup>	0.505
Left Middle Deltoid	SP	2.61 ± 0.57	2.31 ± 0.52	0.105	0.574	0.235	0.008 <sup>e</sup>	0.708
	DL	1.11 ± 0.42	0.75 ± 0.38	0.939	0.161	0.106	0.070	0.537
	SQ	2.04 ± 0.50	1.74 ± 0.46	0.546	0.284	0.285	0.240	0.661
Right Lumbar Erector Spinae	SP	0.08 ± 0.02	0.08 ± 0.02	0.815	0.016 *	0.958	0.012 <sup>f</sup>	0.989
	DL	0.66 ± 0.08	0.42 ± 0.07	0.433	0.126	0.037 <sup>g</sup>	0.000 <sup>h</sup>	0.048 *
	SQ	0.59 ± 0.07	0.39 ± 0.06	0.630	0.023 *	0.435	0.000 <sup>i</sup>	0.056
Left Lumbar Erector Spinae	SP	0.07 ± 0.02	0.08 ± 0.01	0.690	0.346	0.002 <sup>j</sup>	0.063	0.814
	DL	0.71 ± 0.08	0.44 ± 0.07	0.027	0.137	0.001 <sup>k</sup>	0.065	0.033 *
	SQ	0.59 ± 0.08	0.48 ± 0.07	0.420	0.519	0.000 <sup>l</sup>	0.000 <sup>m</sup>	0.314
Right Rectus Femoris	SP	0.12 ± 0.03	0.16 ± 0.03	0.272	0.365	0.696	0.441	0.450
	DL	0.45 ± 0.08	0.33 ± 0.08	0.190	0.138	0.076	0.019 <sup>n</sup>	0.333
	SQ	1.58 ± 0.17	0.89 ± 0.15	0.297	0.407	0.326	0.675	0.014 *
Left Rectus Femoris	SP	0.15 ± 0.04	0.17 ± 0.04	0.056	0.872	0.409	0.531	0.676
	DL	0.39 ± 0.08	0.28 ± 0.07	0.189	0.368	0.569	0.046 <sup>o</sup>	0.325
	SQ	1.46 ± 0.16	0.91 ± 0.14	0.595	0.249	0.553	0.209	0.032 *
Right Biceps Femoris	SP	0.04 ± 0.01	0.03 ± 0.01	0.672	0.504	0.004 <sup>p</sup>	0.126	0.719
	DL	0.64 ± 0.09	0.42 ± 0.08	0.477	0.724	0.159	0.692	0.087
	SQ	0.47 ± 0.08	0.31 ± 0.07	0.136	0.162	0.447	0.003 <sup>q</sup>	0.178
Left Biceps Femoris	SP	0.03 ± 0.01	0.03 ± 0.01	0.442	0.233	0.363	0.002 <sup>r</sup>	0.618
	DL	0.69 ± 0.09	0.41 ± 0.08	0.458	0.357	0.105	0.514	0.045 *
	SQ	0.46 ± 0.06	0.31 ± 0.06	0.104	0.286	0.497	0.873	0.092

Rep, number of repetitions; Tra, number of trainings. Mean ± standard error; level of significance, \* *p* < 0.05. SP, shoulder press; DL, deadlift; SQ, squat. <sup>a</sup> For right middle deltoid, Mauchly's sphericity *p* = 0.009 for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.59, which is lower than 0.75, and after correction, sphericity assumed for training did not reach the significance level, *p* = 0.888. <sup>b,c</sup> Mauchly's sphericity *p* = 0.016 and 0.012 for repetitions and training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.61 and 0.60, which are lower than 0.75, and after correction, sphericity assumed for repetitions and training did not reach significance level *p* = 0.794 and *p* = 0.044, respectively. <sup>d</sup> Mauchly's sphericity *p* = 0.043 for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.65, which is lower than 0.75, and after correction, sphericity assumed did not reach the significant level, *p* = 0.846. <sup>e</sup> For left middle deltoid, Mauchly's sphericity *p* = 0.008 for training, the assumption for the difference in equal variance was not met, and Greenhouse-Geisser Epsilon was 0.58, which is lower than 0.75, and after correction, sphericity assumed for training did not reach significant level, *p* = 0.511. <sup>f</sup> For right lumbar erector spinae, Mauchly's sphericity *p* = 0.012 for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.60, lower than 0.75, and after correction, sphericity did not reach significant level, *p* = 0.194. <sup>g,h</sup> Mauchly's sphericity *p* = 0.037 and 0.000 for repetition and training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon were 0.64 and 0.52, which are lower than 0.75, and after correction, sphericity assumed did not reach the significant level *p* = 0.760 and *p* = 0.703, respectively. <sup>i</sup> Mauchly's sphericity *p* = 0.000 for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.52, lower than 0.75, and after correction, sphericity assumed did not reach

the significant level,  $p = 0.269$ . <sup>j</sup> For left lumbar erector spinae, Mauchly's sphericity  $p = 0.002$  for repetition, assumption for difference in equal variance not met. Greenhouse-Geisser Epsilon was 0.56, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level,  $p = 0.714$ . <sup>k</sup> Mauchly's sphericity  $p = 0.001$  for repetition, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.55, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level  $p = 0.524$ . <sup>l,m</sup> Mauchly's sphericity  $p = 0.000$  for repetition and training, assumption for the difference in equal variance not met. Greenhouse-Geisser Epsilon was 0.53 and 0.50, which were lower than 0.75, and after correction, sphericity assumed did not reach significant levels  $p = 0.155$  and  $p = 0.624$ , respectively. <sup>n</sup> For right rectus femoris, Mauchly's sphericity  $p = 0.019$  for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.62, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level  $p = 0.112$ . <sup>o</sup> For left rectus femoris, Mauchly's sphericity  $p = 0.046$  for training, the assumption for difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.65, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level  $p = 0.143$ . <sup>p</sup> For right biceps femoris, Mauchly's sphericity  $p = 0.004$  for repetition, the assumption for difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.57, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level  $p = 0.556$ . <sup>q</sup> Mauchly's sphericity  $p = 0.003$  for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.56, which is lower than 0.75, and after correction, sphericity assumed reached significant level  $p = 0.027$ . <sup>r</sup> For left biceps femoris, Mauchly's sphericity  $p = 0.002$  for training, the assumption for the difference in equal variance was not met. Greenhouse-Geisser Epsilon was 0.56, which is lower than 0.75, and after correction, sphericity assumed did not reach significant level  $p = 0.296$ .

The Mauchly's sphericity reported that during shoulder press, the left lumbar erector spinae (0.002 <sup>j</sup>), right biceps femoris ( $p = 0.004$  <sup>p</sup>) between repetitions and right middle deltoid ( $p = 0.009$  <sup>a</sup>), left middle deltoid (0.008 <sup>e</sup>), right lumbar erector spinae ( $p = 0.012$  <sup>f</sup>), and left biceps femoris ( $p = 0.002$  <sup>r</sup>) between trainings did not show equal variance in means. The sphericity during deadlift showed no equal variance in means for the right middle deltoid (0.016 <sup>b</sup>), right lumbar erector spinae ( $p = 0.037$  <sup>g</sup>), and left lumbar erector spinae (0.001 <sup>k</sup>) between repetitions and right middle deltoid ( $p = 0.012$  <sup>c</sup>), right lumbar erector spinae ( $p = 0.000$  <sup>h</sup>), right rectus femoris ( $p = 0.019$  <sup>n</sup>), and left rectus femoris ( $p = 0.046$  <sup>o</sup>) between trainings. During squat exercise, the sphericity analysis did not report equal variance in means for the left lumbar erector spinae (0.000 <sup>l</sup>) between repetitions and right middle deltoid ( $p = 0.043$  <sup>d</sup>), right lumbar erector spinae ( $p = 0.000$  <sup>i</sup>), left lumbar erector spinae ( $p = 0.000$  <sup>m</sup>), and right biceps femoris ( $p = 0.003$  <sup>q</sup>) between trainings.

### 3.5. Muscle Strength Pre vs. Post

Values of the muscle strength of the bilateral middle deltoid, lumbar erector spinae, quadriceps (rectus femoris), and hamstring (biceps femoris) are presented in Table 6. The within-subject comparison showed a significant difference for right and left lumbar erector spinae. Meanwhile, all the muscles attained the significance level ( $p < 0.050$ ) for group comparison, except for the left middle deltoid muscle. For the untrained group, the right and left lumbar erector spinae, right and left rectus femoris, and right biceps femoris values increased after the six training sessions, while in the trained group, values of all of the muscles increased after the six training sessions, except for the left rectus femoris and right biceps femoris.

**Table 6.** Two-way repeated measures ANOVA for manual muscle strength (MMT) ( $n = 11$ ).

Parameters	Untrained ( $n = 5$ )	Trained ( $n = 6$ )	<i>p</i> -Value	
			Within Subject Pre vs. Post	Between Groups
Right Middle Deltoid	85.08 ± 22.01	156.28 ± 20.09	0.926	0.041 *
Left Middle Deltoid	85.49 ± 17.82	136.28 ± 16.27	0.951	0.065
Right Lumbar Erector Spinae	130.35 ± 23.14	212.27 ± 21.12	0.016 *	0.028 *
Left Lumbar Erector Spinae	134.93 ± 18.87	207.78 ± 17.23	0.017 *	0.019 *
Right Rectus Femoris	281.61 ± 34.25	426.36 ± 31.27	0.170	0.012 *

Table 6. Cont.

Parameters	Untrained (n = 5)	Trained (n = 6)	p-Value	
			Within Subject Pre vs. Post	Between Groups
Left Rectus Femoris	283.40 ± 34.01	408.43 ± 31.04	0.085	0.024 *
Right Biceps Femoris	208.81 ± 19.06	249.48 ± 17.40	0.569	0.149
Left Biceps Femoris	181.45 ± 22.73	256.76 ± 20.75	0.950	0.037 *

Mean ± standard error; participants; n = 11. Muscle strength was assessed through a dynamometer in newtons (N) two times before and after six training sessions. \* Shows a significant difference  $p < 0.050$ . Mauchly's sphericity and Greenhouse-Geisser Epsilon were equal to 1 for every muscle, so the assumption for the difference in equal variance was met.

#### 4. Discussion

The present study utilized GEE modeling to predict  $VO_2$  for three strength training exercises, including the shoulder press, deadlift, and squat with dumbbells in young participants. For the group models, the right biceps femoris predicted model for  $VO_2$  in the untrained group, while the left middle deltoid in the trained group without attaining the level of significance (Table 3). For the exercise models, the right and left middle deltoid, right and left biceps femoris, and left rectus femoris for the shoulder press, right and left lumbar erector spinae, and right biceps femoris for the deadlift and the right and left lumbar erector spinae, biceps femoris, and left middle deltoid for the squat significantly predicted the GEE models (Table 4a–c).

No single previous study predicted  $VO_2$  by sEMG of individual muscles during moderate-intensity strength exercises. Because there is not a lot of studies on GEE modeling during strength training exercises, it is difficult to compare our computed models with previously available research. One study reported the association between  $VO_2$  and sEMG RMS during cycling exercise and reported global sEMG measured from vastus lateralis muscle as a good predictor of energy expenditure in trained cyclists [32]. In some previous studies, one study reported the relation between  $VO_2$  and sEMG responses of the anterior tibialis (TA), gastrocnemius medial (MG), gastrocnemius lateral (LG), and soleus muscles during different speeds of treadmill walking in young, healthy males; correlations between  $VO_2$  and sEMG were 0.69–0.87 for those muscles [19]. Another study conducted on young males reported the effect of 6 weeks of strength training exercises and whole-body vibration on changes in normalized  $VO_2$  and sEMG; they monitored the rectus femoris muscle during squats and lateral gastrocnemius during heel raises [18].

The main goal of the present research was to calculate GEE models for  $VO_2$  in two categories (1) group and (2) exercise types. For the group models, none of the groups significantly predicted  $VO_2$  from sEMG RMS of individual muscles, but for exercise type models, the shoulder press exercise showed significant relations of the right and left middle deltoid, right and left biceps femoris, and left rectus femoris with  $VO_2$  for the untrained group [QIC = 102, \*  $p = 0.000$ ], while in the trained group, only the left rectus femoris [QIC = 82, \*  $p = 0.000$ ] were significantly correlated with the  $VO_2$ . Lower QIC and significant  $p$ -values for the trained group [QIC = 82 vs. 102 and \*  $p = 0.000$  vs. \* 0.000] are suggestive of a better GEE model than that for the untrained group (Table 4a). For the deadlift and squat exercises, the untrained group models were more predictive than those of the trained group [QIC = 172 vs. 320 and \*  $p = 0.000$  vs. \* 0.026] and [QIC = 76 vs. 348 and \*  $p = 0.000$  vs. \* 0.001] (Table 4b,c) [29,31]. The reason why the correlations between the two groups differed lies in the fact that the untrained group participants had no previous experience of strength training, and their 60% 1RM was lower, so their muscle activation occurred differently than that in the trained group. Another factor that may have affected the results was the gender because the untrained group consisted mostly of female participants and trained group males.

Pre vs. post static muscle strength is shown in Table 6. Some of the muscles' strength increased after six training sessions, but out of the eight muscles, not a single one reached a significant level, because 2-week trainings are not enough to increase muscle strength as adaptation in strength would require about 12 or more weeks of consecutive trainings. However, this was not the concerned objective of this study. The changes in the  $VO_2$  and sEMG\_rms after six trainings reported higher  $VO_2$  values in both groups, particularly during the squat and deadlift exercises, which may have been due to the greater training loads of these exercises. Meanwhile, the sEMG\_rms amplitude was higher for the bilateral middle deltoid during the shoulder press, because it is the major muscle group that is active during that exercise. In the deadlift, the right and left middle deltoid, lumbar erector spinae, and biceps femoris were more active than the rectus femoris, and only the bilateral deltoid and rectus femoris were active during squatting.

Limitations: (1) Because this research's sample size was limited ( $n = 11$ ), generalization of the findings can only be addressed when the study procedure has been tested on a wider population. More studies with a higher sample size are required in the future to investigate these associations, which should include various body muscle strength training exercises with varying cadence and RM loads, as well as training sessions to failure. (2) In order to conduct a well-controlled research study, an adequate dietary evaluation, including BMI and dietary chart, might be incorporated to record a more sensitive link between  $VO_2$  and sEMG. (3) The trained group in this study had just 2 months of strength training experience; however, in the future, individuals with greater training experience might be recruited to establish the study protocol and translate the study outcomes into the general population. (4) The study reported results with mostly females in the untrained group and males in the trained group, which could have biased the results. Therefore, future studies will be conducted by controlling the gender factor to avoid such biases or may test such exercise protocols on a single gender. (5) This is one of the new ideas to use GEE modeling to relate and predict  $VO_2$  using localized muscular activity, so in the future, more studies of this kind may be required to confirm this study's findings and improve exercise prescriptions for health and fitness purposes for the human population.

## 5. Conclusions

It is concluded that  $VO_2$  can be predicted from sEMG RMS during moderate-intensity strength training exercises. Because this study obtained significant  $VO_2$  prediction models during shoulder press, deadlift, and squat exercises using the right and left middle deltoid, right and left lumbar erector spinae, left rectus femoris, and right and left biceps femoris sEMG RMS for the untrained and trained groups.

Practical Implications: These kinds of correlations can help provide a deeper understanding of muscular activity and fatigue during strength training and facilitate relating and predicting metabolic parameters like  $VO_2$  with localized muscular activity. The exercise intensity and volume are two key parameters when designing training programs for all ages, but how various exercise programs alter relationships between oxygen consumption and muscular activity is still an area that future research needs to explore. This study tried to understand the relation between  $VO_2$  and sEMG during moderate-intensity strength exercise, which consisted of three different exercises in a single session. It offers another aspect of exercise prescription in rehabilitation and sports sciences to enhance one's health and fitness of normal, athletic, and chronic disease people.

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**Data Availability Statement:** Not applicable.

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## Abbreviations

ACSM: American College of Sports Medicine; AHA, American Heart Association;  $\beta$ , estimate; BMI, body-mass index; BP, blood pressure; cm, centimeter; CI, confidence interval; DBP, diastolic blood pressure;  $\Delta d$ , difference; EE, energy expenditure; GEE, generalized estimating equation; HR, heart rate; IRB, institutional review board; kg, kilogram; LBF, left biceps femoris; LG, lateral gastrocnemius; LLES, left lumbar erector spinae; LMD, left middle deltoid; LRF, left rectus femoris; MG, medial gastrocnemius; ml/kg/min, milliliter per kilogram per minute; n, number of participants; N, newtons; PAR-Q, Physical Activity Readiness Questionnaire; QIC, quasi-likelihood under an independence model criterion; RBF, right biceps femoris; RER, respiratory exchange ratio; RLES, right lumbar erector spinae; RM, repetition maximum; RMD, right middle deltoid; RMS/rms, root mean square; RPE, Borg rate of perceived exertion; RPM, revolutions per minute; RRF, right rectus femoris; SD, standard deviation; SE, standard error; sEMG, surface electromyography; SBP, systolic blood pressure; SPSS, Statistical Package for the Social Sciences; TA, tibialis anterior; Tr, trained; UTr, untrained;  $\mu V$ , microvolt;  $VO_2$ , oxygen consumption.

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Article

# Inline Skating as an Additional Activity for Alpine Skiing: The Role of the Outside Leg in Short Turn Performance

Vjekoslav Cigrovski <sup>1</sup>, Mateja Očić <sup>1</sup>, Ivan Bon <sup>1,\*</sup>, Branka Matković <sup>1</sup> and Peter Šagát <sup>2</sup>

- <sup>1</sup> Laboratory for Sports Games, Faculty of Kinesiology, University of Zagreb, 10000 Zagreb, Croatia; vjekoslav.cigrovski@kif.unizg.hr (V.C.); mateja.ocic@kif.unizg.hr (M.O.); branka.matkovic@kif.unizg.hr (B.M.)  
<sup>2</sup> Health and Physical Education Department, Prince Sultan University Riyadh, Riyadh 66833, Saudi Arabia; sagat@seznam.cz  
\* Correspondence: ivan.bon@kif.unizg.hr

**Abstract:** The complexity of skiing movements urges recreational alpine skiers and competitors to undertake many specific skill trainings not only during the season but also during the off-season using alternative sports. In AS, the role of the outside leg is crucial for successful turn performance. By measuring kinematic and kinetic parameters, we could define whether there is an objective similarity of the role and the movements of the outside leg while performing a turn in AS to those in the most used additional activity, IS. The sample consisted of ten female alpine ski instructors (age  $31.6 \pm 8.23$ , height  $170.66 \pm 7.32$  cm, weight  $60.16 \pm 7.58$  kg). Overall, 280 turns were analyzed (140 for AS and 140 for IS). For the purposes of this study, the variable sample consisted of 14 variables in total. For the detection of differences between short turn performance in AS and IS, MANOVA was used. The main findings of our study are defined similarities in pressure distribution during IS and AS and noticeable differences in the kinematic parameters of the outside leg between the mentioned activities. Based on the gathered results, recreational alpine skiers should be aware that IS cannot be used for the purpose of AS adoption, but rather as a dry-land additional activity for AS preparation.

**Keywords:** biomechanical analysis; pressure insoles; Xsens motion capture system; performance analysis; recreational skiers; dry-land training

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## 1. Introduction

Alpine skiing (AS) is a specific activity consisting of complex and uncommon movements of the human body [1]. Specific movements that a skier performs are then transferred to ski boots, skis and consequently the snow surface. Finally, the ground reaction force (GRF) then enables the alpine skier to steer the skis and complete turns [2]. In order to control the speed and direction during a descent, the skier has to maintain the optimal posture by timely shifts in the center of their mass, optimally applying pressure and utilizing GRF [3].

In addition, the edging angle is another important factor that secures the skier's reliable support on the snow surface. If all of the abovementioned factors are considered, each phase of the turn will be successfully performed without skidding and sliding [4–6].

The outside leg is more exposed to skidding when compared to the inside leg when the steepness of the terrain and applied pressure on both legs are the same. Therefore, the role of the outside leg is especially important in the conditions of very steep and icy terrain. That is the main reason for shifting the pressure predominantly on the outside leg, which is placed further from the projection of the center of mass [7]. In order to be successful in applying pressure on the outside ski, the skier must be in a specific body position, especially in the finishing phases of a ski turn. This position requires hip and knee flexion, and also a side arc that is achieved by moving the upper and lower leg in the direction of the center of the turn and the upper body in the opposite direction [8–10].



The complexity of the skiing movements urges alpine ski competitors to perform a large number of specific skills trainings not only during the season but also during the off-season using alternative sports. Those must include similar body positions, relations between body segments, coordination structures, general movements and muscle contractions when compared to the primary activity [11].

One of the most frequently used alternative activity for physical and technical preparation for AS is inline skating (IS) [12]. IS enables the execution of short parallel turns that are similar to skiing turns [13,14]. Even though there are clear similarities between the two mentioned activities, the surface, equipment and factors related to both are different [11]. IS is usually performed on a less steep terrain, which is also more solid (concrete or asphalt). Furthermore, during the turn performance in IS, the contact with the terrain surface is made through wheels placed exactly beneath the feet's centers. In AS, on the other hand, the contact is made by the ski edge, which is more lateral. In addition, the speed also varies, which can affect relations between body segments and, finally, forces (GRF and centrifugal force), when observing the same moment of the turn performance in both activities [15].

Comparing the turn execution on roller skates and skis, it can be assumed that alpine skiers can benefit in terms of technical training and physical conditioning by using IS as an additional activity [16]. Research conducted by Kroll et al. [11] suggests a similar sequence of body movements during IS and AS.

In AS, as already mentioned, the role of the outside leg is considered to be crucial in successful turn performance. The inside leg has the role of a support, while the outside leg mostly controls the direction, duration and speed, and also prevents the skier from skidding by adjusting the edging angle in the turn [9]. Research regarding movement analysis in IS is scarce so it is not clear whether the described movement patterns and roles of the inside and the outside legs are the same while executing parallel turns in IS.

To objectively identify and compare the roles of the outside leg in IS and AS, it is necessary to determine the joint angles and pressure forces that occur while performing a turn.

Combined kinematic and kinetic analysis can provide objective evaluation of the outside leg movements and pressure distributions while skating and skiing. By measuring kinematic and kinetic parameters in this research, we could define whether there is an objective similarity of the role and the movements of the outside leg while performing a turn in IS and AS. The results gained emphasize the function of IS as an additional activity for developing and maintaining technical and physical conditioning outside the skiing season for competitive but also recreational level alpine skiers, who do not have the opportunity to spend a great amount of time on the ski slopes. Therefore, alternative activities are even more necessary to prepare them for their short skiing season.

We hypothesized that the short turn execution during AS induces a higher pressure on the outside foot than it does in IS due to the slope steepness. Regarding the pressure distribution, we assumed there would be no significant differences in the load distribution during the turn execution in both activities. Furthermore, we assumed there would be similarities in the kinematic parameters of the lower limbs during short turn performance in IS and AS.

## 2. Materials and Methods

**Participants:** The sample consisted of ten female alpine ski instructors (age  $31.6 \pm 8.23$ , height  $170.66 \pm 7.32$  cm, weight  $60.16 \pm 7.58$  kg). They did not report any prior injuries that could affect their AS and IS technique or kinetic and kinematic variables while performing turns in both activities. All participants had previous experience in inline skating and had finished basic inline skating school. Participants gave their written consent to participate in this study after being informed in detail about the aims and protocol of the research. The Faculty of Kinesiology, University of Zagreb (Croatia) Ethics Committee approved the study, which was performed following the ethical standards of the Declaration of Helsinki.

Variables: The mentioned variables were selected based on relevant studies focused on the kinematic and kinetic analysis of AS technique [6,8–10,17–21]. Based on those studies, the determinants of a successfully executed turn are a proper pressure distribution of certain foot regions in specific phases of the turn and a proper edging angle that requires optimal relations between lower body segments. Considering the fact that lower extremities have a key role in performing a successful turn—especially when observing the outside leg—for the purpose of the kinematic analysis the focus was on parameters that describe the movements of the outside leg. The main movements that enable a skier to steer their skis are flexion and extension (up–down movements), abduction and adduction (lateral movements) and rotations [4]. Our research focused on variables that represent the amplitude of movements in some joints of the lower extremities (knee and hip), with emphasis on flexion–extension and abduction–adduction. Kinetic variables refer to the pressure of the outside leg during the execution of turns. All the available regions of foot pressure enabled by the manufacturer were analyzed. This furthermore enabled the observation of the pressure of the heel and the lateral and medial parts of the foot throughout the turn. The skis are steered throughout the turn mainly by moving the lower extremities. Because of the already mentioned role of the outside leg as a crucial factor in optimal turn execution, it can be assumed that the potential differences or similarities between observed activities would appear in the analysis of the outside leg parameters.

Analyses of kinetic and kinematic parameters were conducted on short parallel turns in two activities—AS and IS. Overall, 280 turns were analyzed (140 for AS and 140 for IS). For the purposes of this study, the variable sample consisted of 14 variables in total. The following kinetic variables were measured: the maximum force of the right foot in the left turn (Max\_R\_LT); the force of the lateral side of the right foot in the left turn (Lat\_R\_LT); the force of the medial side of the right foot in the left turn (Med\_R\_LT); the force of the right heel in the left turn (He\_R\_LT); the maximum force of the left foot in the right turn (Max\_L\_RT); the force of the lateral side of the left foot in the right turn (Lat\_L\_RT); the force of the medial side of the left foot in the right turn (Med\_L\_RT); and the force of the left heel in the right turn (HE\_F\_L\_RT). All results are shown in newtons (N). Pressure data are also presented in terms of mean body weight (MBW= pressure (kg)/mean body weight of all participants (kg)).

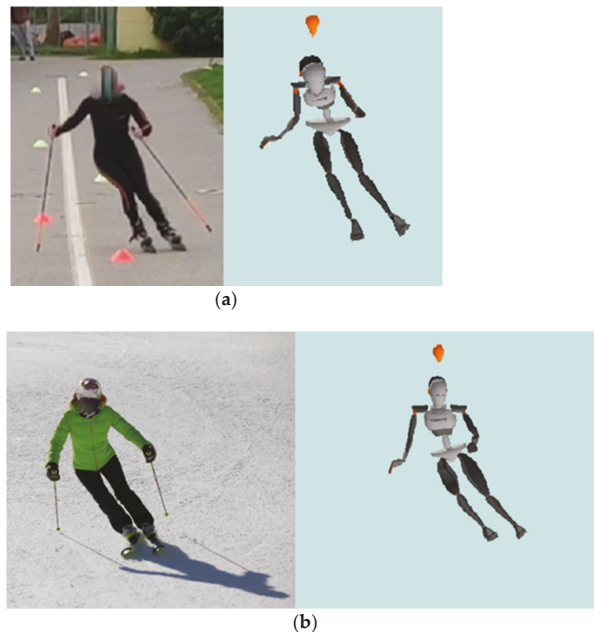
The following kinematic variables were measured ( $180^\circ$  = knee and hip fully extended): the angle of the right knee flexion in the left turn (R\_KNEE\_LT); the angle of the right hip flexion in the left turn (R\_HIP\_F\_LT); the angle of the right hip abduction in the left turn (R\_HIP\_AB\_LT); the angle of the left knee flexion in the right turn (L\_KNEE\_RT); the angle of the left hip flexion in the right turn (L\_HIP\_F\_RT); and the angle of the left hip abduction in the right turn (L\_HIP\_AB\_RT). All results are shown in degrees ( $^\circ$ ).

All data were obtained at the moment when participants' feet were in a position parallel to the fall line (Figure 1). For the purposes of field testing, it is necessary to use systems and equipment that are not invasive and to secure accurate data without affecting the athlete's performance [17–21].

The pressure distribution (kinetic variables) during AS and IS was measured with insoles designed for pressure detection (Novel, Pedar). Due to their construction (2 mm thin and very light), insoles had minimal influence on turn performance during testing of both dynamic activities. For the purposes of this study, output rate was set at 100 Hz and the data were derived from the corresponding software (Novel, Loadpad 25.3.6). The reliability and validity of Novel pressure insoles for analyzing pressure distribution were confirmed in previous studies of similar activities [2,11,22].

Kinematic parameters were measured by an Xsens MVN Link inertial suit system. The system consisted of 17 three-dimensional accelerometers/gyroscopes/magnetometers and a battery. The output rate was set at 240 Hz, which ensures real-time human motion analysis without affecting the movement or rate of motion. Data were derived from the corresponding MVN BIOMECH software (Xsens, MVN Studio 4.4, firmware version 4.3.1, Enschede, the Netherlands). Previous studies confirmed the reliability and validity of the

Xsens kinematic suit for analyzing kinematic parameters (joint angles) in activities similar to those in this study [21,23].



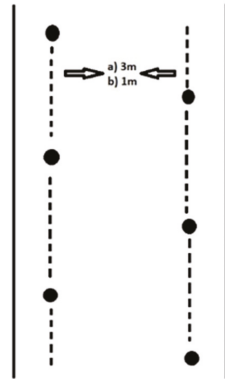
**Figure 1.** (a) Preview of IS turn; (b) preview of AS turn.

**Protocol of investigation:** The protocol was the same for all participants and included measuring anthropometric characteristics, adjusting the Xsens suit and adjusting suitable pressure insoles. Anthropometric characteristics were used for adjusting the sensors and calibration of the Xsens system. The calibration was performed according to the standard of procedure advised by the manufacturer (Xsens technologies B.V., Netherlands). In addition, standard calibration of pressure insoles was performed according to the manufacturer's advice (Novel GmbH, Munich, Germany). All turns were recorded with a video camera (Panasonic GH 5) for the purpose of synchronizing both kinetic and kinematic systems. In order to successfully synchronize the systems, participants were asked to alternately lift their skis or skates two times off the surface just before starting their descent. That was the starting moment when the time lapse of both systems was aligned.

Before testing procedures in AS, participants performed a free warm up run and a trial run in a defined corridor. Then, kinematic and kinetic parameters were measured with corresponding systems as participants performed short parallel turns in the defined corridor. The protocol of investigation was very similar to that used for the IS measurements. After adjusting kinematic and kinetic systems, participants had a free warm up run and then a trial run in the defined corridor. Afterwards, measurement of predefined kinematic and kinetic parameters was conducted while performing short parallel turn performance both on skis and skates. Overall testing was conducted for four days; two days for measuring turns in AS and two days for measuring turns in IS. Testing procedures on the ski slope were performed in the morning hours to secure better snow conditions. The average incline of the slope was 20°. Participants had suitable skiing equipment (slalom skis and adjusted ski boots). Short parallel ski turns were performed in a 3 m wide corridor, according to protocols defined in alpine ski schools. Each turn had to be performed

from one end of the corridor to another. Each participant performed 16 turns, but due to mistakes in motor movement (skidding, sliding, etc.), several turns were excluded from further analysis.

Skating turns were performed in sunny weather conditions in order to secure a dry surface. The corridor width was 1 m and the average incline was 10°. Participants also performed 16 turns, but some of them were excluded from further analysis due to certain mistakes in motor movement. The defined corridor for both skiing and skating turns is presented in Figure 2.



**Figure 2.** Preview of the corridor for short turn execution (a)—corridor width for AS; (b)—corridor width for IS).

**Statistical analysis:** Statistical package Statistica version 13.5.0.17 (TIBCO Software Inc., Palo Alto, CA, USA) was used for data analysis. Basic descriptive parameters for all measured variables were calculated. The normality of data distribution was tested by the Kolmogorov–Smirnov test. MANOVA was used for the detection of differences between turn performance in AS and IS. The results were considered significant when  $p < 0.05$ . With the use of the G\*power program, the sample size (number of turns) was calculated ( $n = 122$ ), which was needed for the measurement procedure with statistical significance of  $p < 0.05$ ; statistical power 0.95; effect size 0.25; 2 groups.

### 3. Results

Differences in kinematic and kinetic parameters between IS and AS were tested by MANOVA; results are shown in Table 1.

**Table 1.** Results of MANOVA for short turns executed in IS and AS.

Test	Lambda Value	F	<i>p</i>
Wilks	0.05	169.1	0.00 *

Legend: \*  $p < 0.05$ .

Results shown in Table 1 show a statistically significant difference between observed short turns executed in IS and AS ( $F = 169.1$ ;  $p < 0.01$ ).

Basic descriptive parameters of each tested kinematic variable along with the results of MANOVA are presented in Table 2.

Table 2 shows differences in all observed kinematic variables between short turns executed during IS and AS ( $p < 0.01$ ). In the left turn, the knee flexion differed significantly between IS and AS, with higher angle values detected in IS than it was in AS (157.43° compared to 139.56°). In addition, the flexion of the right hip was significantly different, with higher angle values determined while performing a turn in AS (145.01° compared to

139.03). Moreover, the hip abduction was significantly greater while performing a turn in AS than it was in IS (163.68° compared to 170.73°).

**Table 2.** Basic descriptive statistical parameters and MANOVA of kinematic parameters for IS and AS.

Variable	IS		AS		F	p
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD		
R_KNEE_LT (°)	157.43 ± 6.47	139.56 ± 2.07	483.64	0.00 *		
R_HIP_F_LT (°)	145.01 ± 6.49	139.03 ± 5.84	32.82	0.00 *		
R_HIP_AB_LT (°)	170.73 ± 2.24	163.68 ± 2.18	354.73	0.00 *		
L_KNEE_RT (°)	159.93 ± 5.42	140.30 ± 2.95	142.87	0.00 *		
L_HIP_F_RT (°)	145.39 ± 6.53	138.33 ± 9.35	20.37	0.00 *		
L_HIP_AB_RT (°)	170.51 ± 1.25	163.00 ± 1.48	707.74	0.00 *		

Legend: \*  $p < 0.05$ ; R\_KNEE\_LT—angle of the right knee flexion in the left turn; R\_HIP\_F\_LT—angle of the right hip flexion in left turn; R\_HIP\_AB\_LT—angle of the right hip abduction in the left turn; L\_KNEE\_RT—angle of the left knee flexion in the right turn; L\_HIP\_F\_RT—angle of the left hip flexion in the right turn; L\_HIP\_AB\_RT—angle of the left hip abduction in the right turn; (°)—degrees.

The results were the same when analyzing right turns. The knee flexion was greater while performing a short turn in AS compared to IS (140.30° compared to 159.93°). The hip was more flexed while performing a short turn in AS than it was in IS (138.33° compared to 145.39°). Regarding the hip abduction, it was determined to be greater during AS than it was in IS (163.00° compared to 170.51°).

Basic descriptive parameters of each tested kinetic variable, along with results of MANOVA, are presented in Table 3.

**Table 3.** Basic descriptive statistical parameters and MANOVA of kinetic parameters for IS and AS.

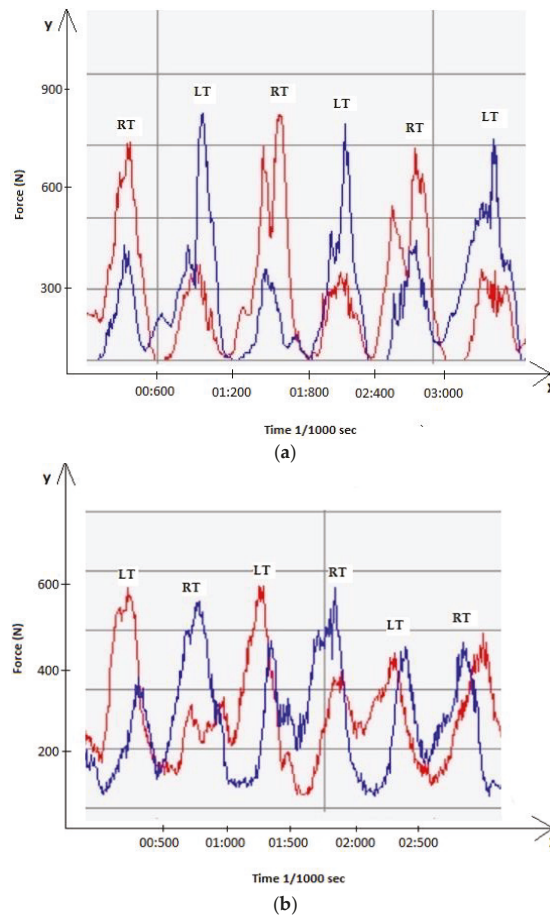
Variable	IS		AS		F	p
	Mean ± SD	IS MBW **	Mean ± SD	AS MBW **		
Max_R_LT (N)	584.82 ± 189.74	0.97	764.79 ± 176.24	1.27	33.81	0.00 *
Lat_R_LT (N)	34.36 ± 31.30	0.06	69.33 ± 86.72	0.12	10.07	0.00 *
Med_R_LT (N)	53.54 ± 61.73	0.09	159.64 ± 131.90	0.27	37.15	0.00 *
He_R_LT (N)	497.82 ± 155.80	0.83	535.82 ± 211.14	0.89	1.47	0.23
Max_L_RT (N)	564.44 ± 160.52	0.94	811.37 ± 124.28	1.35	103.57	0.00 *
Lat_L_RT (N)	38.62 ± 39.91	0.06	77.32 ± 93.98	0.13	10.05	0.00 *
Med_L_RT (N)	65.33 ± 69.87	0.11	161.33 ± 128.84	0.27	30.03	0.00 *
He_L_RT (N)	455.00 ± 130.74	0.76	554.47 ± 207.10	0.92	11.55	0.00 *

Legend: \*  $p < 0.05$ ; \*\*—MBW = pressure (kg)/mean body weight of all participants (kg); Max\_R\_LT—maximum force of the right foot in the left turn; Lat\_R\_LT—force of the lateral side of the right foot in the left turn; Med\_R\_LT—force of the medial side of the right foot in the left turn; He\_R\_LT—force of the right heel in the left turn; Max\_L\_RT—maximum force of the left foot in the right turn; Lat\_L\_RT—force of the lateral side of the left foot in the right turn; Med\_L\_RT—force of the medial side of the left foot in the right turn; He\_L\_RT—force of the left heel in the right turn; (N)—newtons.

Table 3 shows significant differences between short turn performance in IS and AS in seven out of eight measured kinetic variables. In the left turn, pressure force values of the outside foot for AS are higher than those for IS for all observed variables, with the values of the heel pressure force being the only that did not differ significantly ( $p = 0.23$ ). In relation to the MBW, the measured maximum force in AS was 1.27 MBW and in IS it was 0.97 MBW. Regarding the pressure distribution in both activities, the highest values of distribution were noted on the heel (IS—85.12% of maximal foot pressure; AS—70.06% of maximal foot pressure). The next highest proportions of pressure were located the on medial side of the foot (IS—9.15% of maximal foot pressure; AS—20.87% of maximal foot pressure). The

lowest pressure values in the left turn in both activities were measured on the lateral side of the foot (IS—5.88% of maximal foot pressure; AS—9.07% of maximal foot pressure).

Furthermore, the measured forces of the outside foot in the right turn were also higher in AS than those in IS (Figure 3). In relation to the MBW, the measured maximum forces in AS and IS were 1.35 MBW and 0.94 MBW, respectively. In both turns, i.e., in IS and AS, the biggest proportion of foot pressure was noticed on the heel. Concerning the pressure distribution in both IS and AS, the highest values were detected on the heel (IS—80.61% of maximal foot pressure; AS—68.34% of maximal foot pressure). The second-highest proportions of the overall pressure (IS—11.57%; AS—19.88%) were on the medial side of the foot. The lowest proportions of pressure were measured on lateral side of the foot (IS—6.84%; AS—9.53%).



**Figure 3.** (a) Maximal pressure forces on left (red line) and right (blue line) foot during AS, LT—left turn, RT—right turn; (b) maximal pressure forces on left (red line) and right (blue line) foot during IS, LT—left turn, RT—right turn.

#### 4. Discussion

The aim of this study was to objectively determine and compare the roles of the outside leg while performing short turns in IS and AS. The main findings of our study are defined similarities in pressure distribution during IS and AS and noticeable differences in

kinematic parameters of the outside leg between the mentioned activities. Those findings are not completely in accordance with previously stated hypotheses. Therefore, based on the results of this study, recreational alpine skier should be aware that IS cannot be used for the purpose of AS adoption but rather as a dry-land additional activity for preparation for AS.

There were statistically significant differences between IS and AS in almost all of the measured kinetic parameters ( $p < 0.01$ ). The highest values of maximal foot pressure were noticed in AS in both turns (Max\_R\_LT-AS—764.79 N, IS—584.82 N; Max\_L\_RT-AS—811.37 N, IS—564.44 N). Also, when comparing the ratio of maximum foot pressure and MBW, higher values were detected in AS (left turn, AS—1.27 MBW, IS—0.97 MBW; right turn, AS—1.35 MBW, IS—0.94 MBW). The mentioned results can be explained by some objective differences between the two activities. The steepness of the terrain directly affects the speed and gravitational force influencing the skier. Compared to IS, AS is performed usually on steeper slopes which consequently leads to higher speeds and higher centrifugal forces. Therefore, the skier must apply a higher foot pressure to master the speed, control the direction and overcome the high centrifugal force during a turn. As already stated, to successfully perform the turn on a steeper terrain, the skier must apply pressure predominantly on the outside leg [24]. When performing a short turn in IS, the terrain is less steep compared to that in AS, meaning the speed is not as high as it is in AS. The mentioned factors are directly connected to lower values of the applied foot pressure in the outside leg when performing short turns in IS. Furthermore, the friction between the equipment and the surface in the activities differs. It is easier to control and overcome skidding while performing turns on ski slopes by carving edges into the surface. This results in a more stable support, which also leads to higher values of the pressure force on the outside leg.

Similar results regarding differences in MBW and maximal foot pressure of the outside leg between IS and AS were found in a study conducted by Ropret [14]. Maximal values of foot pressure on the outside leg in AS were higher, ~1.5 MBW (in our study they were ~1.3 MBW). In IS, the measured foot pressure was 1.08 MBW (in our study it was ~0.97 MBW). The author concluded that the speed, turning radius and size of the centrifugal force is smaller in IS, which affects a reduced load on the outside leg when compared to AS.

Skis are much longer compared to inline skates, and the length and construction of the specific equipment influences the contact with the surface and differs between IS and AS. Those factors produce slightly different body positions and stances during turns. Despite the differences in equipment and maximal pressure force values, the distributions on foot regions were similar for IS and AS in both turns. The highest values of foot pressure were detected on the heel, followed by the medial side of the foot, and the lowest pressure was detected on the lateral side of the foot.

Dynamic balance is an important factor which has a great impact in AS on the turn performance [25,26]. The same can be stated for IS. Since, as mentioned, a skier adjusts their balance and applies the optimal pressure inside the ski boots and roller blades in the specific phase of the turn. The abovementioned factor is especially evident when both AS and IS are executed in a predefined corridor with the same gate distance and offset. Therefore, although the maximal pressure is higher in AS, the relative pressure and pressure distribution are quite similar.

In addition, the pattern of shifting the center of mass and maintaining a dynamic balance was similar when executing short parallel turns in observed activities. This suggests a positive transfer from developing dynamic balance on skates to applying it when executing turns in AS [27].

In research conducted by Kroll et al. [11], there were similarities but also differences in turn performance between skates and skis. The authors concluded that the main differences are the product of the different speeds of movement, turn radii and centrifugal forces. Furthermore, based on their results, the maximal foot pressure force in AS is on the outside leg while there is no significant difference between the foot pressures on the inside and the

outside leg when observing the turn performance in IS. Even though there are differences, the authors suggest using short parallel turns executed on skates as an additional way to develop AS technique.

Regarding kinematic parameters, even though the coordination of movements and the sequence of lower limb actions is very similar in AS and IS, there were statistically significant differences in all of the measured kinematic variables ( $p < 0.01$ ). Differences between the observed activities were similar in both turns. While performing a turn in AS, it was noticed that the side arc (hip abduction) was greater than that in IS (R\_HIP\_AB\_LT, AS—63.68°, IS—170.73°; L\_HIP\_AB\_RT, AS—163.00°, IS—170.51°). Moreover, the knee was more flexed during AS compared to IS (R\_KNEE\_LT, AS—157.43°, IS—139.56°; L\_KNEE\_RT, AS—140.30°, IS—159.93°). In addition, greater flexion was determined in the hip joint (R\_HIP\_F\_LT, AS—145.01°, IS—139.03°; L\_HIP\_F\_RT, AS—138.33°, IS—145.39°). Based on those results, it can be stated that while performing a short turn a skier's body is in a lower position with an emphasized side arc. These results are in line with those gathered from the kinetic analysis. Due to higher maximal forces on the outside leg, it is necessary to master the forces by gradually flexing the lower body segments and to adopt a body position with an emphasized hip abduction of the outside leg. By maintaining the mentioned side arc throughout the turn, it is possible to maintain the high centrifugal force without skidding and sliding.

Novel research focused on the kinematic analysis of recreational-level IS is scarce. Even more limited are studies concentrating on the specific implementation of IS as an additional activity for AS technique development.

One of the rare studies that was focused on a comparison of AS and IS was conducted by Kroll et al. [11]. The authors concluded that the angle of the body's leaning towards the center of the turn is smaller during IS compared to AS. This can be explained by differences in the contact area between roller skates with the terrain and skis with the terrain. As already mentioned, the contact is more lateral during IS, which leads to a smaller side arc that a person needs to take in order to avoid skidding. Our research also confirms the previous statement, as we observed greater hip abduction (side arc) during AS. Moreover, the kinematic results in our study also support this kinetic analysis. In addition, research conducted by Kroll et al. [28] found that the greater knee flexion enables the skier to generate a higher force and to transfer pressure to the new outside leg more efficiently. The steepness of the terrain affects the skier's body position, i.e., the side arc and the angles of lower body joints. Since AS is executed on a steeper terrain, the skier is in a lower position with a greater flexion of the knee and hip joint, which was confirmed in our study. Optimal flexion for a specific terrain and aligned lower body segments are a precondition for optimal pressure distribution. In our research, the pressure distribution was similar between AS and IS because of the similarity in body position that is necessary to execute a turn. However, the maximal pressure was higher in AS, meaning that a greater knee and hip flexion was required. Therefore, if the body position is optimal (in accordance with the demands of the terrain) the pressure distribution will be similar regardless of the maximal forces that occur.

Even though we determined some clear differences in kinematic aspects of the turn performance between IS and AS, based on our and some similar studies it can be concluded that the mechanism of turn execution is similar [17,29–31]. The skier has to maintain a lateral and frontal balance at the same time, which results in similar pressure distributions in IS and AS. Moreover, in both activities it is necessary to bring the center of mass low in relation to the inside of the turn radius. However, because of the specific conditions in which AS is performed, the skier has to be in a lower position in order to overcome higher foot pressure forces and master GRF when executing a turn. It is not possible to apply the same slope steepness and the corridor width when measuring IS and AS due to objective general differences between the mentioned activities. In the case of an identical slope steepness between IS and AS, the skier would not be able to control the speed during IS turns. This could cause compensatory movements and the executed turns



would evidently differ from turns executed on skis. Therefore, the measured values would not be valid for comparison. In addition, the injury risk would be significantly higher while executing IS on terrain that was as steep as the ski slope. Another factor that would disable the control of the speed, and which would lead to inability to execute the turn in an identically set corridor, is the difference in the friction of the wheels compared to the skis with asphalt and snow, respectively. Moreover, wheels are exactly below the center of the foot, unlike ski edges, which are lateral in relation to the foot. Therefore, the lever is bigger on the roller skates and, in order to make a turn in the exact same direction, a smaller movement of the leg is required when executing a turn in roller skates. Considering proportionate adjustments to the terrain steepness and the length of the skis and roller skates, a narrower width of the corridor would be more appropriate for IS than for AS measurement. In that case, the subject executing the turn would have to apply almost the same movement amplitude on roller skates as they would on skis to achieve the same length, duration, rhythm and speed of the turn. The steepness of the terrain would be proportionally adjusted in order for the subject to achieve the same above-mentioned turn features with the same amplitude of joint movements. All of the above explains the reasons for choosing an AS slope with an inclination of  $20^\circ$  and a slope for IS with an inclination of  $10^\circ$ . If the AS slope steepness had been set to the same angle as in the IS measurement ( $10^\circ$ ), turns would not have been executed optimally compared to the ski slope regarding duration, turn, rhythm and speed. The same can be stated vice versa for a higher inclination of the IS slope. The observed additional activity (IS) can be used in training to simulate the conditions of the main activity (AS) with consideration of the already mentioned differences regarding terrain and equipment.

When observing IS as an additional activity for AS competitors, IS is also performed on less steep slopes. In such a way, the speed and the direction can be controlled while the rhythm and the tempo are similar to those achieved during AS. In our research, the focus was on recreational skiers whose level of skiing technique was not as high as that of skiing competitors. From that standpoint, it is even more necessary to adapt the IS training to the conditions and environments that are appropriate for recreational skiers. This enables them to perform IS training by themselves on available slopes in parks or other environments. Regarding training and developing ski technique, based on the above mentioned it is recommended to use IS as an additional activity in the preparation period of the season. During that period, alpine skiers train simpler course settings and elements. Turns are basic left–right with the same gate distance and gate off-set, while the courses are usually set on a flat terrain. Therefore, inline skating with its already mentioned characteristics could be used as an extra technical training with similar goals. When discussing recreational level alpine skiing, IS can provide the greatest benefits when used in periods when recreational level alpine skiers are in the first phase of short turn adoption. During that phase of the learning process, it is usually executed on a less steep terrain. In that case the forces are smaller and it is more suitable for the learning process of a short turn. Information about the timing of specific body movements that ensure the optimal control of the direction and speed could be crucial for accomplishing the aim of alpine ski schools—to enable skiers to be independent in mastering complex skiing elements. The obtained similarities and differences in some of the observed parameters of AS and IS can contribute to the development of the teaching process of each skiing technique. Since the results indicate similarities in pressure distribution when comparing AS and IS, the implementation of IS can help in mastering the pressure transfer in each phase of the turn. In addition, the coordination of movements and the sequence of lower limb actions are very similar in both activities. However, it has to be emphasized that the final values of kinematic parameters are slightly different due to the terrain and equipment, so the execution of the turn cannot be the same. Considering the mentioned differences, it is important to include IS as an additional activity only when recreational skiers have mastered the basics of AS turn performance. Only then can these skiers recognize the real contribution of IS in the learning process of pressure transfer and distribution. If the above-mentioned condition is

not fulfilled, the skiers may aim to excessively simulate the movements during IS, which could maximize the potential injury risks and interfere with the learning process of AS. IS could also be used to master the rhythm for easier and smoother turn performance and for the timely performance of each movement, from the initial to the final phase of the turn. One of the problems that can be solved by using IS in the preparation period is oversteering. Oversteering can lead to a reduced control of the direction and an impaired dynamic balance, which can cause falls and injuries on the ski slope. IS helps in gaining a feel for steering, which prevents the skis turning faster than they do in their “natural” radius. Severe knee injuries can be caused if the skis turn faster due to an excessive edging angle that is not appropriate to the slope steepness and gate offset. Therefore, and as already mentioned, due to the different position of wheels in relation to the foot in IS, a bigger lever is created for the same knee movement compared to the AS. Hence, IS could be a useful tool in learning the optimal amplitude for executing turns and consequently prevent serious injuries.

### *Limitations*

The presented study was focused on the kinematic and kinetic analysis of the role of the outside leg when performing short parallel turns in AS and IS. In order to come to a clear conclusion whether it is justified to use IS as an additional activity for recreational alpine skiers, it is necessary to conduct an overall biomechanical analysis of the whole-body movement, including both the inside and outside leg and trunk movements. In addition, an electromyography analysis of the lower body’s muscle activity should be conducted to get an insight into the sequence of movements in each phase of the turn. Furthermore, to make an objective comparison of the two activities, it is necessary to completely replicate the conditions and the protocol in which the measurements are conducted. The corridor width and steepness of the slope differ due to general differences in the equipment and technical structure of the turn performance of each activity. Usually, training sessions in IS are performed in accordance with a temporal and geometrical extension of AS turns. However, due to abovementioned characteristics of both activities, the steepness of the terrain and corridor width cannot be exactly the same for both activities. In this way, the risk of potential injuries caused by an inadequate terrain steepness can be minimized.

### **5. Conclusions**

The results of the conducted kinematic and kinetic analysis of the role of the outside leg in AS and IS pointed to clear similarities in the ratio of pressure distribution while performing short parallel turns. Furthermore, one of the main findings is determined differences in the kinematic parameters of the outside leg between AS and IS. The kinematic analysis pointed to differences in the knee and hip angle of the outside leg, which has a role in controlling the speed and direction during the descent. The kinematic and kinetic analyses were in accordance with each other. The maximal pressure was higher during AS, and it was to be expected that greater knee and hip flexion and hip abduction were to be found in AS. On the other hand, the pressure distribution was similar in AS and IS, meaning that the outside leg had the role of maintaining a dynamic balance and stability during short turn execution in both activities.

Although the motor knowledge necessary for the adoption of AS and IS is similar, it is important to be aware that there are objective differences which need to be taken into consideration when using IS as an additional activity for recreational skiers. IS can be used for solving more complex problems of skiing technique and adopting some specific high-level motor skills. Therefore, we recommend IS as an additional activity for skiers who have developed at least a basic AS technique in order to avoid interference with the learning process.

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draft preparation, V.C., M.O. and I.B.; writing—review and editing, V.C., P.Š. and B.M.; supervision, P.Š. and B.M. All authors have read and agreed to the published version of the manuscript.

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

**Data Availability Statement:** The data presented in this study are available on request from the corresponding author. The data set is not publicly available due to its huge size and participants' privacy protection.

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Article

# The Sagittal Integral Morphotype in Male and Female Rowers

Jose Ramón Alvero-Cruz <sup>1</sup>, Fernando Santonja-Medina <sup>2,3,4,\*</sup>, Jose Manuel Sanz-Mengibar <sup>2,5</sup>  
and Pilar Sainz de Baranda <sup>2,6</sup>

- <sup>1</sup> Andalucía Tech, Faculty of Medicine, Campus de Teatinos, University of Málaga, 29071 Málaga, Spain; alvero@uma.es
  - <sup>2</sup> Sports and Musculoskeletal System Research Group (RAQUIS), University of Murcia, 30100 Murcia, Spain; jmsmengibar@hotmail.com (J.M.S.-M.); psainzdebaranda@um.es (P.S.d.B.)
  - <sup>3</sup> Department of Surgery, Pediatrics, Obstetrics and Gynecology, Faculty of Medicine, University of Murcia, 30100 Murcia, Spain
  - <sup>4</sup> Department of Orthopaedic Surgery and Traumatology, “Virgen de la Arrixaca” University Clinical Hospital, 30120 Murcia, Spain
  - <sup>5</sup> Centre for Neuromuscular Diseases, National Hospital for Neurology and Neurosurgery, University College London Hospitals NHS Foundation Trust, London NW1 2BU, UK
  - <sup>6</sup> Department of Physical Activity and Sport, Faculty of Sport Sciences, University of Murcia, 30100 Murcia, Spain
- \* Correspondence: santonja@um.es; Tel.: +34-868-88-7159

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**Abstract:** The goal of this study was to describe the integrated spinal assessment of the sagittal morphotype in rowers to determine whether the intense practice of rowing causes a modification of the sagittal curvatures of the spine, its relationship with the rowing technique, and training background. The second goal was to analyse how the dorsal and lumbar curves behave in the three phases of the rowing gesture, and to determine which phases can be detrimental to the correct development of the spine during growth. We analysed the spine curvatures in the sagittal plane of 29 females and 82 males, which were measured with an inclinometer in standing, slump sitting, maximal trunk flexion and during rowing performance. The average value of thoracic kyphosis in the rowers was 30° (mean, 30 + 8.27°). Thoracic hyperkyphosis was found in only two rowers (1.8%). Lumbar lordosis was within normal range in 84.1% of the males (mean, 27 + 9.57°) and 75.9% of female rowers (mean, 33°). Functional thoracic hyperkyphosis was observed in 57.4% of the males and 17.1% of the females. Functional lumbar hyperkyphosis was observed in 28 of the 69 males (40.5%) and five of 22 females (17.2%). Rowing seems to provide adequate spine alignment in the sagittal plane on standing. The integrated spinal assessment of the sagittal morphotype showed that half of our rowers presented with functional thoracic hyperkyphosis, and 43.2% presented with functional lumbar hyperkyphosis. Spine behaviour during the rowing technique shows that the thoracic kyphosis (98.2%) and lumbar spine (91%) perform within normative ranges and could explain the adequate positioning of the spine in the sagittal plane on standing. Years of rowing training tend to reduce thoracic kyphosis in males.

**Keywords:** anatomy; spine; thoracic spine; low back; lumbar spine; biomechanics; rowing

## 1. Introduction

Research shows that the spine seems to adapt to the biomechanical requirements of different sports [1–3], primarily as an engine for the execution of the specific skill, and consequently producing structural changes. Specific training improves performance and may modify the spine curvature according to its intensity [4]. Research has described spinal profiles in artistic [3] and trampoline gymnasts [4], swimmers [5–7], climbers [8], tennis players [9], bodybuilders [6,10], skiers [11], dancers [12], and also, riders [13]. The sagittal curvatures of the spine have also been studied in team sports, such as football,

rugby [6], hockey players [1], volleyball, basketball, and handball [14]. This adaptative mechanism and its potential consequences [15] have also been studied in rowers, including pain [16–19], muscle dysfunction due to fatigue [20], and stresses on the intervertebral joints and discs [21]. The wedging of immature vertebral bodies is another potential consequence in high-performance athletes and swimmers [7,22,23] due to repetitive trunk flexion movements [3,24]. The combination of high forces acting on the rower and high training volume put rowers at risk for injury [16]. Kinematics and biomechanical measurements show posterior pelvic tilt and lumbar spine flexion at the catch position [25,26], and fatigue seems to increase this range of motion of the spine [27,28]. Greater bone mineral density [29] has also been used to quantify the mechanical loading produced during rowing.

Research on the sagittal integral morphotype (SIM) in male and female rowers has not been found to exist, neither about the relationship between postural adaptations nor their years of training. At present, thoracic and lumbar kyphosis during rowing technique have not been related to the impact of a standardised clinical spine assessment. The specific impact of this sport on spine alignment requires an assessment that allow comparisons with other disciplines [1,3,4,10,12,13,30–33].

The movement of the lumbar spine in the sagittal plane during the rowing technique has been previously studied [25]. In contrast, the postural adaptation of the spine as a result of rowing training has not been described. Lower back dysfunction due to fatigue [20,27] and stress on the intervertebral joints and discs [21] described in rowers may have an impact on the basal positioning of the spine in the sagittal plane. Sagittal spinal curvatures play an important role in health, and therefore, both clinical assessment and research in this topic seems to be relevant, as in other sports [1,3–7,10,13,30–33].

The “sagittal integral morphotype”, according to Santonja [34], allows a more comprehensive assessment of the thoracic and lumbar curvatures. The curvatures are assessed according to vertebral disposition by three different tests [34–36]. This noninvasive assessment of the sagittal curvatures with the inclinometer in standing position, slump sitting, and maximal trunk flexion provides good reproducibility, reliability and correlation with radiographic measurements [37,38]. Pastor [7] compared the diagnosis of thoracic hyperkyphosis, made with an inclinometer, to radiographic findings in young swimmers. A sensitivity of 81.6% and specificity of 100% was found, with an ICC of 0.86 in male swimmers and 0.84 in females.

The integrated spinal assessment of the sagittal morphotype has proven to be useful in the assessment of the normal status of the spine of different sportsmen [1,3,4,10,12,13,30–33,35], as well as scholars [35,36,39–41] and adolescents [42]. The advantage of this standardised assessment is the possible comparison of the spinal adaptations among different sport techniques. As previously described, many other sport techniques with no similarities with these tests showed compensatory strategies of the spine [1,3–5,13] to cope with its biomechanical requirements. The sagittal curves of the spine tend to increase with time, especially in adolescence [7,35,36,39–42].

The goals of this study were to describe the integrated spinal assessment of the sagittal morphotype in male and female rowers, as well as its relationship with each sport technique, years of training and other disciplines. Our hypothesis was that there is a specific adaptation of the spine to the biomechanical requirements of rowing, and this spinal adaptation depends on the years of rowing training. In addition, there exist different adaptations of the integrated spinal assessment of the sagittal morphotype between male and female rowers.

## 2. Materials and Methods

### 2.1. Study Design and Approvals

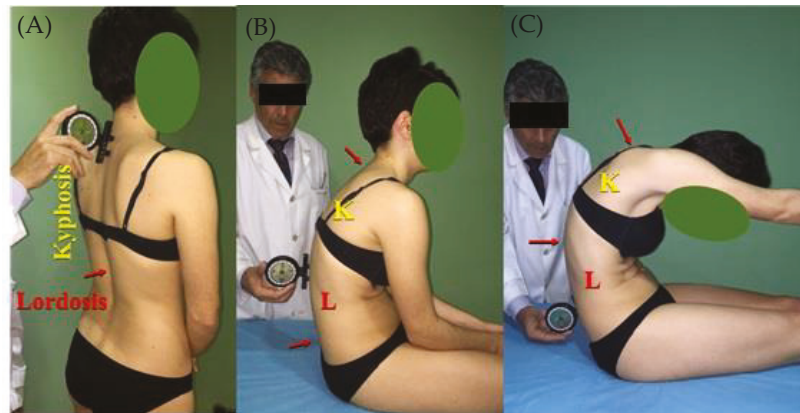
A cross-sectional analysis was designed to confirm or rule out our hypothesis, describing and correlating the spinal measurements with the years of training of male and female rowers. The journal’s ethical standards were met [43], and this study was approved by the Ethics and Scientific Committee of the University of Murcia (Spain) [ID: 1702/2017].

## 2.2. Participants

Participants were recruited from four different clubs associated with rowing federation in the southern region of Spain. Rowers, parents, and coaches were informed of the study procedures before the assessments were performed and all study participants provided written informed consents. Inclusion criteria were: belonging to a male or female rowers' club with membership in a rowing federation, participating in regional or national competitions, and having trained 18 h per week or more during the last six months [3]. Exclusion criteria were the presence of scoliosis ( $>20^\circ$  Cobb angle) or any spine deformity on the sagittal plane that required orthopedic treatment [3]. The final sample included 111 rowers; 29 females and 82 males. The mean age ( $\pm$ standard deviation) was  $17.43 \pm 3.25$  years (men: minimum 14, maximum 35, average 17.2; women: minimum 14, maximum 26, average 17.8) and the mean years of training was  $4.98 \pm 3.77$  (men: minimum 0.5, maximum 20, average 4.72; women: minimum 0.5, maximum 15, 5.73 average).

## 2.3. Measurements

Integrated spinal assessments of the sagittal morphotype were quantified in all participants [1,3] while wearing undergarments and barefoot. Thoracic and lumbar spine curvatures in the sagittal plane were measured in standing position, slump sitting (SS), and maximal trunk flexion (MTF), according to the assessment protocol defined by Santonja [34]. Measurements were performed by an orthopaedic consultant with 15 years of expertise, and athletes were assessed and data from every rower were obtained during the same session. Quantification of the sagittal curvatures of the spine was carried out with a Unilevel inclinometer (ISOMED, Inc., Portland, OR, USA). Upon standing, the inclinometer was placed at T1 to begin with, and then slid down to the end of the thoracic kyphosis, where the greatest value was observed. After resetting the inclinometer, the lumbar curve was quantified using the same procedure at this vertebral level. In order to quantify the thoracic and lumbar curves in SS and MTF, the inclinometer was placed at T1–T2, T12–L1 and L5–S1 [34–36]. Measurements were repeated twice to guarantee reliability (Figure 1).



**Figure 1.** Sagittal integral morphotype of the thoracic and lumbar spine (A) Standing assessment: the inclinometer is placed at the beginning of the thoracic kyphosis before zeroing, and it is then slid down to the greatest curve angle is observed. Once the inclinometer is zeroed, the lumbar curve will be quantified with the same procedure. (B) Slump sitting quantification: cranial arrow points to the initial placement; the inclinometer was zeroed at T1–T2, before sliding it down to T12–L1 to quantify the thoracic curve. The inclinometer was zeroed again at T12–L1 and slid down to L5–S1 to quantify the lumbar curve. (C) Assessment in maximal trunk flexion: both arrows show the thoracic kyphosis limits. K = thoracic kyphosis; L = lumbar curve.



The reliability of the researcher who carried out the measurements of the SIM was reflected as ICC: kyphosis in standing = 0.98; lordosis in standing = 0.94; kyphosis in sitting = 0.96; lumbar in sitting = 0.97; kyphosis in flexion = 0.87 and lumbar flexion = 0.97.

Normal values are considered when the measurements are within normative ranges in standing position, SS and MTF tests [34,35] and are summarised in Table 1. If the thoracic kyphosis was greater than 45°, the measurements in self-correction of the kyphosis were also quantified. “Postural hyperkyphosis” was considered when the hyperkyphosis was reduced to ≤30° during the self-correction tests [34,44].

**Table 1.** Classification of the sagittal thoracic and spine curvatures according to the normative data [34,35].

	Sagittal Morphotype	Thoracic Curvature	Lumbar Curvature
Standing	Normal range	20–45°	(–)20–40°
	Hyperkyphosis/hyperlordosis	>45°	>(–)40°
Maximal trunk flexion in sit and reach test	Normal range kyphosis	40–65°	10–30°
	Thoracic hyperkyphosis	>65°	
	Functional lumbar hyperkyphosis		>0°
Slump sitting	Normal range kyphosis	20–45°	±0–20°
	Thoracic hyperkyphosis/ Functional lumbar hyperkyphosis	>5°	>0°

The rowing stroke may be divided into drive and recovery phases. The drive phase includes the ‘catch’ when the oar enters the water, and the lumbar spine is fully flexed at this point of main work [20]. At the end of the drive phase, the oar is withdrawn, switching the lumbar spine to a relatively extended position. The ‘finish’ phase is a mid-way position during the drive and extension phases. Thoracic and lumbar spine measurements were also taken during the sport technique performance on the ergometer and these measurements were obtained during training against the usual resistance used with the ergometer. The measurements were at: (a) ‘catch’: maximal flexion of hip and trunk with the seat placed forward; (b) ‘finish’: participants sat upright on the seat, placed at full knee extension and elbows flexed towards the chest; (c) ‘extension’ position: maximal extension of the spine at the end of the stroke [20,27]. The inclinometer was placed and zeroed at T1–T2, before sliding it down to T12–L1 to quantify the thoracic curve. The inclinometer was zeroed again at T12–L1 and slid down to L5–S1 to quantify the lumbar curve. This procedure was carried out in the three positions in order to quantify both curves (Figure 2) [1,3,5,10,12,30,35,44].

Age, gender, and year of training of each rower were also noted in order to correlate these factors to the sagittal plane spine measurements.

#### 2.4. Statistical Analysis

All statistical analyses were carried out using MedCalc Statistical Software version 19.0.3 (MedCalc Software bvba, Ostend, Belgium). Initially, an exploratory analysis of the data was carried out, in which central tendency (median and mean), dispersion (95% confidence interval [CI] and standard deviation). Measures were calculated after performance of Shapiro–Wilk’s test to verify normality. The percentages of rowers with spine curvatures outside the normative values were also calculated. An independent samples t-test or Mann–Whitney U test examined differences between genders. The correlations among the variables were examined using the Spearman’s rank correlation coefficients (rho). The magnitude of the correlations was evaluated as trivial ( $r < 0.10$ ), small ( $0.10 \leq r < 0.30$ ), moderate ( $0.30 \leq r < 0.50$ ), large ( $0.50 \leq r < 0.70$ ), very large ( $0.70 \leq r < 0.90$ ), and perfect ( $r \geq 0.90$ ) [16]. The acceptable type I error was set at  $p < 0.05$  [45].



**Figure 2.** Rowing technique phases. Thoracic and lumbar sagittal curves were measured during the three phases of rowing: catch (**top**), finish (**middle**) and extension (**bottom**). Thoracic kyphosis was quantified placing the inclinometer at T1–T12 and lumbar spine at T12–L5.

### 3. Results

The demographic features of the participants, significant differences between male and female rowers, and the average descriptive values of the sagittal curves can be found in Table 2. The independent t-test or Mann-Whitney U test showed no differences in age ( $p = 0.35$ ) or years of training ( $p = 0.49$ ) between the genders. The female participants showed greater lumbar lordotic angles than the males in standing position ( $p = 0.0008$ ), but lower angles of lumbar kyphosis in SS ( $p = 0.028$ ), and MTF ( $p = 0.0026$ ) than the males. Thoracic kyphosis values were similar in standing position between the genders, but lower values in women were observed in SS ( $p = 0.003$ ), and MTF ( $p = 0.045$ ).

**Table 2.** Descriptive values: demographic data of the participants and sagittal spine curvatures (significant differences according to gender).

Variables	Men (n = 82)			Women (n = 29)			p-Value
	Mean $\pm$ SD	Median	95% CI	Mean $\pm$ SD	Median	95% CI	
<b>Demographics</b>							
Age (years)	17.28 $\pm$ 3.23	16	16.0–17.0	17.86 $\pm$ 3.34	16.5	16–18.0	0.35
Training (years)	4.72 $\pm$ 3.42	4	3.0–5.0	5.73 $\pm$ 4.64	4	3–6.6	0.49
<b>Standing position</b>							
Thoracic ( $^{\circ}$ )	30.21 $\pm$ 8.27	30	29.3–33	30.62 $\pm$ 9.03	32	26–35.0	0.82
Lumbar ( $^{\circ}$ )	27.01 $\pm$ 9.57	26	25.0–28.0	33.14 $\pm$ 9.13	34	30–37.2	0.0008
<b>Slump sitting</b>							
Thoracic ( $^{\circ}$ )	47.83 $\pm$ 50	10.58	45.6–50.7	39.52 $\pm$ 9.48	39	35–45.2	0.003
Lumbar ( $^{\circ}$ )	20.13 $\pm$ 9.04	20	16.9–23.0	14.03 $\pm$ 11.5	12	7.3–15.0	0.028
<b>Maximal trunk flexion</b>							
Thoracic ( $^{\circ}$ )	63.7 $\pm$ 9.76	65	61.6–65.0	59.38 $\pm$ 10.2	59	55–65.0	0.045
Lumbar ( $^{\circ}$ )	29.35 $\pm$ 10.6	26	25.0–30.0	24.31 $\pm$ 10.7	22	18–26.0	0.026

Table 2. Cont.

Variables	Men (n = 82)			Women (n = 29)			p-Value
	Mean ± SD	Median	95% CI	Mean ± SD	Median	95% CI	
<b>Ergometre position</b>							
Thoracic catch (°)	34 ± 12.5	31	30.0–35.0	28.31 ± 13.5	25	21–30.2	0.026
Thoracic finish (°)	37.05 ± 11.3	35.5	34.0–40.0	30.07 ± 12.2	26	24.7–35.2	0.0086
Thoracic extension (°)	35.28 ± 11.2	35	30–38.02	36.55 ± 13.3	35	26–42.0	0.86
Lumbar catch (°)	22.24 ± 8.52	22	20.0–25.0	15.45 ± 5.93	16	11.7–20	<0.0001
Lumbar finish (°)	17.67 ± 6.72	18	16.0–20.0	11.79 ± 6.66	12	8.0–15.0	0.0001
Lumbar extension (°)	7.64 ± 8.77	7	5.0–10.0	−5.55 ± 11.1	−5	−8.0–0.0	<0.0001

Abbreviations: SD: standard deviation; CI: confidence interval.

The sport technique analysis showed that the values of thoracic kyphosis during the three phases of rowing (95% CI at ‘catch’, ‘finish’ and ‘extension’) were within the normal range (Table 2) in trunk flexion ( $\leq 65^\circ$ ), and were reduced in females during ‘catch’ ( $p = 0.0026$ ) and ‘finish’ ( $p = 0.008$ ). Thoracic extension values are similar in the male and female participants. Only one rower reached  $74^\circ$  during the ‘catch’ phase and  $66^\circ$  during the ‘extension’ phase. Another rower reached  $68^\circ$  during the ‘extension’ phase, but the rest of the participants showed thoracic kyphosis curves within the normal values.

The 95% CI of the lumbar curve angles were within normal ranges during trunk flexion ( $10\text{--}30^\circ$ ) and were significantly lower in women than in men during the three phases of rowing ( $p = 0.0001$ ). Ten rowers exceeded the normative  $30^\circ$  during the ‘catch’ phase (9%), and a lordotic curve was observed in 25 rowers during the ‘extension’ phase. Table 3 summarises the number of cases according to the classical diagnosis in the standing position of the thoracic and lumbar spines, the sagittal morphotype of the spine integrating the three tests (in the last column of the table), and the percentage of rowers whose measurements were outside the normative values. In total, 91.5% of the males and 86.2% of the females possessed thoracic kyphosis in standing position within the normative values ( $20\text{--}45^\circ$ ). Thoracic hyperkyphosis ( $>45^\circ$ ) was observed in only one male ( $50^\circ$ ) and one female rower ( $55^\circ$ ), but both showed a curve reduction to  $<30^\circ$  during the self-correction test [34,44] (postural hyperkyphosis). Of the male participants, 84.1%, and 75.9% of the females showed lumbar lordosis within the normative ranges. Only two males and six female rowers had hyperlordosis ( $>40^\circ$ ) and all of them presented with postural hyperlordosis (lumbar curve in flexion with kyphosis of  $10\text{--}30^\circ$ ) [34,35].

Regarding the integrative sagittal morphotype, 57.4% of the male rowers had “functional thoracic hyperkyphosis” (normal range kyphosis in standing position with adopted hyperkyphotic attitude during SS and/or MTF; Figure 3), and this finding was three times less frequent in females (17.1%). Twenty-eight of 69 male rowers (40.5%) and five of 22 female rowers (17.2%) with normal lordosis in standing were diagnosed with functional lumbar hyperkyphosis (lumbar kyphotic posture was quantified  $>30^\circ$  in the MTF and/or  $>20^\circ$  SS tests) (Figure 4).

Correlation values between the studied variables can be found in Table 4 and Figure 5. A weak correlation between thoracic kyphosis and years of training has been observed in the male rowers ( $\rho = -0.258$ ;  $p = 0.02$ ), but it was not significant in the female rowers. Lumbar lordosis in standing position in the male rowers tended to reduce with age ( $\rho = -0.302$ ,  $p < 0.01$ ). Thoracic kyphosis during the three phases of rowing performance tended to reduce with age in the males (‘catch’:  $\rho = -0.419$ ,  $p = 0.0002$ ; ‘finish’:  $\rho = -0.229$ ,  $p = 0.04$ ; ‘extension’:  $\rho = -0.337$ ,  $p = 0.0031$ ); and in the ‘finish’ ( $\rho = -0.385$ ;  $p = 0.04$ ) and ‘extension’ ( $\rho = -0.561$ ;  $p = 0.0019$ ) phases in the female rowers.

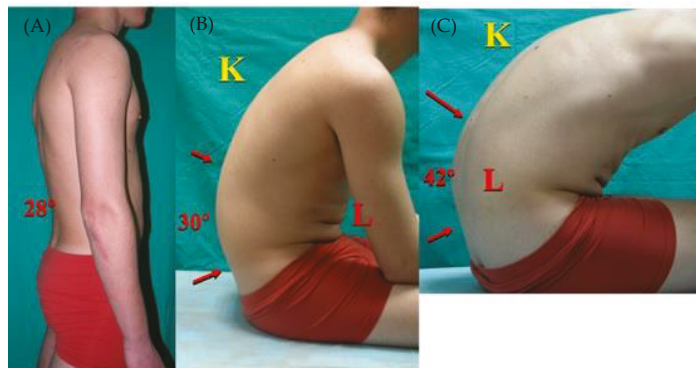
**Table 3.** Distribution according to the classical thoracic and lumbar morphotypes in standing position and according to the integrated spinal assessment of the sagittal integral morphotype [34,35].

CATEGORY	SUBCATEGORY	Standing	Slump Sitting	Maximal Trunk Flexion	Male		Female	
					n = 82	%	n = 29	%
<b>CATEGORY Thoracic Morphotype</b>					<b>Thoracic Morphotype</b>			
Hypokyphosis or hypokyphotic attitude	Standing	Hypokyphosis (<20°)	Normal (20–45°)	Normal (40–65°)	6 (2 > 45° SS)	7.3	3	10.3
Hypomobile kyphosis		Normal (20–45°)	Normal (20–45°)	Hypokyphosis (<40°)	0		0	
Normal kyphosis		Normal (20–45°)	Normal (20–45°)	Normal (40–65°)	28	34.1	20	68.9
Hyperkyphosis	Total	Hyperkyphosis (>45°)	Hyperkyphosis (>45°)	Hyperkyphosis (>65°)	1	1.2	0	
	Standing	Hyperkyphosis (>45°)	Normal (20–40°)	Normal (40–65°)	0		0	
	Static	Hyperkyphosis (>45°)	Hyperkyphosis (>45°)	Normal (40–65°)	0		1	3.4
	Dynamic	Hyperkyphosis (>45°)	Normal (20–40°)	Hyperkyphosis (>65°)	0		0	
Functional thoracic hyperkyphosis	Static	Normal (20–45°)	Hyperkyphosis (>45°)	Normal (40–65°)	40	48.8	4	13.7
	Dynamic	Normal (20–45°)	Normal (20–40°)	Hyperkyphosis (>65°)	3	3.7	1	3.4
	Total	Normal (20–45°)	Hyperkyphosis (>45°)	Hyperkyphosis (>65°)	4	4.9	0	
<b>CATEGORY Lumbar morphotype</b>					<b>Lumbar morphotype</b>			
Hypolordosis	Lumbar hypomobility	Hypolordotic attitude (<20°)	Normal (0 ± 20°)	Normal (10–30°)	1	1.2	0	
Normal lordosis		Normal (20–40°)	Normal (0 ± 20°)	Normal (10–30°)	41	50	17	58.6
Functional lumbar hyperkyphosis	Static	Normal (20–40°)	Hyperkyphosis (>20°)	Normal (10–30°)	9	11	0	
	Dynamic	Normal (20–40°)	Normal (0 ± 20°)	Hyperkyphosis (>30°)	3	3.6	0	
	Total	Normal (20–40°)	Hyperkyphosis (>20°)	Hyperkyphosis (10–30°)	16	19.5	5	17.2
Hyperlordosis	Postural or attitude	>40°	Normal (0 ± 20°)	Normal (10–30°)	2	2.4	6	20.7
	Structural	> 40°	Normal (0 ± 20°) or lordotic (<−20°)	Hypokyphosis (<10°)	0		0	
Lumbar hypermobility		Hyperlordosis (>40°)	Normal (0±20°) or hyperkyphosis (>20°)	Normal (10–30°) or hyperkyphosis (>30°)	1	1.2	0	
Lumbar kyphosis		Hypolordosis or kyphosis (< 20°)	Hyperkyphosis (>20°)	Hyperkyphosis (>30°)	9	11	1	3.4
Structural lumbar kyphosis		Lumbar kyphosis	Hyperkyphosis (>20°)	Hyperkyphosis (>30°)	0		0	

Note: comparisons according to the classical thoracic and lumbar spine morphotypes in standing position only and the integrative assessment of the sagittal morphotype of the spine (standing, slump sitting and maximal trunk flexion) in male and female rowers.



**Figure 3.** Functional thoracic hyperkyphosis. Normal thoracic kyphosis ( $30^\circ$ ) in standing was observed in the rower displayed (normal range  $20\text{--}45^\circ$ ). In maximal trunk flexion,  $80^\circ$  of thoracic kyphosis was quantified, resulting in a functional thoracic hyperkyphosis.



**Figure 4.** Sportsman with functional lumbar hyperkyphosis (A) lumbar lordosis is quantified with the normal range in standing ( $28^\circ$ ); however, (B) lumbar kyphosis is increased in slump sitting ( $30^\circ$ ), (C) as well as in maximal trunk flexion ( $42^\circ$ ). K = thoracic kyphosis, L = lumbar curve.

**Table 4.** Correlations between the sagittal spine curvature values and years of training, age and training technique.

Variable	Men		Women	
	Training Years	Age	Training Years	Age
<b>Standing position</b>				
Thoracic spine	−0.258 *	0.021	0.018	0.033
Lumbar spine	−0.204	−0.302 **	−0.161	−0.045
<b>Slump sitting</b>				
Thoracic spine	−0.069	−0.038	−0.031	0.086
Lumbar spine	0.097	−0.034	0.304	0.15
<b>Maximal trunk flexion</b>				
Thoracic spine	−0.249 *	0.135	−0.203	−0.293
Lumbar spine	0.115	−0.031	0.041	−0.266
<b>Ergometre position</b>				
Thoracic catch	−0.419 **	−0.216	−0.261	−0.173
Thoracic finish	−0.229 *	0.053	−0.385 *	−0.24
Thoracic extension	−0.337 **	0.083	−0.561 **	−0.309
Lumbar catch	−0.026	−0.168	0.104	0
Lumbar finish	0.056	0.008	−0.237	−0.118
Lumbar extension	0.212	−0.009	0.503 **	0.047

Note: \*  $p < 0.05$ , \*\*  $p < 0.01$ .

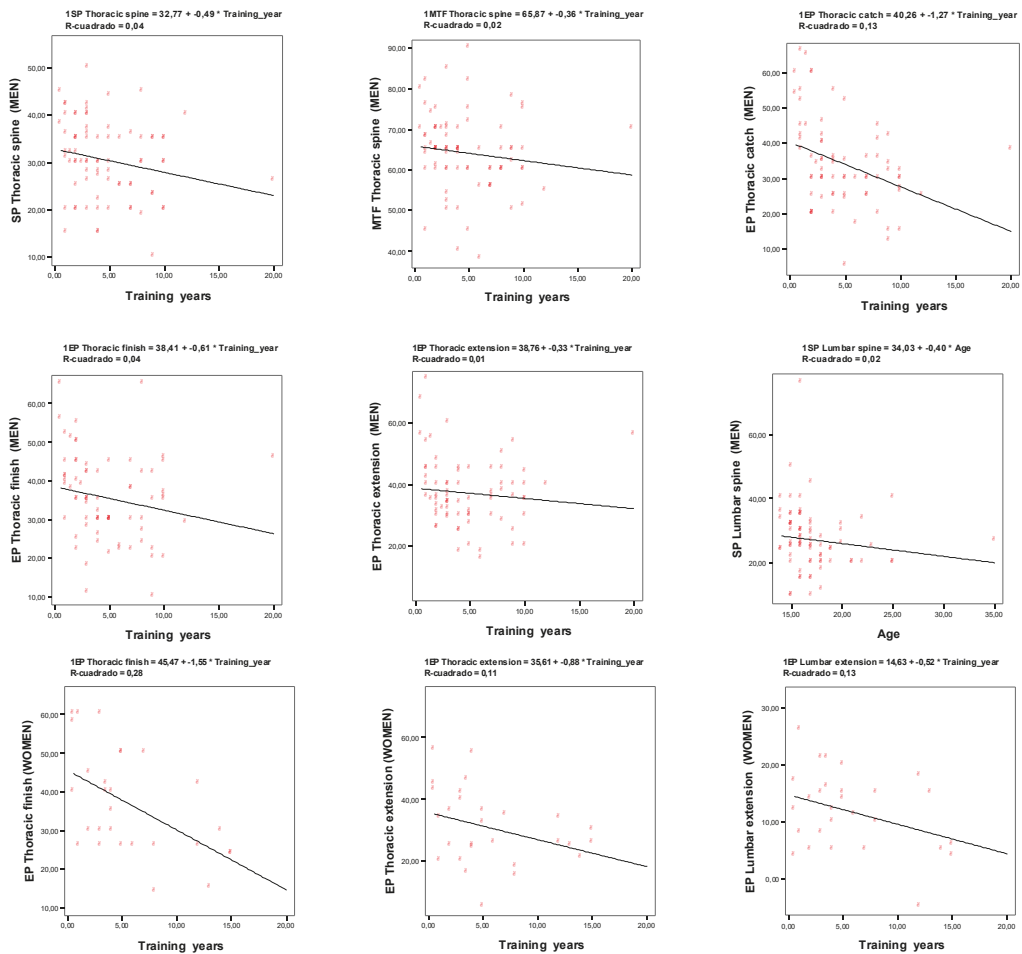


Figure 5. Correlations significative between the sagittal spine curvature values and years of training, age and training technique.

#### 4. Discussion

This research is the first work describing the effect of rowing training on thoracic kyphosis and lumbar lordosis in standing, as well as the effect of rowing training on the SIM of the spine in this sport. Our main relevant findings showed how the kyphosis and lordosis adapt to the rowing technique training. A high percentage of functional thoracic hyperkyphosis was observed (57.4% in the male rowers; three times less in women [17.1%]). Functional lumbar hyperkyphosis was observed in 34.1% of the male rowers, while only half of the female rowers showed this lumbar spine adaptation (17.2%). The female gender seemed to play a protective role for the dynamic adaptation of the spine. Also, our research compared the behaviour of the thoracic and lumbar segments during the three phases of rowing, and our results showed that the thoracic and lumbar spine performed within normal ranges. This could be useful to understand the relationship between back pain and spine adaptations, as well as the need for preventive programmes in this sport.

Low frequencies of thoracic hyperkyphosis in male (1.2%) and female (3.4%) rowers were observed in our study and all cases of thoracic hyperkyphosis were flexible when assessed with self-correction tests [34,44]. This adequate positioning of the thoracic spine

in the standing position suggests that rowing is a protective sport against thoracic hyperkyphosis in men and in women. In contrast, 51.8% of hyperkyphosis has been reported in high-level swimmers younger than 15 years old [5,7]. A low frequency of lumbar hyperlordosis was also observed in the male participants (3.7%), indicating that rowing may be beneficial in men with lumbar hyperlordosis.

#### 4.1. Thoracic Kyphosis

The average value of thoracic kyphosis in standing position was lower in rowers (30°) than in other sports (Table 5). This value was lower than those of artistic [3] and trampoline gymnasts [4], climbers [8], swimmers [5], weightlifters [10], tennis players [31], and hockey players [1]. These values are even more favourable than those of scholars [35,39–41] adolescents [42], and non-athletic adults (controls) [12,46]. Thoracic kyphosis of rowers in standing position showed similar values to rhythmic [46] and aesthetic group gymnasts [47]. Only dancers [12,48] showed lower values of thoracic kyphosis in standing, probably due to the posture work and body scheme required in this discipline. No significant differences between genders in this measurement was uncommon.

#### 4.2. Lumbar Spine

The average values of lumbar lordosis in standing were reduced in rowers (males, 27°; females, 33.1°) (Table 2), in comparison to trampoline (40.3°) [4] and beginner rhythmic gymnasts (40.3°) [46], ice-hockey players (38.1°) [22], and swimmers (36.3°, and even 50.9° when measured on radiographs) [7]. Our average values were similar to previous studies in hockey [1], aesthetics gymnasts [47], tennis players [31], kayakers [49], weightlifters [10], swimmers [5] and non-athletic adults [12]. It is important to note that the lumbar hyperlordosis was six times more frequent in females than in males (20.7% vs 3.7%) (Table 5). In contrast, lower values of lumbar hyperlordosis in hockey players (1.4%) [1], show jumping riders (5%) [13], and scholars (9%) [41] have been observed. All cases of lumbar hyperlordosis found in rowers were non-structural.

#### 4.3. Sagittal Integral Morphotype

The SIM [34] included the quantification of the curves in at least three different positions (standing, SS and MTF), and implies the definition of new diagnostic concepts.

#### 4.4. Thoracic Morphotype

It is important to note that 46.8% of our rowers (57.5% males, 27.6% females) had normal thoracic spines in standing, but increased position during the flexion and/or sitting tests. This was described as functional thoracic kyphosis by Bado [50]. These percentages were higher than in hockey players (18.9%) [1] and aesthetic gymnasts (25.4%) [47], but lower in artistic gymnasts (men, 65.2%; women, 75%) [3]. The percentages of functional kyphosis in scholars and adolescents without intense sport training were lower, as observed in previous research: 29.7% [42], 36.8% [35], 38.5% [51], and 43.2% [39].

The diagnosis of functional thoracic hyperkyphosis is clinically relevant during pubertal growth spurs because curves could become structural, according to Bado [50]. In our rowers, the functional thoracic hyperkyphosis due only to hyperkyphosis in SS represented 84.6% of total functional thoracic hyperkyphosis in rowers, suggesting bad postural hygiene in sitting. In contrast, the functional thoracic hyperkyphosis due to hyperkyphosis in MTF (15.4%) could denote an adaptation to the sport performance. The wedging of immature vertebral bodies is another potential consequence in high-performance rowers, as observed in other young athletes [7,22,23], due to repetitive trunk flexion movements [24].

If the sagittal spinal assessment would have been carried out in standing position only, the majority of the rowers would have shown thoracic and lumbar curves within the normative values. This supports the integrated spinal assessment of the sagittal morphotype as a more specific screening tool.

**Table 5.** Summary of research order by year of publication, including average values of thoracic kyphosis in standing, sitting and maximal flexion of the trunk during the sit and reach test.

Group	Thoracic Spine					Thoracic Morphotype					Lumbar Spine					Lumbar Morphotype			Demographics	
	SP	SSP	MTF	↓	Normal	↑	FTH	SP	SSP	MTF	↓	Normal	↑	FLK	Age, Mean or Range (Years)	n				
Rowers	Male	30.2°	47.8°	63.7°	7.5%	91.3%	1.2%	57.5%	27°	20.1°	29.3°	12.5%	83.8%	3.7%	46.3%	17.2 (14–35)	82			
	Female	30.6°	39.5°	59.3°	10.3%	86.2%	3.4%	27.6%	33.1°	14°	24.3°	3.4%	75.9%	20.7%	20.7%	17.8 (14–26)	29			
Scholars [36]	Both genders				2.2%	70.4%	27.4%	36.8%				1.9%	89.1%	9%	82.3%	8–12	731			
In-line hockey [1]	Both genders	38.5°	45°	53.7	1.4%	60.8%	37.8%	18.9%	28.7°	28.7°	31.5°	9.5%	89.2%	1.4%	66.1%	8–15	74			
Dressage riders [13]	Both genders	39.2°	34.9°	50.7°	0	61.5%	38.5%	23.10%	40.4°	10°	27.4°	0	46.10%	53.9%	38.50%	9–17	13			
Show jumping riders [13]	Both genders	43.8°	44.4°	54.2°	0	50%	50%	40%	43.2°	15.4°	27°	0	50%	50%	40%	9–17	10			
Artistic gymnasts [3]	Male	39.6°	26°	62.9°	0%	73.9%	26%	65.2%	27.7°	15.5°	26°	4.3%	78.2%	17.3%	13%	8–30	24			
	Female	31.8°	49.3°	61.4°	8.3%	87.5%	4.16%	75%	30.5°	15.7°	27.7°	0%	83.3%	16.6%	29%		24			
Scholars [40]	Both genders	35.7°	41.9°	53.9°		71.3%	28.7%	-	32.9°	24.4°	33.4°		73.6%	26.4%	-	8–13	688			
Skiers [11]	Both genders	41.2°							33.4°							16–19	51			
Aesthetic group gymnastics [47]	Female	29.3°	47.9°	69.1°	22.3%	67%	9.6%	25.4%	32.9°	15.9°	26.4°	6.4%	77.7%	16%	-	10–18	94			
	Male	36.8°	43.7°	55.4°	2.3%	70.2%	27.4%	-	30.9°	26.4°	33.1°	1.9%	89.1%	9%	-	10–18	741			
Ballet dancers [48]	Male	18.5°	6.3°	42.6°	48.6%	51.3%	0%	-	24.7°	1.7°	34.5°	23.7%	75%	1.3%	-	13.2	76			
	Female	43.8°	36.1°		0%	37.5%	62.5%		27.5°	32.6°		4.2%	83.35	12.5%	-	13–18	40			
Scholars [51]	Male	35.5°	43.1°	64.8°	0%	76.5%	23.5%	38.5%	33.9°	9.2°	19.5°	0%	88.2%	11.8%	20.5–23.9%	11–12	39			
	Female	37.5°	49°	68.8°					32.4°	8.7°	16.8°					11–12	46			
Teenagers [42]	Male	37.6/47°	43/55.1°	66/80.7°	0%	44.5%	54.5%	29.7%	29/35.7°	7.3/12°	16.6/23°	1.2%	90.5%	8.3%	26.2%	13–18	119			
	Female	35/42.5°	37.2/43°	64/73.3°	2.6%	68.6%	29%		34/40.3°	5.8/10°	16.6–18°	3.5%	65.7%	30.8%		13–18	103			



Table 5. Cont.

Group	Thoracic Spine				Thoracic Morphotype				Lumbar Spine				Lumbar Morphotype				Demographics	
	SP	SSP	MTF		↓	Normal	↑	FTH	SP	SSP	MTF		↓	Normal	↑	FLK	Age, Mean or Range (Years)	n
Trampoline gymnasts [4]	46.9°	51.3°	62.8°					32°	21°	30.3°							14.9	34
	43°	49.2°	53°					40.3°	14°	25.2°								35
Scholars [52]	49.4°			3.5%	24.1%	72.4%	-	49.3°				17.2%	65.5%	17.2%			6–14	58
Weightlifting [10]	40.5°	42.7°	61.6°	0%	72.8%	27.2%	-	31.9°	15.4°	25.4°		0%		18.1%	47.5%		22.8	22
Kayakers [49]	42.5°		72.2°					28.6°		35.8°							14–17	30
Dancers [12]	28.3°	33.1°	49.7°	18.2%	85.8%	0%	-	35.1°	8.3°	19.8°		0%	84.8%	15.2%	24.3%		17–28 (22.7)	33
	22.8°	30.9°	49.4°	48%	52%	0%	-	33.8°	8.3°	19.4°		0%	93.9%	6.1%	12.2%		16–29 (22.1)	33
Rhythmic gymnasts [46]	37.5	39.7°	71.9°	0%	69.7%	30.3%	-	40.3°	5.5°	15.7°		0%	58.8%	41.2%	9.3%		17–29 (22.7)	33
	33.4°	37.6°	56.7°	3.7%	82.5%	13.8%	-	40.3°	16.2°	25.1°		1.2%	57.5%	41.3%	-		6–18	81
Swimmers [5]	28.3°	38.5°	50.4°	14.6%	80.5%	4.9%	-	35.8°	16.8°	26.3°		3.7%	62.2%	34.1%	-		6–18	82
	33.5°	39.5°	59.5°	55	70.9%	24.1%	-	35.3°	13.8°	22.9°		11.4%	63.3%	25.3%	-		6–18	79
Swimmers [7]	40.4°		78.4°	1.2%	47%	51.8%	-	31.2°		24.6°		2.3%	82.3%	15.4%	-		9–15	345
	39.5°		73.4°					36.3°		21.6°								
Scholars + intervention programme [39]	53.3°			0%	18%	82%	-	43.5°				0%	42%	58%	-		9–15	99
	48.6°			0%	38.8%	61.2%	-	50.9°				0%	18.45	81.6%	-			
Scholars [53]	34.1°	46°	60.4°	5.5%	77.8%	16.7%	43.2	29.1°	16.5°	24°		5.5%	94.5%	0%	59.7%		10–11	18
	35.3–36°	42°	64°	6.1%	66.7%	27.2%		24.8–40°	15–16.5°	28°		7.4%	82.7%	9.95			10–11	81
Adults [54]	42.3°	48.1°	56.6°	5	65.9%	34.1%	-	34.8°	17.2°	28.1°		2.4%	87.8%	9.8%	-		18–24	772
	46.7°		67.4°	0%	24.4%	75.6%	-	32.9°		22.6°		2.4%	81.9%	15.7%	-		19–22	126
Weightlifting [55]	46.3°			0%	42.5%	57.5%	-	32.3°				3.8%	83.9%	12.3%	-		18–24	772

Note: Distribution according to the classical thoracic and lumbar morphotypes in standing position and according to the integrated spinal assessment of the sagittal integral morphotype [34,35]. Abbreviations: SP; standing position; SSP; slump sitting position; MTF; maximum trunk flexion; FTH: functional thoracic hyperkyphosis; FLK: functional lumbar hyperkyphosis; C; control; †; hyper (kyphosis or lordosis); ‡; hypo (kyphosis or lordosis).

#### 4.5. Lumbar Morphotype

The most frequent lumbar integrative sagittal morphotype observed in sportsmen is functional lumbar hyperkyphosis, which involves normal lordotic angles in standing, but excessive kyphotic angles in sitting and/or flexion tests, as described by Santonja et al. [34,35,56]. The behaviour of the lumbar segment in rowers also seems to differ according to gender (Tables 2 and 5). Functional lumbar hyperkyphosis was observed in 43.2% of our rowers (twice more in men than women). A similar percentage has been observed in weightlifters (47.5%) [10], dressage riders (38.46%), and show jumping riders (40%) [13]. A higher frequency has only been found in hockey players (66.1%) [1]. Lower percentages were present in scholars (20.5–23.9%) [51], artistic gymnasts (12% in men, and 29% in women) [3], and 26% of adolescents [42]. The lowest percentages have been observed in flamenco dancers (flamenco, 12%; ballet, 24.3%) [12].

#### 4.6. Curve Adaptation According to Sport Technique, Age, and Years of Training

##### 4.6.1. Age and Years of Training

Thoracic kyphosis tends to reduce with years of training in male rowers ( $\rho = -0.258$ ), but not in female rowers. This positive effect could explain why the majority of the male rowers' thoracic kyphosis measurements (91.3%) fell within the normal range. This is an unusual finding because thoracic kyphosis tends to increase with age [57], and should make us think about the potential benefits of this sport. Lumbar lordosis was not correlated to years of training and tended to reduce with age, only in male rowers ( $\rho = -0.302$ ).

##### 4.6.2. Sport Technique

Reduced thoracic kyphosis during the three rowing phases in males and during two phases in females ('finish' and 'extension') was observed. Technique performance seems to perfect with age, leading to less kyphotic positioning; thus, explaining our adaptative results.

Lumbar lordosis increased only in females during the 'extension' phase ( $\rho = 0.503$ ) ( $X = -5.55 \pm 11.1$ ; 95% CI =  $-8.0-0.0$ ;  $p < 0.0001$ ). Again, this finding supports the benefits of this sport on the spine position in the sagittal plane. Wilson et al. [27] have also shown higher maximal flexion values between L2–L4 during rowing stroke (mean maximum flexion angle,  $54.38 \pm 9.48^\circ$  (range,  $35.88-75.98^\circ$ ). Differences in spine positioning during rowing between males and females have been observed in our sample in regards to lumbar spine performance, but no differences in age or years of training have been observed.

The SIM studies the baseline postural adaptation as a result of years of a specific training, not spinal movement during the sport performance. This methodology allows comparison between different sport disciplines, as well as plotting to normative [1,3] and developmental data [58].

The lumbar spine was kyphotic during the three phases of the rowing technique in the male rowers, and in the 'catch' and 'finish' phases in the females, but all average values were within the normative value range for flexion tests. The average convexity values were greater during 'catch', and progressively reduced during the 'finish' and 'extension' phases for both genders. The male rowers showed significantly more lumbar flexion during the whole performance, and females even showed some degrees of lumbar lordosis at the end of the stroke.

Our large sample size and no significant differences in terms of age and years of training between the genders allowed us to observe the specific distribution of the sagittal morphotype of the spine in rowers, which has been previously described in other sports (Table 5). Future studies understanding the development of this integrative spinal assessment in regards to years of training, as well as its relationship with back pain in rowers, are recommended. Lumbar curvatures in the slump sitting and trunk forward bending positions, together with height, have been recently found as predicting factors of sciatica

history in female classical ballet dancers [32]. Recurrent lower back pain has been related to the lumbar curve during trunk flexion, as well as reduced hamstring extensibility [33,59]. Hamstring length and demands are factors to consider in future analyses of rowers.

Nugent et al. [60] found that rowers without lower back pain, or considered “healthy”, have different kinematics (including flatter low back spinal position at the ‘finish’ phase) than those with lower back pain (larger lumbar kyphosis). McGregor et al. [61] found that rowers with lower back pain had significantly less range of motion at the L5/S1 level (in the ‘catch’ position:  $7.5 \pm 1.3^\circ$  in normal;  $4.8 \pm 1.2^\circ$  in previous history of lower back pain groups; and  $2.8 \pm 5.5^\circ$  in current lower back pain group). Recently, Cejudo et al. [59] found a relationship between lower back pain and static functional lumbar hyperkyphosis, and structured hyperlordosis in male and female team sports players.

Functional adaptations of the spinal sagittal curves seem to be the response to biomechanical stresses [62] and does not always depend on the years of training, but appear to be specific to each sport technique [1,3,8]. Future studies may also describe the effect of preventive intervention for the functional adaptations described.

The limitations of our study include the lack of radiographic assessment of the three positions, to establish the correlation with inclinometer quantification. Secondly, it has not been possible to measure the sagittal curves dynamically during the rowing performance, as there are no reliable measurement systems that would allow to quantify the curves of the back subjected to the workloads. On the other hand, the reliability of the inclinometer has already been studied against radiographs [7,37,38,63], and ethical limitations were found to irradiate healthy adults for non-clinical reasons.

The clinical relevancy of our research was to present that rowers are less hyperkyphotic than the general population and other sportsmen (Table 5). This was also observed in the lumbar spine, indicating that rowing may have a lumbar kyphosis-reductive effect in females, while this finding has been found to frequently increase in scholars and teenagers [35]. Regardless of this, spine alignment remained within normative ranges of lumbar kyphosis during the ‘extension’ and ‘finish’ phases of the rowing technique. A total of 9% showed lumbar kyphosis larger than  $30^\circ$  that has been related to lower back pain [60].

Future research will require the determination of the relationship of back pain to the integrated spinal assessment of the SIM in rowers. Also, establishing a specific threshold for lumbar kyphosis during the ‘catch’ and ‘extension’ phases will guide the sport technique to prevent back pain due to functional lumbar hyperkyphosis.

## 5. Conclusions

Rowing seems to provide appropriate spine positioning in the sagittal plane in the standing position, since 90% of high competition rowers showed kyphosis and lordosis values within the normal range. The SIM indicated that half of the male rowers had thoracic functional hyperkyphosis, and almost half presented with functional lumbar hyperkyphosis. In contrast, lumbar hyperlordosis is the rowing adaptation more often observed in female rowers (20.7%), while functional thoracic hyperkyphosis and functional lumbar hyperkyphosis are less frequently observed in female rowers.

Spine behaviour during the rowing technique shows that thoracic kyphosis (98.2%) and the lumbar spine (91%) perform within normative ranges, which could explain the adequate positioning of the spine in the sagittal plane upon standing. Years of rowing training tend to reduce thoracic kyphosis in males.

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Article

# Relationships, Decisions, and Physical Effort in the Marro Traditional Sporting Game: A Multimodal Approach

David Martín-Martínez<sup>1</sup>, Pere Lavega-Burgués<sup>1</sup>, Cristòfol Salas-Santandreu<sup>1</sup>, Conxita Duran-Delgado<sup>1</sup>, Queralt Prat<sup>1</sup>, Sabine Damian-Silva<sup>1</sup>, Leonardo Machado<sup>1</sup>, Pablo Aires-Araujo<sup>1</sup>, Verónica Muñoz-Arroyave<sup>1</sup>, Manuel Lapuente-Sagarra<sup>2,3</sup>, Jorge Serna<sup>1</sup> and Miguel Pic<sup>4,\*</sup>

- <sup>1</sup> Motor Action Research Group (GIAM), INDEST, National Institute of Physical Education of Catalonia (INEFC), University of Lleida, 25192 Lleida, Spain; davidmartin.eurofitness@gmail.com (D.M.-M.); plavega@inefc.es (P.L.-B.); csalas@inefc.es (C.S.-S.); cduran@inefc.es (C.D.-D.); querian@hotmail.com (Q.P.); sabrinedamian@hotmail.com (S.D.-S.); leonardoed.fisica@hotmail.com (L.M.); pab\_aires@yahoo.com.br (P.A.-A.); veronicarroyave15@yahoo.es (V.M.-A.); jserna@gencat.cat (J.S.)
  - <sup>2</sup> Smart Performance & Sport Science, Faculty of Education and Sport, University of the Basque Country (UPV/EHU), 48940 Vitoria-Gasteiz, Spain; lapuente.manuel@gmail.com
  - <sup>3</sup> Development & Innovation on Conditioning & Exercise (DICFE) Research Group, National Institute of Physical Education of Catalonia (INEFC), University of Lleida (UdL), 2192 Lleida, Spain
  - <sup>4</sup> Motor Action Research Group (GIAM), South Ural State University Chelyabinsk, 454080 Chelyabinsk, Russia
- \* Correspondence: pic.aguilar.90@ull.edu.es; Tel.: +34-645480564

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**Abstract:** The purpose of this study was to examine the players' decisions-making in Marro (a Traditional Sporting Game) through a multimodal approach. Each player's decision-making assumes specific accelerations and decelerations associated with different effort. The research objectives were: (i) to study the decision-making associated with the roles of Hunter and Hare; (ii) to know the physical effort by the roles (Hunters and Hares); (iii) to reveal T-Patterns in the multimodal strategic approach (integrated with decisions and different physical effort) with a direct incidence on the scoring by roles. The study was performed with 22 male and 2 female players aged 18 to 25 ( $M = 19.4$ ;  $SD = 1.3$ ). The Marro game was played by two groups for eight minutes. An observational methodology was used, through a type III design. The observational design was nomothetic, one-time, and multidimensional. An 'ad hoc' tool was built to ensure the data quality. Univariate analyses were performed using Crosstabs Command, with adjusted residuals (AR), Classification Trees (Chaid model) and T-Pattern Analysis (TPA). Significant differences were found between matches using the scoring ( $p < 0.001$ ;  $ES = 0.26$ ), role ( $p < 0.001$ ;  $ES = 0.31$ ), or the organic variables of the study, the speed ( $p < 0.001$ ;  $ES = 0.73$ ), the metabolic power and the acceleration/deceleration the speed ( $p = 0.023$ ;  $ES = 0.43$ ), while the predictive model pointed to the variable role ( $p < 0.001$ ) as the main factor responsible for the model growth. TPA ( $p < 0.005$ ) revealed differences attributable to internal logic in the yellow (first match) and orange (second match) teams, while organic variables were more changeable in the violet (first match) and green (second match) teams. This study advances the individualization of the decision-making process. These results may be useful to better understand the internal of functioning of the Marro game 360° since the use of various methodologies and variables (multimodal approach) provided original findings.

**Keywords:** motor decisions-making; GPS; T-Patterns; acceleration; motor praxeology; role

## 1. Introduction

Traditional sporting games (TSGs) constitute a unique family in the field of physical activity. TSGs are fundamental for the acquisition of basic life skills: cognitive, social, and emotional competences, and values and attitudes that define socially responsible citizens [1]. These are motor manifestations that result from cultural tradition, and therefore they often give rise to relationships and rules in accordance with habits and local customs. One of



these TSGs is the Marro game (Prisoner's Bar), practiced since the Middle Ages in Europe, in which two teams play against each other [1]. Players who leave their home can capture opponents who have gone out before them. However when a player is performing the role Hunter, trying to catch an opponent (Hare), he/she should notice that another opponent could have left home as a Hunter, in which case our protagonist would become his/her Hare. So, a player could be at the same time the Hunter of an opponent and Hare of another opponent. Once a player is captured, they are taken to the prison where they can be released by their teammates. The scoring depends on the number of captured players; so, there are two key actions in the game: capturing opposing players and saving teammates prisoners. To do this, it will be necessary to use individual and team motor strategies when leaving home and going through the roles of Hunter or Hare. TSGs, as Marro game, are based on a democratic agreement or, a social contract [1]. In order to play a TSG, all participants should respect the rights and prohibitions by the rules of the socio-motor game, which are organized in teams and which require players to engage in constant dialogue with others, whether they are members of the same team or rivals [2].

Any TSG has an identity card, an internal organization pattern, or internal logic, that is structured in relation to space, time, material and players, which allows its protagonists to continually adapt to a new motor situation [1,3].

The Marro game's internal logic established by the rules requires the participants to perform motor interactions with the other protagonists according to the role in which they participate. At the same time, players are asked to use each of the spaces on the playing field in an intelligent way (e.g., to calculate the risky zones to be used when they became Hunters or Hares) in order to head to the opposite zone of the field to save prisoner teammates. Finally, it will also be necessary to conduct an intelligent relationship with time (each decision will have to be taken at the most appropriate moment, calculating the risk to be taken in each motor action, depending on the scoring, that is, the number of players captured by the player's own team and the opposing team).

The Marro game becomes a laboratory of decisions, which in turn involve interpersonal relationships [1]. This is a game in which the motor conduct interpretation of opponents and colleagues, as well as the emission of messages to be decoded by other participants, are some examples of the processes that are activated. In this game, the adaptation to unforeseen events caused by the players' information uncertainty confirms the high demand for the use of reflexive or cognitive skills [4].

The science of motor action or motor praxeology offers a theoretical framework to reveal the internal order that any game generates in aspects of as much interest as decision-making. Some key concepts are the universals or operational models that contain the internal logic of any game. For example, from the Scoring System model it is understood that the Marro is a zero-sum game, which generates complete but imperfect information according to game theory [4].

Through the universal corresponding to the network of changes in sociomotor roles, it is possible to identify the decisional burden of any TSG as the Marro. A role corresponds to the limitations, rights and prohibitions prescribed by the rules for one or several players [1]. In the Marro game there are three roles: Home (being in the protected area), field player (alive), and Prisoner.

Unlike team sports, in which attention is directed towards the decisions that are triggered around the ball (relationship with the material), in the Marro game, the players's decisions are conditioned by an excellent management of timing. In this game, the "moment" of leaving home suggests the chance of having Marro (to be a Hunter) or receiving Marro (to be a Hare) over the opponents. In addition, participants may potentially become a Hunter (from a "x" rival Hare) and a Hare (from a "y" rival Hunter who has gone out later) at the same time. In these circumstances, players should choose the most suitable role in each sequence of play.

Reading (coding) and interpreting (decoding) the motor conducts of opponents and team mates confirm the importance of communication and processes involved, which at the same time are linked to extraordinary decision making. Reflective or cognitive abilities [5].

In the Marro game, decision making is associated with transitions of roles, which in turn lead to different relationships with team mates and opponents. Each of these roles triggers a set of minimal decision units (subroles). For this purpose, it may be convenient to incorporate the notion of a strategic role referring to a role integrating different groups of decisions. This is the case of the alive role, where any player could perform the strategic role of Hunter (and decides to pursue an opponent), Hare (when fleeing from the opponent), Neutral (when the decision on the opponents is not clear enough) and in Conflict (when some dispute and interruption occur between players). In this game, systematic observation allowed the identification of different subroles for each of the roles.

At the same time, these decisions and relationships trigger different energy commitments, associated with changes in space using accelerations and decelerations. The Marro game, like other collective games, is considered an acyclic or discontinuous game that requires the simultaneous participation of the aerobic and anaerobic systems to carry out with guarantees to the demands of the periods of sprint (maximum efforts) and moderate run (sub-maximum efforts). In the characteristic effort of this game, high intensity races are alternated with periods of rest or low intensity continuous races. This is the clue that it is to be considered a hybrid game, as far as conditional requirements are concerned [6].

Some adaptations on the organism could be related to the use of intervallic training with TSGs, as there are phases with very high intensities and periods of time where there are hardly any actions of medium to high intensity [7].

Normally, actions at maximum intensity are linked to changes in the score and, therefore, are determining actions when interpreting a relevant factor in the game as the final scoring.

Despite the advances in recent years, however it seems to be still scarce the line of research that addresses TSGs from the energy approach seems to still be scarce. According to [2], we will go further than previous investigations, to delve into different variables involved in the organic participation through Speed, Acceleration/Deceleration, and Metabolic Power (*MetPow*). These variables belong to players' external and Internal load, understanding external load as the parameters of the activity: Speed, Acc (acceleration), Dec (deceleration), and internal load as the physiological parameters of the player. The metabolic power contains, the internal load obtained indirectly, combining the external variables of speed and acceleration. For practical purposes, it cannot be considered as an internal load, despite the fact that the values obtained express the physiological parameters of the player.

The acceleration is the first variable corresponding to the mechanical intensity signals, which cause a change in speed over time, i.e.,  $Acc (m/s^2) = (V1 - V0)/(t1 - t0)$ . It could also be understood as the amount of speed that changes in a given time. The characteristics of the concept of acceleration, are related to changes in speed that occur with respect to the time evolution. It would be relevant to know if the beginning of the action was from a static position, or from a specific speed. For example, since the absolute value of acceleration will be lower if the player is in movement than if the action begins at a lower speed or from a static position.

The player can obtain the same value of acceleration at two different initial and final speeds. However, the intensity generated would be very different, since at the same absolute value of acceleration it will tend to be lower when the initial speed is lower. The capacity of the player to change speed when moving is directly related to the capacity to manifest force in a specific way at high speeds, that is, it depends on coordinative and conditional aspects.

Acceleration, as a study variable, also provides the actions with the negative value: decelerations or braking of the player. When the decelerations are of maximum intensity, there is a direct relationship between the initial speed with which the action starts.

According to [8], the maximum Acc. is achieved in the first second and, on the other hand, the maximum deceleration is achieved between 0.5'' and 1''. The intensity is defined not only by the value of the Acc, but also by the time it takes to reach it, since if the player manages to make the deceleration in a shorter period of time, it means that he/she manages to apply that same force, but in a faster way, and therefore the player achieves higher power values, and a higher intensity. It also seems that players usually reach a speed of 3–4 m/s at maximum accelerations, although it can reach up to 5 m/s in those players who probably have more capacity for speed. The maximum deceleration is usually reached between 2 and 5 m/s, obtaining higher values with those players who show greater values of explosive force in eccentric contractions.

*Metabolic power* (MetPow) is another's of the variables in our study. MetPow is an intensity signal [9] derived from the player's instantaneous speed [10] and acceleration [11], which indirectly determines the amount of adenosine triphosphate (ATP) required by the activity being performed.

If the speed of the action is slow and constant, the metabolic power values are low. On the other hand, when the changes in speed is higher both the values of the Acc and the MetPow are increased too. When the player slows down, instead of being 0, the MetPow values obtained are lower due to the low energy requirement [9].

Through this MetPow intensity signal, it is also possible to calculate the oxygen consumption (VO<sub>2</sub>) in an indirect way. This data allows to associate it with a concept of energy expenditure and to analyse the values obtained in order to interpret the game and to know which energy aspects it develops at an aerobic or anaerobic level.

These values allow a progressive evolution of the load for the physiological demands of this game, based on the intermittence of the player's effort. These actions are classified in the three categories with MetPow values above 20 W-Kg<sup>-1</sup>. The training schedule determines that less than 5% of the actions performed by the sample of players in eight minutes are of high intensity. However, on the other hand, a relationship is established between these actions and the score variable that determines the final result of the game. With the data obtained, a training of the players can be proposed by simulating the energetic manifestations of the relevant situations of the game as Campos and Lapuente's proposed (2018) (adapted from [7], the short interval training (<45''): Sprint Interval Training (SIT), and Repeated Sprint Training (RST).

It is possible to define individual intermittence profiles, in order to individualize this training according to the physiological demands obtained. These demands will be variable, depending on various factors such as the scoring, rival attitude, teammate attitude, etc. However, if similar conducts are established in different games, individualized training sessions can be created for each player according to his intermittence profiles. These individualized training tasks are obtained from the relationship that exists between different actions where the player exceeds a threshold of high energy expenditure and the pause that exists between both actions. This information allowed us to program the type of training that the player has to experience, to establish a direct relationship with the strategic needs during the game by each player.

Physical effort, decisions and motor interactions are different dimensions of the same multimodal or polyhedral phenomenon: the motor conduct promotes a multidimensional 360° approach: through an organic, decisional and relational nature [2].

From this theoretical framework, the objectives of the present study were performed according to two matches (four teams): (a) to study the decision-making associated with the roles of Hunter and Hare; (b) to know the physical effort in the motor actions of the roles of Hunters and Hares; and (c) to reveal T-Patterns in the multimodal strategic chains (integrated with decisions and different physical effort) in the motor actions of the Hunter and Hare with a direct incidence on the scoring.

## 2. Materials and Methods

The design was based on an Observational Methodology [12,13] already contrasted by studies in sports and TSGs [2]; located in its quadrant III [14,15]. The selection of this quadrant was justified by the following reasons: (i) the study of the decisional collectivity (teams) when playing suggests a nomothetic character; (ii) the inexistence of a monitoring plan (just although a specific recording was made); and (iii) the use of different criteria (roles) and categories (subroles) reminds that it is a multidimensional recording system [15,16].

### 2.1. Participants

This study involved 24 players (girls = 2, 8.3%; boys = 22, 91.6%), undergraduate students in the first year of physical activity and sports sciences at the University of Barcelona, Spain, who were enrolled in the pedagogy course. To practice the Marro game (two matches) players were randomized in order to form teams. The participants' decisional and energetic intervention was studied for eight minutes (480 seconds) during the game.

### 2.2. Variables

In this research six categorical variables were examined: scoring, strategic roles, subroles and speed, acceleration/deceleration and metabolic power were used.

The *Score* variable used the following coding: tie (ZE), +1 (ON), +2 (TW), +3 (TH), +4 (FO), +5 (FI), +6 (SI), +7 (SE), +8 (EI), +9 (NU), +10 (TE), -1(NO), -2(NT), -3(OH), -4(NFO), -5(NFI), -6 (NS), -7(OE), -8 (NE), -9 (NU), -10 (NT).

The strategic role variable was coded into five categories:

- (a) *Home* (HM), a player who is at home area. In this specific area, players can neither capture nor be caught.
- (b) *Prisoner* (PR), a player who has been caught while they were out of home. Prisoners players were located on one of the sides (1.5 m from the home area of the opposing team).

For the living role (being away from home), systematic observation made it possible to identify four strategic roles associated with different decision-making by the living player:

- (c) *Hunter* (HN), a player who chases an opponent.
- (d) *Hare* (HR), a player who runs away to avoid being captured.
- (e) *Neutral* (N), a player who is in a neutral situation, without directing his decision towards capturing or running away.
- (f) *Conflict* (CF), a player in a dispute against an opponent, being interrupted in his/her participation in the Marro game.

The variable *Subrole* corresponds to the minimum decision units.

When the decision-making does not change the score by roles, it was decided to identify them with the name NI (no impact on the score). In this way, the following categories were coded:

Home Role (HM):

- (a) Subrole NI: no incidence on score;

Hunter (HN):

- (a) ZNI: no impact on score;
- (b) ZC Hunter that is at the moment of capturing.
- (c) ZP is caught: Hunter who is chasing (running).
- (d) ZS Saving: a Hunter who touches the hand of one of the prisoners who is joined in a chain.

Hare Role (HR):

- (a) LNI: no incidence on score;
- (b) LC Capture: Hunter who catches a rival player;
- (c) LP She is caught: Hare that when fleeing is caught by a rival Hunter;

- (d) LS saving: Hunter who is currently saving his/her fellow Prisoners.  
Neutral (N):
- (a) No impact on score (NNI)  
Prisoner Role (PR):
- (a) NIP: no impact on scoring;  
Conflict Role (CF):
- (a) FF Conflict: Players who, for whatever reason, do not play. The stoppage of the game can be partial (players who are in conflict, but the rest are still playing) or total (the entire game is paralyzed). Whether it is total or partial, it will be categorized “in conflict” until the moment the game is restarted with the motor decision-making that is taking place at that moment of restarting the game.

By using devices with a global positioning system (GPS), three variables were recorded referring to the biological or energetic dimension of the players: Speed, Acceleration/Deceleration, and Metabolic Power (MetPow).

The variable Speed (m/s) originated six categories of thresholds (Table 1). The values were differentiated for both genders according to [17].

**Table 1.** Thresholds set in the Speed variable according to gender.

	Code	Km/h (Women)	Km/h (Men)
Low	LS	−6	−7
Medium	MS	6–12	7–13
High	HS	12–16	13–18
Very high	VS	16–18	18–21
Maximum	XS	18–20	21–24
Extreme	EN	+20	+24

The *Acceleration* variable corresponded to the mechanical intensity signals that caused a change in speed over time, i.e.,  $Acc (m/s^2) = (V1 - V0)/(t1 - t0)$ . Table 2 shows the thresholds observed and codified for this variable.

**Table 2.** Thresholds of the variable Acc/Dec.

Code in the Database	m/s
A	0 to −1
B	−1 to −2
C	−2 to −3
D	−3 to −4
E	−4 to −5
F	<−5
G	0 to 1
H	1 to 2
I	2 to 3
J	3 to 4
K	4 to 5
L	+5

The metabolic power (MetPow) was another variable related to an intensity signal derived from the player’s instantaneous speed and acceleration. MetPow indirectly determines the amount of ATP required by the activity being performed. The thresholds corresponding to this variable are identified in Table 3.

**Table 3.** Thresholds of the Met Pow variable.

	Code	w/kg
Low	LP	0–10
Medium	MP	10–15
High	HP	15–20
Very high	VP	20–35
Maximum	XP	35–55
Extreme	EP	>55

### 2.3. Procedures and Instruments

#### 2.3.1. Application of the Marro Game

A protocol was applied to homogenize the application of the procedure in practice. The explanation of the game was written and presented to the teacher for the groups before carrying out the experience in order to neutralize the influence of the explanation of the game.

Although Marro can be played in different ways, in this study, it was performed with the following rules: two teams with an equal number of players were placed in a protected area (home) behind a line at one end of a rectangular field. Each player who left the house could chase and capture (as a Hunter) the opponents (as Hares) who had left before him/her. However, if an opponent left his/her house after him/her, the latter had “Marro” on him/her and would become his/her Hunter, so he/she would become his/her Hare. When a player caught a Hare, he/she would take he/she to the prisoner area, in a side at 1.5 m where he/she would be placed forming a chain (holding hands) with the rest of the prisoners of his/her team. If a fellow Hunter or Hare managed to touch a prisoner on the chain, he saved all the prisoners, although they could be recaptured by any opponent, before returning home. The team that had captured all the opponents before, or that had captured a larger number of prisoners after eight minutes, won [2].

Before starting the match, participants performed a series of exercises to adapt to the intensity of the physical effort of the match. An 8-min Marro game was then played and recorded with two cameras for observational analysis.

On the other hand, during three sessions prior to the final recording, simulated recording cameras were placed to reduce the reactive effect produced in individuals exposed to filming cameras. The students experienced the game for the first time during the session prior to the day of the recording, and ten minutes were made available so that doubts related to strategic interpretation or rules of the game could be dispelled.

#### 2.3.2. Use of Cameras, GPS Devices, and Specific Software

After explaining the game rules, a GPS device was handed out to each player having identified the device ID and the student ID.

GoPro cameras and STAT Sports GPS devices, model Apex Pro, were employed; while the Excel tool, the data analysis programs Spss v.25 and Theme v.6 [18,19] were used.

Once the data were collected by the location systems, the StatSports software was employed to download the records. When the data were exported, the different drills, or time interval cuts from the different parts of the practice, were performed. The first cut made was from the time the device was switched on until the starting of the warm-up. All participants performed a pre-practice activation, which was logged and directed by those responsible for the study. The second record was obtained by recording five vertical jumps by each of the participants, to avoid any errors, and to make it easy to be observed the beginning of the Marro game. Finally, the end of the recording was made when the game ended, after eight minutes all players performed five vertical jumps.

Once we had the different cuts of the data export, we proceed to analyse the drill of the game, and each of the data obtained in those eight minutes.

### 2.3.3. Creation of a Database Obtained from GPS

Recording events were placed on an excel sheet. To build up this database, the first column corresponded to ‘time’, each row being reserved for the second scale. Therefore, 480 rows were used per player (8 min). While no changes were made to the original format of the database in order to apply the statistical strategy of the decision trees, nevertheless, all organic variables were transformed into a category format before the crosstab command were set up. In order to perform TPA, the events occurrences repeated in more than one consecutive occasion were deleted [2]. Thus, if during the first minute, the categories (events) appeared (for example, A, A, B, B, C, C) it would mean that the player performed the category A for 3 s, the category B for 2 s and category C for 3 s. For this reason, given that the concrete second the category (occurrence) began. It would be possible to calculate its specific time (Interval duration) since the end of a category coincided with the beginning of the next one. In this way, the repetitions of categories were deleted from raw data following two unavoidable premises (Table 4): (a) the first time that each event appeared was considered original and, therefore, included in transformed data, and on the other hand, (b) Keeping the original temporal reference [20]. This transformation allowed the use of TPA as well as a significant database reduction. This type IV data is particularly relevant when it comes to knowing the T-data structure [21], already applied in the TSGs scenario [22]. This procedure was applied with Python’s programming tool [2].

**Table 4.** Transformation performed to convert sequential data to T-Data removing repeated events from original database (second by second) in order to be analyzed by THEME v.6.

Time Scale	6 Categories (7 Events)	Duration (Interval)	6 Categories (2 Events)
10 s	TW HN NI IS A IP	→ 10 (2 s)	TW HN NI IS A IP
11 s	TW HN NI IS A IP	→ 12 (5 s)	TW HN NI IS G IP
12 s	TW HN NI IS G IP		
13 s	TW HN NI IS G IP		
14 s	TW HN NI IS G IP		
15 s	TW HN NI IS G IP		
17 s	TW HN NI IS G IP		

Note: Following previous studies (Muñoz Arroyave et al., 2021) the order data corresponding to the 7 events, one per second (One Row = One Second) (from 10 to 17 s) were transformed into two intervals (One Row = Time Interval: 2''; 5'') (from 10 to 17 s) to be analyzed as T-Data.

### 2.3.4. Observation System Validation by Roles, Subroles, and Data Quality

Different strategies were followed to address the data quality in events (occurrences). Firstly, the mixed ‘ad hoc’ system was built with the expertise supported by the GIAM research group. All the categories and criteria were defined (concept and his opposite), then put into practice by identifying them in the recordings. The game events were done directly on an excel sheet (Microsoft, 2010). Finally, two expert observers were selected to carry out a focal follow-up of each player, with a duration of 8 min. Highly reliable statistical correlation coefficients were achieved. The inter and intra Cohen’s kappa coefficient ( $\kappa$ ) were found to be higher than 0.80, thus ensuring the data quality of the events. On the other hand, to verify the quality of the GPS devices, instantaneous signal quality indicators were used, with a number of satellites above nineteen, among other indicators. It is also important to assess the facilities where the signal was obtained, in case they have metallic structures, high buildings, stadiums, etc. and this study was carried out in an outdoor sports facility without buildings or stands around.

## 2.4. Data Analysis

### 2.4.1. Use of Crosstabulations

In the present study, crosstab command ( $p < 0.05$ ) by pairs of variables were applied, taking into adjusted residuals (ARs)  $> 1.96$  or  $< -1.96$  [23] as well as effect sizes through

Cramer's V test. According to [24], to interpret the value of the effect size it was followed: 0.10 = small effect, 0.30 = medium effect, and 0.50 = large effect.

#### 2.4.2. Decision Tree

A supervised learning model or decision tree was applied, (Chaid model) [25]. Starting from the initial variable (dependent variable), the rest of the variables were considered predictive variables of the model; next requirements were followed: (i) cross validation, (ii) Pearson's chi-squared was used, (iii) the maximum levels of the model was 5, (iv) the number of cases grouped in parent and child nodes fluctuated between 100 and 200 cases. All the analyses were carried out with the SPSS v.25 tool (SPSS Inc., Chicago, IL, USA).

#### 2.4.3. T-Pattern Analysis

T-Pattern Analysis (TPA) [22,26–28] was used to reveal structural regularities, invisible to a superficial eye. TPA is a novel algorithm to find out both sequential and temporal regularities. TPA is a multivariate technique based on the lengths interval (time) and sequences of events (occurrences). Following [26] 'if A is an earlier and B a later component of the same recurring T-pattern, then, after an occurrence of A at t, there is an interval  $[t + d1, t + d2]$  ( $d2 \geq d1 \geq d0$ ) that tends to contain at least one occurrence of B more often than would be expected by chance'. The search parameters when applying TPA were: (i) Level of significance for the critical interval ( $p < 0.005$ ) [29,30], (ii) The most complex T-Patterns were based on 5 occurrences; (iii) Selection of free heuristic critical interval setting [31], (iv) TPA detection were validated by simulation, through data randomization [29,32].

#### 2.4.4. Frequency Areas

Finally, in order to construct the frequency areas the accumulations of a minimum of 35 frequencies between both team were selected, in their respective matches, with differences between team greater than ( $\pm 4.0$  adjusted residuals).

### 3. Results

Descriptive tables (Crosstabs) allowed us by pairs the associative significance ( $p < 0.05$ ) and effect size. On the other hand, the classification tree showed the strength of the variables to predict the similarities or differences by matches and teams. The use of TPA ( $p < 0.005$ ) identified to 360° multimodal strategic chains (score, role, subrole and physical values) of both Marro matches, according to different scores and roles. Finally, the frequency areas showed the differences between the matches and the teams from 360° multimodal approach. Below, the results of each of these sections were described.

#### 3.1. The Scoring Variable in Both Matches

Two Marro's matches were taking into account for analysed (11,100 s) through the 4 different teams' scoring: (Orange vs. Green, OG; Yellow vs. Violet, YV). Both matches were divided into 5760 s in the NV match and 5340 s in the AL match. This second match (AL) ended before the 8 min due to one of the teams achieved the goal of capturing all their opponents.

Significant differences were found ( $p < 0.001$ ; Cramer's V, Effect Size (ES), =0.26) when comparing the scores of the two matches OG and YV (see Figure 1). In both matches, the most frequent result was ZE (O, tie)  $n = 3600$ ; 67.41% (YV match:  $n = 1608$ ; 30.11% and OG match:  $n = 1992$ ; 34.58%). The second most found score in both matches were ON (+1) and NO (-1). Both scores were found more times in the match OG, ON (1) 996 and NO (-1) 996; (34.58%). The third most important results ( $n = 1326$ ) were TW (2) and NT (-2) (YV:  $n = 1104$ ; 20.67%; and OG:  $n = 1548$ ; 26.87%). The OG match always had a greater presence of these five types of scores (ZE, ON, NO, TW, NT), with respect to the YV match. Finally, it was noted that some scores (EI, FI, FO, etc.) were only observed in YV match.



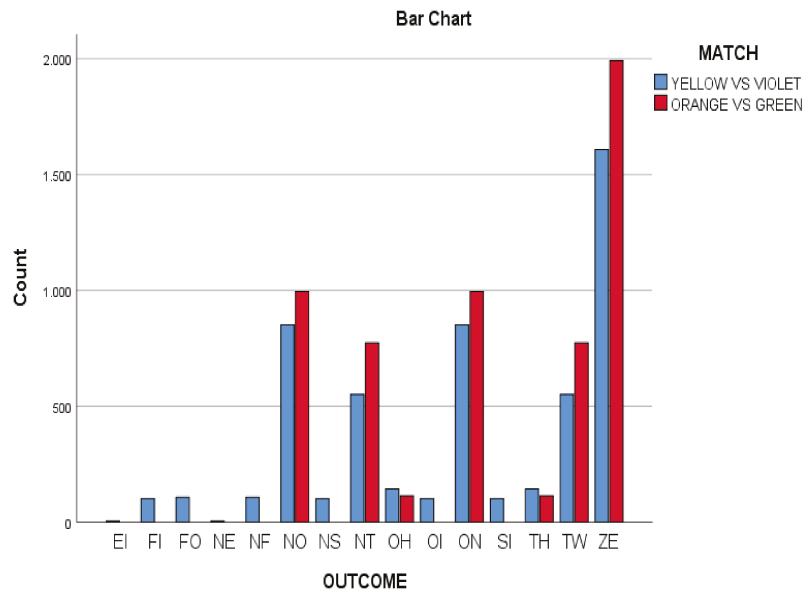


Figure 1. Outcome variable in both matches (Yellow vs. Violet, Orange vs. Green).

In order to characterize the teams, situations with the greatest margin of difference in the score were investigated. The emergence of the most complex T-patterns (dendrograms) emerged in the first match with TW (score 2) and not (−1), while in the second match the TW score was found again (2), but also on (1) and NT (−2).

### 3.2. Roles Used by Both Matches

Significant differences were observed ( $p < 0.001$ ;  $ES = 0.31$ ) when comparing the scores of the two matches OG and YV (see Figure 2). The HM (home) role was the most used in the YV match ( $n = 1709$ ; 32%), while the N (neutral) role was the most used in the OG match ( $n = 2430$ ; 42.18%). It should notice that HM and N roles are passive roles, although the type of decisions-making are potentially unequal in the N and HM roles. In the N role, the player was out of home, and at any moment he or she can go on the run or chase. In contrast, in the HM role, the player was protected at home and cannot make either decision.

YV and OG were also different in the second role. In YV the players were more active in decision-making, since a superiority of the role HN (Hunter) was found ( $n = 1341$ ; 25.11%), with respect to the roles PR (prisoner) ( $n = 933$ ; 17.47%) and N (neutral) ( $n = 817$ ; 15.29%).

In both matches, the HN (Hunter) role was more present than the HR (Hare) role. Despite sharing that regularity, the HN role was more prominent in the YV match than in OG (Hunter role: YV:  $n = 1341$ ; 25.11% vs. OG:  $n = 682$ ; 11.84%). These differences between both matches were less in the HR role (Hare YV role:  $n = 483$ ; 9.04% vs. OG:  $n = 408$ ; 7.08%).

In both matches, the role of PR (prisoner) was similar (Prisoner role: YV:  $n = 935$ ; 8.42% vs. OG:  $n = 802$ ; 7.22%).

Another finding of the study neither of the two matches were conflicting. The role of CF (conflict) was scarce (YV:  $n = 55$ ; 1.02%, vs. OG  $n = 143$ ; 2.48%).

Regarding subroles, it was observed how the decisions that did not directly affect the score (NI = no changes were detected on the score) were the most prominent.

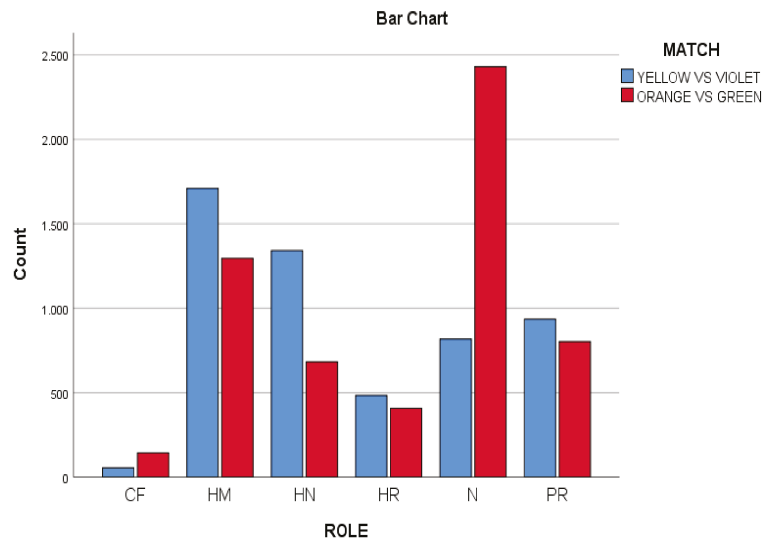


Figure 2. Role variable distributed by scoring in both matches (Yellow vs. Violet, Orange vs. Green).

### 3.3. Subroles Performed in Both Matches

Concerning the subroles (see Figure 3), the decisions-making without direct incidence (changes) on the scoring (NI) were the most frequent ( $n = 10,874$ ; 97.96%). This regularity was similar in both matches (YV:  $n = 5274$ ; 98.76%, vs. OG  $n = 5600$ ; 97.22%). The subroles CH (capture) ( $n = 28$ ; 0.25%) and S (save) ( $n = 10$ ; 0.09%) decisive for scoring only occurred on 38 occasions. This scarce presence was also observed in each match CH (YV:  $n = 13$ ; 0.24%, vs. OG  $n = 15$ ; 0.26 %) and S (YV:  $n = 13$ ; 0.05%, vs. OG  $n = 15$ ; 0.12%).

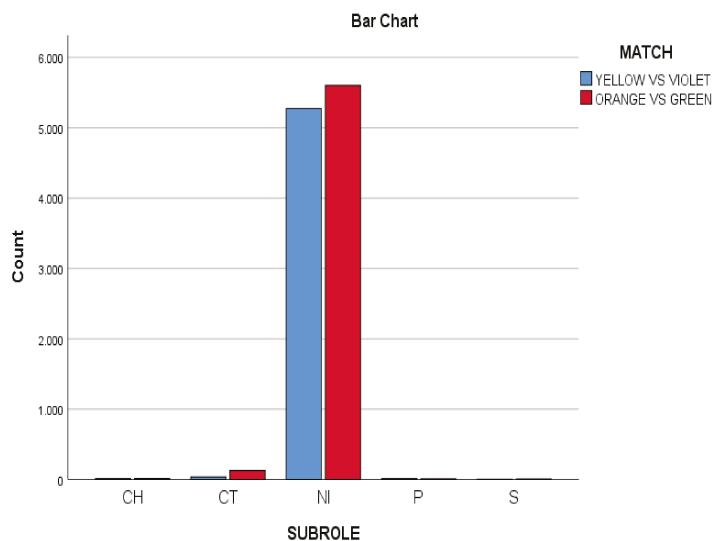


Figure 3. Subrole variable in both matches (Yellow vs. Violet, Orange vs. Green).

### 3.4. GPS Devices in Both Matches

It was observed a great predominance of the less intense values (>80%) in the three variables: Speed (Figure 4), Acceleration/Deceleration (Figure 5) and Metabolic Power (Figure 6), in both matches (YV and OG).

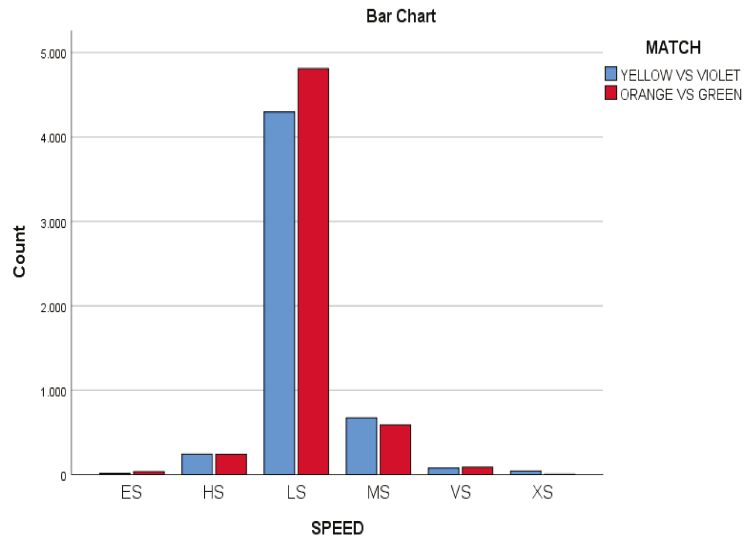


Figure 4. Speed in both matches (Yellow vs. Violet, Orange vs. Green).

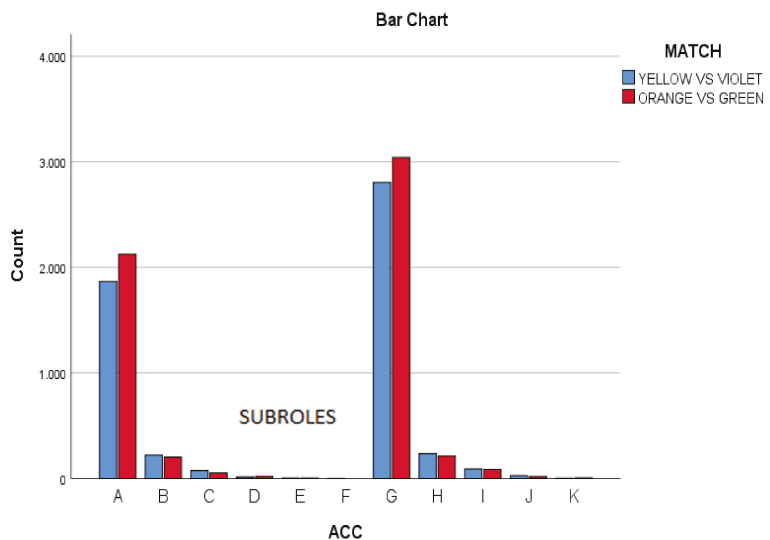
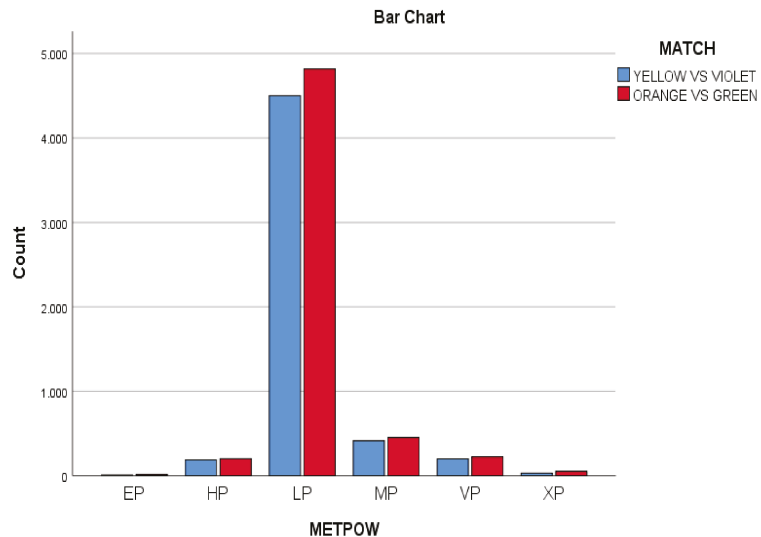


Figure 5. Acc in both matches (Yellow vs. Violet, Orange vs. Green).

The minimum values of the three variables were higher than 80% of the total sample in each of the variables. Speed actions, there are less than 7 km/h per second (6 km/h in the case of girls), ( $n = 4297$ ; 80.46%) in the YV match and ( $n = 4811$ ; 83.52%) in the OG match. In both matches, the MS (medium speed, between 7 and 13 km/h) and HS (high speed,

between 13 and 18 km/h) actions had a similar protagonism (YV match: MS (medium speed)  $n = 670$ ; 12.54%; OG match:  $n = 587$ ; 10.19%). The three maximum speed categories (VS, XS and ES), in the two matches represent less than 2.5% of the total actions, (YV match:  $n = 131$ ; 2.27%; OG match:  $n = 123$ ; 1.10%).



**Figure 6.** MetPow in both matches (Yellow vs. Violet, Orange vs. Green).

Regarding the speed variable, it was observed that most of the match actions (>80%) were performed at low speed (LS = men < 6 km/h; women < 7 km/h) in both matches (LS: YV:  $n = 4297$ ; 80.46%, vs. LS OG  $n = 4811$ ; 83.52%). The average speed values (MS = 7 to 13 km/h) corresponded to 10% of the playing time of both matches (MS: YV:  $n = 670$ ; 12.54%, vs. MS OG  $n = 587$ ; 10.19%). Finally, high speed actions (HS from 13 to 18 km/h) were less than 3% of the actions of both matches (HS  $n = 131$ ; 2.27%, vs. MS  $n = 123$  times; 1.10%).

In the section on accelerations and decelerations, it was described how the same manifestations of the players were observed, despite they were playing in two different contexts, with different decisions-making and motor actions. In the two matches studied, the actions with less speed (−1 m/s to 1 m/s) occupy 88.61% of the actions with 9836 times of the total ( $n = 11,100$ ). Actions with acceleration range 3 to 5 km/h and those with deceleration range −3 to −5 km/h represent 0.83% of the total actions analysed ( $n = 93$ ), indicating that actions with maximum intensity of this variable do not often appear.

It was also observed that in both matches the values of accelerations and decelerations had a similar protagonism. Most actions were of lower speed (AC/DC Low −1 m/s to 1 m/s:  $n = 9836/11,100$ ; 88.61%). Acceleration actions from 3 to 5 km/h, and deceleration actions from −3 to −5 km/h were very scarce ( $n = 93$ ; 0.83%).

The relationship between the values of maximum acceleration and maximum speed at that particular moment, obtained from the players in the study, describes a linear relationship that allows us to identify a linear slope, to find out, the acceleration capacity with respect to their speed. With this data, not only the performance of the players could be evaluated, but also it could be prescribed training to each subject.

Relationships were established in the study of the values of metabolic power obtained in the players by two matches. Where the average difference of the variables studied was 0.20, it is then determined that the same exact values were reproduced, despite studying two different situations of the same game. The LP category (low power, from 0 to 10 w/kg) appears in more than 83.95% of the total, being (4501) times; 84.28% in the YV match, and

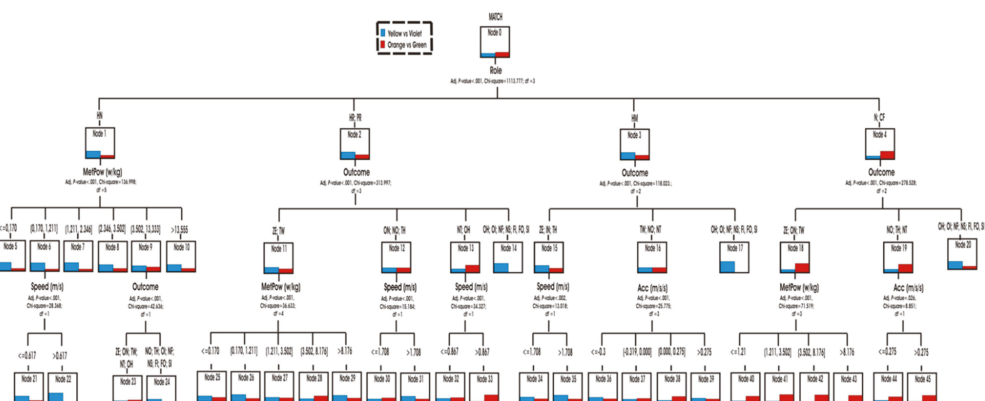
(4818) times; 83.64% in the OG match. In the comparison with each of the variables, there was a direct relationship between the two matches, where the greatest difference between the values of MetPow (metabolic power) was 0.66 in the variable that appears most in LP (low power). The values in the YV heading were LP ( $n= 4501$ ; 84.2%), MP (medium power) ( $n= 415$ ; 7.77%), HP (high power) ( $n= 187$ ; 3.5%), VP (very high power) ( $n= 200$ ; 3.74%), XP (maximum power) ( $n= 30$ ; 0.56%), and EP (extreme power) ( $n= 7$ ; 0.13%), unlike the OG match where the values of LP ( $n= 4818$ ; 83.6%), MP ( $n= 451$ ; 7.82%), HP ( $n= 202$ ; 3.50%), VP ( $n= 223$ ; 3.87%), XP ( $n= 51$ ; 0.88%), and EP ( $n= 15$ ; 0.26%).

The two matches yielded the same values for the different categories: LP (low power), MP (medium power), HP (high power), YP (very high power): YV match ( $n= 4501$ ; 84.2%); OG match ( $n= 4818$ ; 83.6%).

### 3.5. Predictive Capacity Variables by Teams

Multimodal results 1. Predictive capacity variables on the multimodal conduct in the two matches.

Through the hierarchical segmentation technique or classification tree, the predictive capacity of the score variables (role, speed, accelerations/decelerations and metabolic power) was explored to reveal the similarities or differences in the multimodal behaviour of both matches (Figure 7).



**Figure 7.** Predictive capacity of the scoring variable (two matches). Note. For easier reading of this figure, this classification tree has been segmented into different images, which are shown below.

The role was the first predictive variable ( $p < 0.001$ ; chi-square 1,113,777,  $df = 3$ ) in both matches. The teams in YV match, spent more time in the Hunter role HN (node 1) and being at home HM (node 3) (Figures 8 and 9) than the teams in OG match. They also spent slightly more time in the roles of Hare HR and Prisoner PR (Node 2). In contrast, the teams in the OG match participated more in the Neutral N and Conflict CF roles (node 4) (Figure 10).

The second most predictive variable when comparing both matches was the score. Score was the explanatory variable in three of the previous nodes.

The physical variables were the least present in the tree as predictive variables: metabolic power (three times, once at the second level), speed (three times at the third level) and acceleration/deceleration (twice at the third level).

The main predictor variable for the roles Hare HR and Prisoner PR (Node 2) was the score ( $p < 0.001$ ; chi-square 313.997,  $df = 3$ ). It was observed that the teams in the AL match participated for longer than those in the OG match with the Z and TW ties scores (Node 11). In this case, the next predictive variable of both matches was the metabolic power (nodes from 25 to 29).

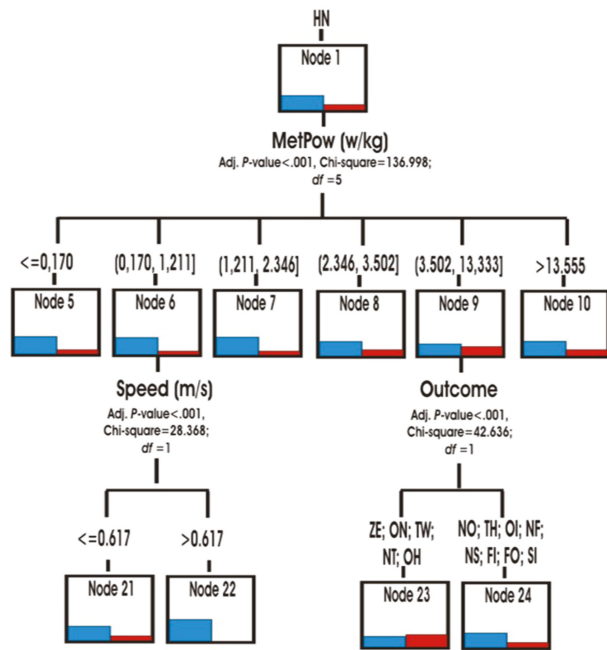


Figure 8. Predictive capability of the Hunter role.

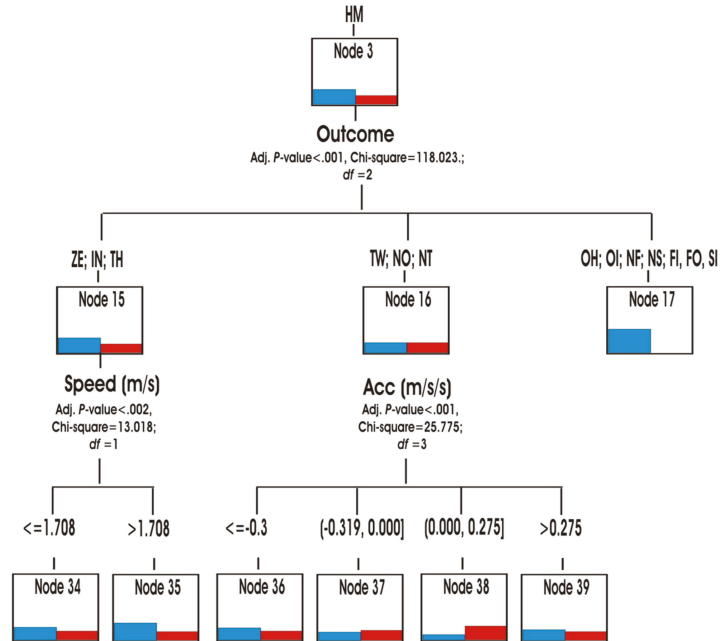


Figure 9. Predictive capability at Home Role.

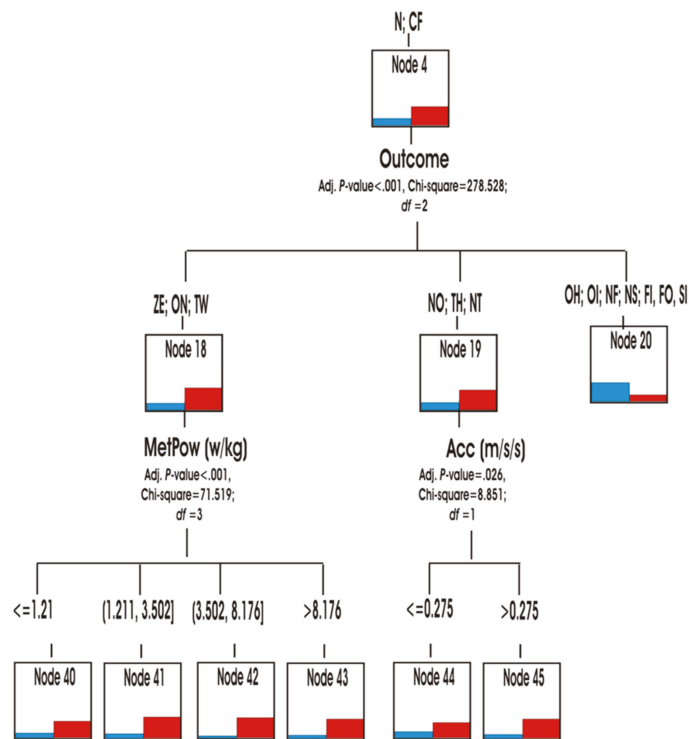


Figure 10. Hare and Prisoner Role Predictability.

In parallel, the YV match participated much more than OG in actions with the scores OI, NF, NS, NE, FI, FO, SI (node 14). The OG match played longer than YV with the NT and OH scores (node 13). The two matches had a similar prominence when the score were ON, NO, TH (node 12). The regularities found in the nodes 12 and 13 were specified with more detail when the following speed prediction variable was used.

When the teams were in the HM home role, the tree identified the score variable that originated three types of behavior (nodes 15, 16 and 17) according to different scores ( $p < 0.001$ ; chi-square 118,023,  $df = 2$ ). The teams of the YV match spent more time than those of the OG match in the role of HM with the scores of ZE, ON, TH (node 15). In this case, the next predictor variable was the speed to go from one place to another in the home or to go out. It was observed that, in the YV heading, the actions were done with greater speed ( $V > 1708$  m/s) than in the OG heading ( $p = 0.002$ ; chi-square 13,018,  $df = 1$ ). The teams participated in different accelerations/decelerations when they intervened with TW, NO, NT (node 16) scores ( $p < 0.001$ ; chi-square 25,755,  $df = 3$ ).

It was also observed that the teams of the OG side spent more time than YV adopting the neutral N or conflicting CF roles (node 4) ( $p < 0.001$ ; chi-square 278,528,  $df = 3$ ). This superiority was also shown in two types of scoring: ZE, ON, TW (node 18) and NO, TH, NT (node 19). In contrast, the trend was reversed with the other scores OH, OI, NF, NS, FI, FO, SI (node 20). In order to predict the transition to other roles, the tree predicted the metabolic power (node 18) or the acceleration/deceleration (node 19) variables as predictive in both matches (Figure 11).

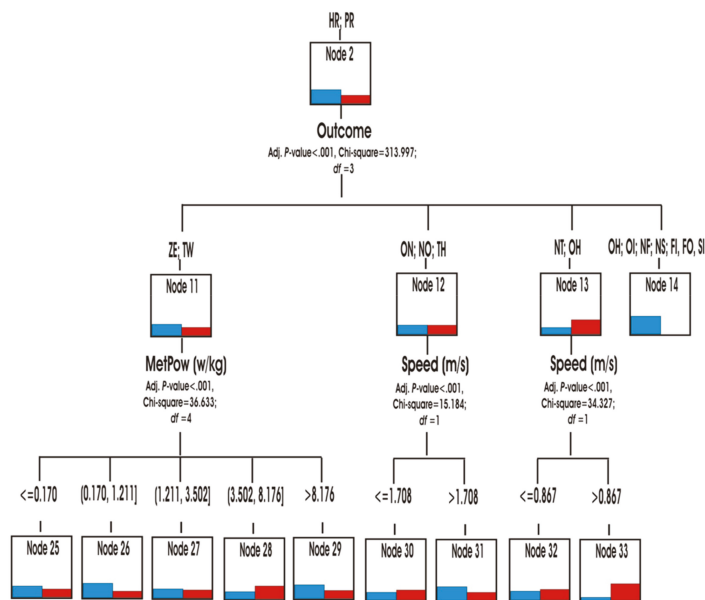


Figure 11. Role Neutral and Conflict Predictability.

Finally, the main predictive variable for the Hunter role HN was the metabolic power ( $p < 0.001$ ; chi-square 136.998,  $df = 5$ ). The match YV showed recorded higher MetPow values in most occasions with respect to what happened in the OG match. This superiority was mainly identified in energy actions with values between 0.1 and 1.21 w/kg (node 6). For this node, speed also intervened as a predictive variable. It was observed that the speed of the actions was higher in the match YV with respect to the OG ( $p < 0.001$ ; chi-square 26.368,  $df = 1$ ). In the remaining cases, the two matches intervened with a similar MetPow in motor actions with values ranging from 3.50 to 13.5 w/kg (node 9).

Multimodal results 2. T-Patterns through the strategic multimodal by teams in the Marro game.

Figures 12 and 13 shows the strategic multimodal chains in both matches. In match YV with the TW scoring (+2), the variable chains (frequencies) most used by the Yellow team are (tw, hn, ni, ls, g, lp), therefore, with TW score (+2) and HN role (Hunter) and subrol NI no incidence on the score, with minimum values of speed, acceleration and metabolic power.

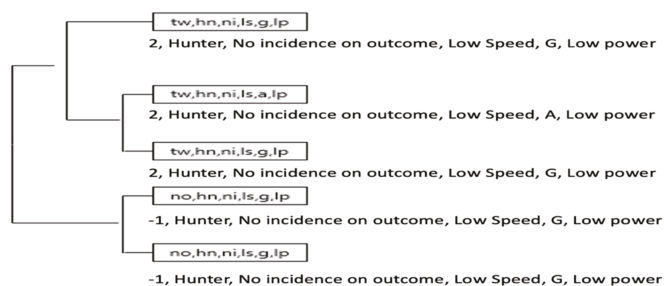
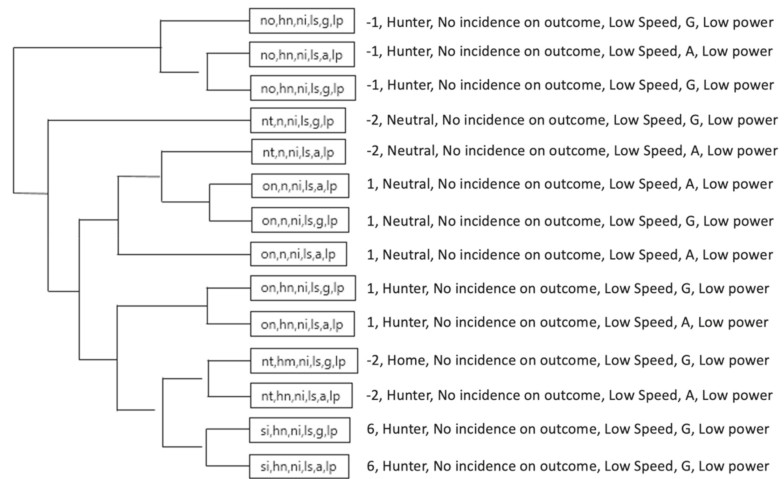


Figure 12. The most complex T-Patterns detected by Yellow team.





**Figure 13.** The most complex T-Patterns detected by Violet team.

The strategic chain (no, hn, ni, ls, g, lp) by the Violet team, in which the Hunter role appears with a NO scoring (−2), and with the minimum values of speed, acceleration and metabolic power. On the Violet team, the ON (1) score also appear, linked to a N (neutral) role (Hunter).

Description of the T-Pattern analysis:

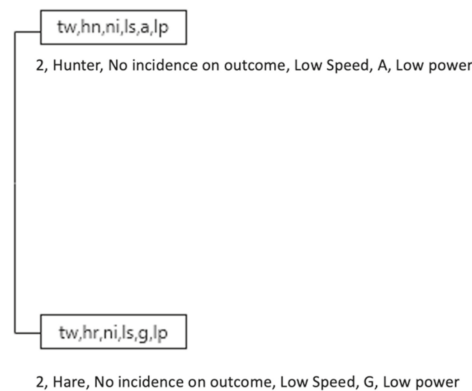
In the Yellow team, 2 strings were identified with result in favor of 2 (result +2, Hunter role, subrole no incidence score, low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg), after which followed the sequence of result in disadvantage of 1 (result −1, Hunter role, subrole no incidence on outcome (scoring), low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg).

Violet Team:

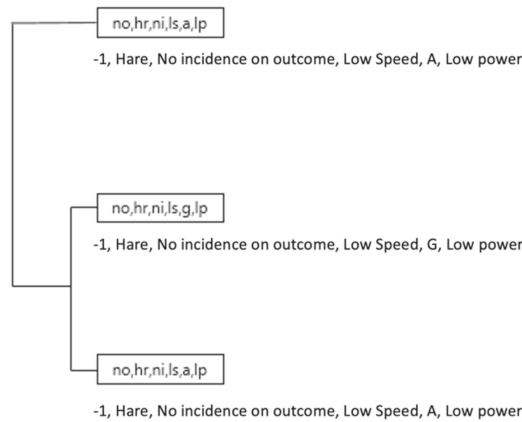
In general, there was some regularity to be used the Neutral role through the following outcomes (scoring) −1, −2 and +1, while the Hunter role in favor +1 outcome (scoring). Finally, there was a tendency for the home role to be used if the result was at a disadvantage of 2.

Score:

On the other hand, while in the Yellow team, with result TW (+2) the Hunter role, also the HR (Hare) role appeared (Figures 14 and 15).



**Figure 14.** The most complex T-Patterns detected in Yellow team by Hare roles.



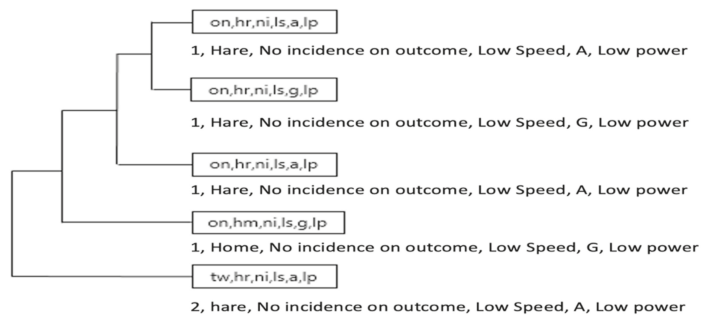
**Figure 15.** The most complex T-Patterns detected in Violet team by Hare roles.

In the Yellow team 2 chains were identified with result in favor of 2 (result +2, Hunter role, no incidence subrole, score, low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg), after which followed the sequence of the same result with the appearance of the role Hare (+2, Hunter, no incidence outcome, low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg and (+2, Hare, no incidence outcome, low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg).

The Violet team offered its own particularities with a regularity in the Hare role with the marker at –1 and the subrole of no incidence outcome, accompanied by physical effort variables of low speed, Acc/Dec 0–1 m/s, MetPow 0–1 w/kg.

In the OG, it was mainly observed that the strategic chains of the Orange team (on, hr, ni, ls, a, lp) and (on, hr, ni, ls, g, lp), associated an ON score (+1) and HR Role (Hare) as well as minimum values of speed, acceleration and metabolic power. Then, the same score appeared in Role HM (home) and HR (Hare) unlike the Violet team with strategic chains that prioritized a NT score (–2) associated to Role N (neutral) and Subrole NI (no incidence in the score).

Figures 16 and 17 illustrates a more complex dendrogram in Green team than Orange team. However, T-Patterns (categories) in Orange team were more dynamic by involving two types of scoring and two different roles (on, hr, ni, ls, a, lp), (on, hm, ni, ls, g, lp) and (tw, hr, ni, ls, a, lp), while in the Green team, only the acceleration and deceleration thresholds would vary (nt, n, ni, ls, a/g, lp).



**Figure 16.** The most complex T-Patterns detected in Orange team.

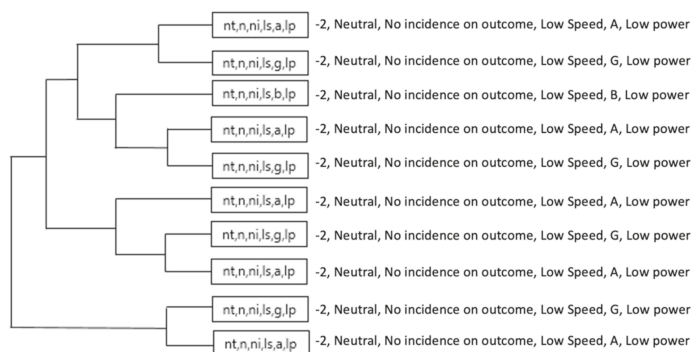


Figure 17. The most complex T-patterns detected in green team.

Multimodal results 3. Frequency areas of strategic multimodal chains of Marro team and consignments

Figures 18 and 19 illustrate the 360° multimodal strategies of the twenty-four participants in OG and YV matches. Combinations of Categorical variables with frequencies ( $\geq 35$ ) or were considered, so combinations below these values were eliminated ( $n < 35$ ). Also, values between  $\pm 4$  of the residual adjustments were used as an exclusion requirement. Therefore, all values ( $>4$ ) and ( $<-4$ ) were included.

The number of most repeated chains in the two matches was different. A higher frequency of repeated strings was found in the teams of the OG match than in the teams that faced each other in the YV match. OG Match (Orange Team  $n = 132$  and  $112$ ; Green Team  $n = 80, 139, 152$  and  $158$ ) and AL match (Yellow Team  $n = 70$  and  $80$ ; Violet Team  $n = 61, 62$  and  $70$ ).

In the Marro game the score was constantly modified (since the number of prisoners captured and saved changed without any established order). It was observed that the strategic chains that each team originated were activated in the face of unequal scores (Orange Team TW =  $+2$ ; Green Team NO =  $-1$  and NT =  $-2$ ; Yellow Team ON =  $+1$ ; Violet Team NO =  $-1$ ).

The four teams (two matches) started their strategic chains from a different role, depending on whether their scoring was favourable or unfavourable. With a favourable score (TW or ON), the teams originated chains from the home or neutral role; while when the score was unfavourable ( $-2$  or  $-1$ ), the more numerous chains were activated from the prisoner (PR), neutral (N) and also home (HM) roles. Only one chain was identified associated with the conflict role (FC). The subroles corresponded to decision units with little energy relevance in the three variables: low speed (LS), low acceleration/deceleration ( $G = \text{Acc/Dec } 0\text{--}1 \text{ m/s}$ ); and low Met Pow ( $0\text{--}10 \text{ w/kg}$ ). In addition, in general with little motor relevance (neutral role, house, prisoner associated with decisions without incidence on the score).

The multi-modal chains most present in the different team were the following:

Match OG:

Orange Team:  $+2$  TW, N, NI, LS, G, LP ( $n = 132$ );  $+2$  TW, HM, NI, LS, G, LP ( $n = 112$ )

Green Team: Tie ZE, HM, NI, LS, G, LP ( $n = 152$ );  $-1$  NO, HM, NI, LS, G, LP ( $n = 158$ );

$-1$  NO, PR, NI, LS, G, LP ( $n = 139$ );  $-2$  NT, PR, NI, LS, G, LP ( $n = 80$ )

Match YV:

Yellow Team:  $+1$  ON, HM, NI, LS, G, LP ( $n = 85$ );  $+1$  ON, HM, NI, LS, A, LP ( $n = 70$ )

Violet Team: ZE, N, NI, LS, G, LP ( $n = 70$ );  $-1$  NO, N, NI, LS, G, LP ( $n = 62$ );  $-1$  NO, N, NI, LS, A, LP ( $n = 61$ )

#### Orange Team vs. Green Team Duel

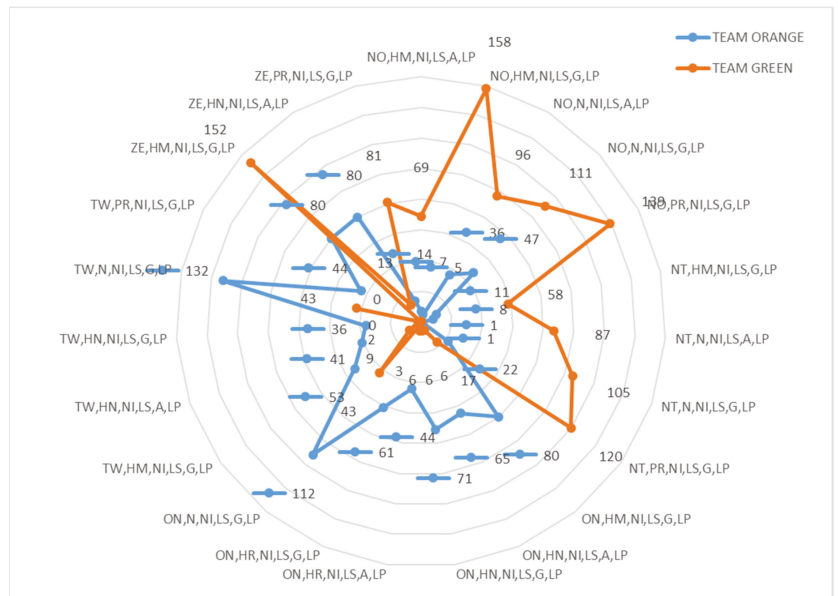


Figure 18. Frequency areas team Orange vs. team Green.

### Yellow Team vs. Violet Team Duel

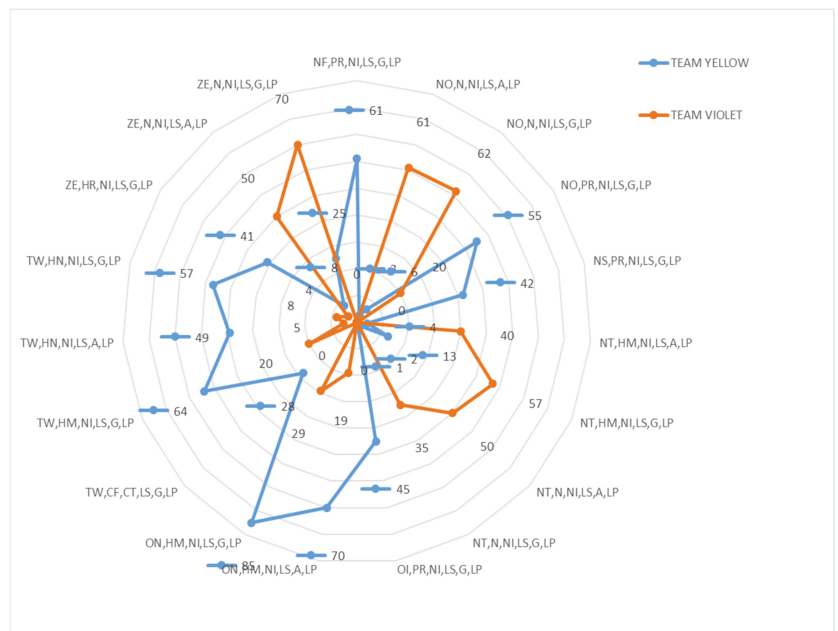


Figure 19. Frequency areas Yellow Team vs. Violet Team.

## 4. Discussion

The aim of this research was to study of two matches of Marro game by using a 360° multimodal approach [2]. This multi-approach methodological premise made it possible to determine with enough specificity according to organic and decisional conducts (direct observation). The notion of 'match', as variable was novelty in research through TSGs.

### 4.1. First Results, Ready for a Multimodal Approach: The Case of the Marro Game

The multidimensional approach Marro game notes the importance of attending different dimensions in an intertwined way. Statistical analyses complement each other and allow us the 360° multimodal approach, considering roles, decisions or subroles and energy implications (speed, acceleration/deceleration and metabolic power). All of this was based on a key element of the internal logic of the Marro game: the scoring that was constantly changed. The most frequent result was a draw, followed by other scores such as ON (+1), NO (-1), TW (2) and NT (-2). In this game a team can reach tied, reaching tied again, or having a one-point advantage or it can even happen that a team manages to finish the game early, having reached the limit score, as happened in the match Yellow against the Violet team. This is another original feature of the game's rule, which is confirmed by players. The teams of both matches experienced two different game adventures, where the scores during the game were different. The findings show the unpredictable and changing nature associated with a varied decision-making process, depending on the information uncertainty generated mainly by the opponents [4].

It has been observed that the two matches have followed different dynamics, so the Yellow and Violet teams of the YV match participated longer in the HN (Hunter) role than in the prisoner (PR) or neutral (N) roles. This superiority led to participation in motor actions of greater commitment or decisional risk. This does not mean that the HR role was not present in the OG role, since in the four team duels (YV and OG) the HN role was more present than the free HR role (Hare).

The multimodal vision facilitates the integrated approach of decisions with relationships. In this case, the conflict role played very little in both matches. Therefore, we can affirm that we are dealing with participants very respectful each other.

The most frequent (subrole) decisions (NI) do not have a direct impact on the score. We might think that this is a rather undynamic game. However, we may think that, above all, strategic interventions predominate, which serve to prepare the game's high points: the capture of an opponent or the release of prisoners from our team.

The results from Speed, accelerations/decelerations and metabolic power complement the previous findings. Low values were observed in all three variables, which requires us to interweave the different classes of variables (scoring, role, subrole, speed, acceleration/deceleration and metabolic power). The data allowed us to estimate by an iterative procedure the time course of the speed and hence of the acceleration yielding an energy cost value such that when multiplied by the speed yields the actual metabolic power [33].

Metabolic power is not the panacea for team-sport analysis, but it is a very useful tool. By estimating the cost of acceleration in activity comprising perpetual changes in speed, it addresses a fundamental flaw of existing approaches. Consequently, it provides a more comprehensive—yet still incomplete—measure of intensity and volume for variable-speed locomotion, allowing for improved monitoring of training and competition loads and thus represents a step in the right direction in the quest to understand the demands of team sports. Metabolic power is not the panacea for team-sport analysis, but it is a very useful tool. By estimating the cost of acceleration in activity comprising perpetual changes in speed, it addresses a fundamental flaw of existing approaches. Consequently, it provides a more comprehensive—yet still incomplete—measure of intensity and volume for variable-speed locomotion, allowing for improved monitoring of training and competition loads and thus represents a step in the right direction in the quest to understand the demands of team sports. Metabolic power is not the panacea for team-sport analysis, but it is a very

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Finally, the technology for data collection must be properly selected: when dealing with accelerations, any sample rate below 10 Hz is highly questionable. Additionally, signal filtering should be considered to smooth the accelerations/decelerations of the center of mass in sync with the stride frequency to reduce noise, without losing information [35].

Metabolic power is proposed [36] as a more accurate way of calculating the intensity because its calculation integrates both the speed and acceleration that the player demonstrates at each moment, instead of considering them separately.

#### 4.2. Decision-Making Importance and Scoring: The Marro 360° Multimodal Approach

The classification trees establish a hierarchical order of the variables when it comes to finding the strategic specificity of both matches. The first variable Role (which integrates the different subroles) offers greater explanatory force. Secondly, the scoring intervenes, which confirms the importance of time in this game. In Marro, the player who comes out “last” has an advantage over the others (it is a relationship with time); the game is based on moving from one role to another (it is also a relationship with time) in a constant roles changes [1]. Finally, unlike sports, the scoring can move forward, backward or even a team can win before the agreed time has elapsed (again, the relationship with time is key).

The player must constantly detect, interpret and understand; he or she must pay attention to ongoing relationships and bring out new ones, in turn generating new organizations and creating a distribution of the game. The player is constantly in motion [37].

The strength of the decisional and relational temporality of the game justifies that the other three energy variables remain in the predictive background and appear in the last place in the hierarchy of the classification tree. This indicates that the decision takes precedence, and the energetic implication is at the service of the decision-making.

This type of observation has been less explored in sports competition [38] which is more interested in determining sports performance indicators. The paths followed by the matches obey the same internal processes that occur within the teams. However, it is up to the physical education teacher to increase a global, interpretative and reflexive approach, in order to fill his or her decisions with contextualised content, without excluding the fulfilment of current competences [39] TSGs.

With all this, the second methodological approach invites us to reveal multimodal strategic temporal regularities in the four teams, that is, in both matches.

#### 4.3. In Search of T-Patterns through Strategic Multimodal Chains

Knowing the decisional anatomy of the teams or matches in any TSG requires addressing a range of connectivity that nests on roles and subroles to guide the study of game action, and thus build a comprehensive coding system; composed by criteria (roles) and categories (subroles), in line with observational studies, themselves considered mixed methods [32]. However, how to address the spontaneity of motor actions through TSGs? Linked to the mixed approach [40], in-depth analysis by specialists is required as a prerequisite for understanding the identification of the specificity of roles to play [41].

The TPA approach has revealed strategic regularities (T-Patterns) used by the four teams. In addition, the frequency areas provide a graphical (visual) aid to identify the most commonly used multi-modal chains by teams. These two complementary strategies suggest a more integrated view of the game dynamics, and allow us to advance a multimodal interpretation of the phenomenon being studied.

In the Marro game [42] is unavoidable to consider the temporal relationship of the players actions, understood as an unavoidable strategic indicator [2]. This approach distinguishes from other studies, more interested for sociological issues [43]; and thus, pay attention on increasing the understanding of the dynamics of matches and teams while playing. The real interest of this study has been ambitious, overcoming the isolated and fragmented vision of some data of the game: to reveal interconnected pieces of the two matches understood as a puzzle of interconnected relationships. How and when to send coded messages between partners, undecipherable by the rival threat?

The preference of actions aimed at capture over the rival in the role of Hunter should be understood as the visible part of the strategic iceberg of both matches. The findings from the TPA perspective reveal regularities of multimodal strategic time patterns reflected in the dendrograms of the four teams. The dendrograms reflect the selection of the most complex multimodal strategic chains with the highest impact on the scoring. The most relevant fact does not point towards the identification of regularities in the Hare role in both teams, but the favourable score that rested on the Yellow team (+2) could be due to the identification of the transition of roles [1] not identified in the Violet team. The same fact was detected in both matches, when the passive of the role in the Green team could facilitate the overcoming in the team's score.

This is a game with an important decision-making process because the actions are fast-paced. In this sense, decision-making could be even more decisive than metabolic indicators. Speed, acceleration and deceleration, and metabolic power can be a consequence of the team's plan. This action plan, as shown by the dendrograms and the frequency areas, devotes most of the game to motor interventions that do not directly affect the score. Players continually switch relationships to capture opponents or to save playmates. The game contains great originality in the rules, decisions and relationships and energy management.

These findings provide us a different view of the ludic context of TSGs, recognised as a scenario characterised by complexity [44] and playful spontaneity [45]. Within the apparent disorder that originates the superficial look of the game dynamics, there is a deeply strategic and temporal order [18,19,26] which suggests to take a distance from the identification of decision-making as random process (Limayem, and [46,47]).

Playing TSGs as Marro game can be beneficial for battling the problems of physical non-activity in modern western societies but also to serve as a link between the society and its citizens. By playing such traditional sports games, the players re-enact similar experiences of the local culture that were played by people of other generations in the past.

## 5. Conclusions

In this study, the responses of the players have been observed in depth according to the score in the form of strategic decisions, and how these affect the physical effort of the player.

The two groups were different because their strategic multimodal chains were unequal, despite certain similarities when analysing the two matches in different contexts. The physical effort has been similar in both matches. The findings confirm that Marro game could be categorized into intermittent efforts, and determinants in the high intensity score, but with a higher percentage of time in unemployment or with low physical activity.

We have observed that teams play similarly on certain scores, and it is intuitive that this may be determined by the risk assumed with a score for or against. But we must be cautious as no direct link is established between the risk to be assumed by the player and the analysis of the complexity of the action or the requirement of the opposing team.

Finally, no link is established between the modification of the score and the conditional response, since, in the Blue vs. Violet match, the score was more dynamic than in the Orange vs. Green match, but the energetic values were similar in both matches without evident differences in this sense.

## 6. Limitations and Future Prospects

One of the limitations of this study is the lack of a priori knowledge of the team's strategic plans in order to evaluate the interventions in a richer context. On the other hand, addressing the emotional state of players would favour the understanding of the 360° vision of players' motor conducts, in accordance with what is proposed by the science of motor action [1]. Finally, conducting interviews with players in their respective teams and rivals would help to understand the intentionality and significance of the findings around the participants' strategic chains.

The original 360° multimodal vision provided by this line of research, almost unheard of in observational studies, proposes as future prospects to continue advancing in this type of study that facilitates the interpretation of the processes that occur in the development of a game, that is, to reveal the networks of interconnections that originate the implementation of the internal logic of any TSG.

As indicated by [33]:

“Each player is the agenda of a behaviour of motor interrelations to which he/she gives meaning within the confrontation of two teams. We are therefore at the heart of a group dynamic whose originality is to express itself through movement”

(2018, p. 91).

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Article

# Do Blood Lactate Levels Affect the Kinematic Patterns of Jump Shots in Handball?

Ivan Belcic <sup>1,\*</sup>, Sasa Rodić <sup>1,2</sup>, Vedran Dukarić <sup>1</sup>, Tomislav Rupčić <sup>1</sup> and Damir Knjaz <sup>1</sup>

<sup>1</sup> Faculty of Kinesiology, The Laboratory for Sports Games, University of Zagreb, 10000 Zagreb, Croatia; sasa.rodic@kif.hr (S.R.); vedran.dukaric@kif.hr (V.D.); tomislav.rupcic@kif.hr (T.R.); damir.knjaz@kif.hr (D.K.)  
<sup>2</sup> High School Jastrebarsko, 10450 Jastrebarsko, Croatia  
\* Correspondence: ivan.belcic@kif.hr

**Abstract:** The aim of this study was to determine whether the dynamic motor stereotype of movement (shooting technique) is violated under conditions of an increased lactate concentration in a player's blood after a 30–15 intermittent fitness test. The hypotheses was that there would be statistically significant differences in ball speed and shooting accuracy in jump shots on the goal before and after the occurrence of fatigue in the player. The sample of respondents consisted of 10 top-level handball players of the highest competition rank in Croatia. The results showed significant differences before and after the fatigue protocol in the run-up speed ( $F = 5.66$ ;  $p = 0.02$ ), in the maximum speed of the forearm ( $F = 5.85$ ;  $p = 0.02$ ) and the hand ( $F = 4.01$ ;  $p = 0.04$ ), in the speed in the shoulder joint ( $F = 5.39$ ;  $p = 0.02$ ) and wrist joint ( $F = 4.06$ ;  $p = 0.04$ ), and in the ball shooting speed ( $F = 5.42$ ;  $p = 0.02$ ). The accuracy of the shot was, on average, lower (36.20 vs. 33.17 cm) but not significantly so. High blood lactate levels affect changes in certain kinematic parameters during the performance of a jump shot in handball. Consequently, this reduces the speed of the shot, which can affect situational performance as one of the two significant parameters of scoring success.

**Keywords:** movement analysis; performance; handball shot; internal load; shot precision

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## 1. Introduction

The evolution of handball is leading handball in the direction of an even faster and more dynamic game in which speed, agility, speed endurance, and explosive power are increasingly important for success [1–3]. Under these conditions, in order to achieve a logical objective that leads to victory (i.e., scoring more goals than the opponent) [4,5], attackers try to obtain and ensure the optimal position for a shooter. This is achieved by using fast movements on the field at short distances with the help of fast and strong changes in movement direction of the body, with or without the ball, using different offensive actions [6]. Handball is a physiologically demanding game that places medium-to-high demands on a player's aerobic system, while also placing significant loads on the anaerobic energy system [7,8]. In a study conducted by Chelly et al. [9] on a sample of 18 handball players, national team members of the average age of 15.1 years, it was found that the average heart rate was  $172 \pm 2$  beats per minute and the average blood lactate concentration is  $9.7 \pm 1.1$  mmol/L during the monitored championship matches. This indicates extremely demanding physiological loads during a match, in which aerobic and anaerobic modes of operation alternate.

Force production, coordination of movement, motor control precision, time of muscle reactions, and proprioception are directly negatively affected by the occurrence of fatigue [10]. Considering research on the influence of fatigue on changes in certain kinematic parameters in various specific motor movements, research in the field of football, basketball, and baseball is in the lead. Previous studies in these sports have proven certain changes in the observed kinematic parameters due to higher physiological load [11–13].

Cortes et al. [14] found, in a sample of football players, that physiological load significantly affects the kinematic performance of the two-foot jump from running approach and sidestep-cutting task, as seen from the increase in knee and hip extension in pre- and post-load performance. In a study of the effects of a specific exercise imitating a 90-minute football match, a group of authors [15] found that increased physiological load reduced the shot accuracy by 25%, while passing accuracy did not decrease significantly, but passing speed decreased by 7.8%.

Shooting on the goal is considered one of the most important technical skills in competitive handball, because it is the main determinant of all actions that players take during the game [16]. Thus, a jump shot is the most common shooting technique in handball, with a total of approximately 74% of all shots during a game [4]. The kinematic parameters of the specific motor skills of shooting in the field of handball have been observed for many years. These parameters have been observed by various systems for monitoring the performance of shot movements. Much of the research is devoted to the analysis of the kinematic parameters of ground and jump shot performance and determining model values, as well as analysis of different shots on the goal [4,17–20]. A small amount of research can be found in the literature related to the effects of fatigue on the performance of the kinematic chain of ball shooting in handball. Akyuz et al. [21] researched if skeletal muscle fatigue affects shooting accuracy and ball speed in handball and found no significant differences. The authors used the following criteria for exhaustion: Maximum heart rate, plateau in  $VO_2\max$ , and a blood lactate concentration over 8 mmol/L. Thorlund et al. [22] determined the influence of fatigue on the mechanical properties of muscles and their neuromuscular activity on a sample of 10 top-level handball players during a simulated handball match. They found that reducing the ability to exert maximum force has a negative impact on performing fast movements such as acceleration, sprints, and lateral movements. In the situational conditions of a handball game, the reduced height of the jump affects the reduction in the possibility of blocking the opponent's shot, but also the shot over the block. The effect of local fatigue on the performance of upper extremities in handball jump shots was researched by Plummer and Gretchen [23]. No significant differences in kinematic performance were found in the observed shoulder and elbow segments before and after the fatiguing protocol with the local load caused by throwing medicine balls. The authors hypothesized that the fatigue protocol loading the entire kinematic chain could cause significant changes in the shooting technique. This was confirmed by another study, which determined the effects on the kinematics of the jump shot after the functional load by a progressive load test on the treadmill [24]. Although no significant changes in performance were found before and after the load protocol, there are still visible differences after the load protocol in the angular positions of the hip and torso segments during the jump shot.

The aim of this research was to determine whether the dynamic motor stereotype of movement, i.e., the shooting technique, is violated under the conditions of increased lactate concentration in the player's blood after a 30–15 intermittent fitness test, with hypotheses that there are significant differences in ball speed and shooting accuracy in jump shots on the goal before and after the influence of fatigue on the player.

## 2. Materials and Methods

### 2.1. Confirmation of the Ethics Committee

This research, undertaken in accordance with the Declaration of Helsinki, was approved by the Ethics Committee of the Faculty of Kinesiology, University of Zagreb. Prior to the start of the measurements, the respondents were given detailed information about the measurement protocol, the benefits, and the risks of the research. Moreover, upon arrival at the measurement venue, all respondents signed a consent form to participate in the research and to the use of their personal data.

## 2.2. Sample of Respondents

The sample of respondents (Table 1) consisted of 10 top-level handball players of the Premier Croatian Handball League (the highest-ranking competition in Croatia). The criterion for inclusion in the research was the level of playing, i.e., the players had to be members of the highest competition rank in the Republic of Croatia. The respondents did not have any health problems in the past year, which was the most important criterion for participation in the study. To avoid the impact of fatigue on the results of measurements, the respondents had a reduced training volume and intensity and did not play any matches two days prior to the testing. Additionally, the respondents were asked and warned not to take any stimulant before testing.

**Table 1.** Basic descriptive statistics of the respondents ( $n = 10$ ).

Variable	Valid N	Mean	Minimum	Maximum	Std. Dev.
Age (years)	10	19.32	18.02	21.96	1.21
Height (cm)	10	188.35	179.00	197.50	5.61
Weight (kg)	10	86.42	71.40	106.20	9.62
Fat mass (%)	10	12.44	6.60	20.90	4.15

## 2.3. Measurement Protocol

### 2.3.1. Materials

A Seca 213 portable stadiometer was used to measure body height, and a TANITA BC-545n (Bioelectrical Impedance Analysis) scale was used to measure weight and fat mass percentage. Heart rate (HR) was measured with a heart rate monitor (Polar H10, manufacturer: Polar, Kempele, Finland). Blood lactate level was measured with a portable lactate meter (Lactate Scout 3, manufacturer: SensLab GmbH, Leipzig, Germany). A blood sample was taken from the fingertip of the ring finger. Before taking the blood sample, the finger was cleaned with antiseptic solution and wiped with a liner. After puncture with a lancet, the first two drops of blood were discarded and the third was used for analysis. The blood sample was taken with Lactate Scout SENSOR (EKF diagnostics, Senslab, Code 14) and inserted into a portable lactate meter. The measurement of kinematic parameters was performed with the XSENS Awinda system for kinematic analysis. This system consisted of 17 wireless sensors attached with Velcro straps to subjects body (foot, lower leg, upper leg, pelvis, sternum, shoulder blade, upper arm, forearm, hand, and head). The Awinda wireless system records with a 60 Hz frequency, a 30 ms latency, and a 1000 Hz internal sampling rate. The MVN BIOMECH software package (MVN Studio 4.4, firmware version 4.3.1) was used to analyze the data from the kinematic suit. Previous research has determined the metric characteristics and the possibility of applying this system in sports games [25,26]. The speed of the ball (IHF Official ball size 3) was measured by a speed radar (Stalker ATS II, manufacturer: Stalker Sport, Texas, USA). A progressive discontinuous load test 30–15 [27] to failure was used for the fatigue protocol. The test was interrupted when the respondent failed to reach the 3 m zone three times in a row. Upon completion of the fatigue protocol, the respondents rated the subjective feeling of load by using a modified scale [28] from 0 to 10 degrees, known as the rating of perceived exertion (RPE). The shot was filmed with a high-speed camera (Panasonic GH5, 180 FPS). Video analysis was performed using the Kinovea (v.0.8.15) software package. Video analysis provided information about ball distance from goal frame, which was used to determine the shot accuracy. Previous research [5] has justified the use of this method for measuring accuracy.

### 2.3.2. Experimental Procedure Timeline

The measurement protocol included measuring anthropometric characteristics, warm-up, pre-load protocol measurement, fatigue test, and post-load protocol measurement. Upon arrival at the sports hall, the respondents' height, weight, foot length, ankle height, knee height, leg length, pelvis width, shoulder height, shoulder width, and arm span

were measured. These basic anthropometric characteristics were needed to describe the respondent sample and to calibrate the system for measuring kinematic parameters (Xsens technologies BV, Netherlands). Upon arriving at the sports hall, the respondents were asked to lay down on a mat and stay calm for 5 min, and resting heart rate was measured. Then, the data on lactate concentration were taken and measurement of the anthropometric characteristics was carried out. After this, the respondents began to warm up. The warm-up protocol lasted 15 min and consisted of running with tasks, dynamic warm-up with the ball, passing the ball in pairs, and 10 shots on the goal with a progressive increase in speed and strength of the shot. After the warm-up, the respondent started with the first series of (5) shots on the goal (Figures 1 and 2). Between each shot was a 30 s difference, which allowed subjects to return to the starting position and prepare for the next run-up and shot. Right-handed subjects were asked to aim at the right top corner, and left-handed subjects to aim at the top left corner of the goal. The lactate concentration was then measured again, after which the respondent proceeded with the fatigue protocol. Immediately upon completion of the test, the lactate concentration was measured again, and the respondents began a second series of (5) shots.

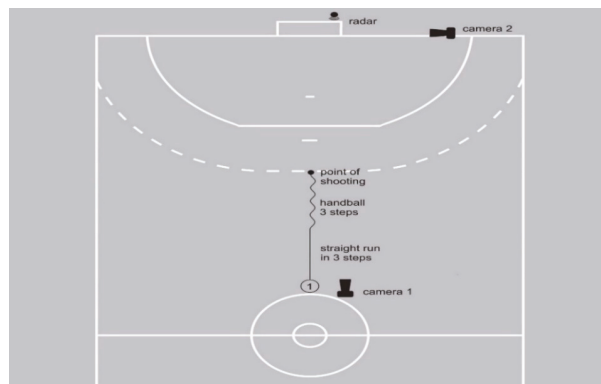


Figure 1. Sketch of the workplace.

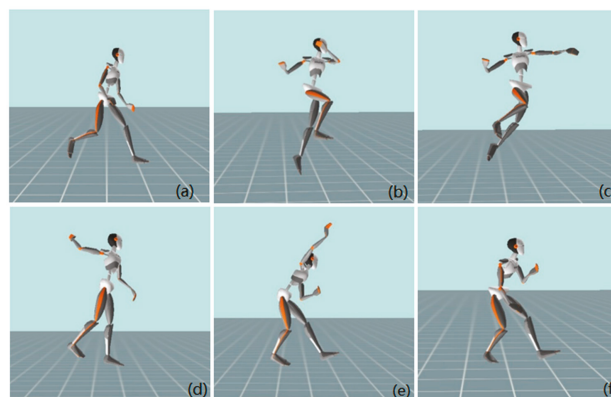


Figure 2. Kinogram of the final phase of the jump shot on the goal (a) final step; (b) take-off, (c) and (d) preparation for the shot; (e) shot, and (f) landing.

#### 2.4. Variables

In the run-up phase, the variable of maximum pelvis velocity (Pelvis\_Vmax) (m/s) was observed. In the take-off phase, the variables of the lower extremities (take-off foot) were observed: Maximum angular velocity in the ankle joint (Ankle\_AVmax) ( $^{\circ}$ /s), maximum angular velocity at the knee joint (Knee\_AVmax) ( $^{\circ}$ /s), and maximum angular velocity at the hip joint (Hip\_AVmax) ( $^{\circ}$ /s). In the ball shooting phase, the parameters of the dominant side of the body (the shooter's dominant arm) were observed: Maximum shoulder speed (Shoulder\_Vmax) (m/s), maximum upper arm speed (Upper\_arm\_Vmax), (m/s) maximum forearm speed (Forearm\_Vmax) (m/s), maximum hand speed (Hand\_Vmax) (m/s), maximum angular velocity at the shoulder joint (Shoulder\_AVmax) ( $^{\circ}$ /s), maximum angular velocity at the elbow joint (Elbow\_AVmax) ( $^{\circ}$ /s), and maximum angular velocity of the wrist joint (Wrist\_AVmax) ( $^{\circ}$ /s). After the ball shooting phase, the speed of the ball (Ball\_Vmax) (km/h) and the accuracy of the shot (ACCURACY) (cm) were measured.

#### 2.5. Data Processing Methods

G\* power analysis calculated the total ( $N = 66$ ) sample (number of shots) required to conduct the study with an error of  $p < 0.05$ , a statistical power of 0.8, an effect size of 0.25, and two groups. The Statistica v.13.05.0.17 (TIBCO software Inc) software package was used for statistical data processing. Basic descriptive parameters (mean, minimum, maximum, and standard deviation) were calculated for all observed variables. Multivariate analysis of variance (MANOVA) was used to test the statistical significance of the kinematic parameters of the shot before and after the fatigue protocol. Additionally, to test significant differences between each observed parameter, ANOVA for repeated measurements was used. A total of 100 shots were measured, of which 92 (46 before and 46 after the fatigue protocol) were used for statistical analysis. Due to errors in the performance of the motor movement (shooting technique) of the jump shot on the goal, eight shots were not analyzed.

### 3. Results

Prior to warm-up, the players were measured (with basic descriptive parameters in Table 2) for lactate concentration parameters ( $1.3 \pm 0.52$  mmol/L) and resting heart rate ( $83.70 \pm 7.81$  bpm). Moreover, the maximum heart rate was measured during the execution of shots before the fatigue protocol ( $163.00 \pm 10.91$  bpm), then during ( $195.40 \pm 8.30$  bpm) and after the fatigue protocol ( $171.40 \pm 7.04$  bpm) during the execution of shots under physiological load. The level of blood lactate concentration was measured after the first shots ( $2.07 \pm 1.16$  mmol/L) and after the fatigue protocol ( $11.88 \pm 3.33$  mmol/L). The maximum heart rate was 204 bpm, and the lactate concentration was 18.4 mmol/L. During the 30–15 fatigue protocol test, the respondents achieved an average running speed of  $18.20 \pm 1.01$  km/h. The minimum running speed was 16 km/h, while the maximum was 20 km/h. The fatigue test was rated, on average, as very severe activity (8.2 scores on a modified Foster subjective feeling scale [29]).

Table 3 shows the basic descriptive indicators and results of the ANOVA between the kinematic parameters of the shot before and after the fatigue protocol. The position of the hand during the performance of the jump shot was lower (2.18 cm) after the exhaustion protocol. The highest position of the shooter's hand was not significant ( $p = 0.39$ ). The speed of the run-up also decreased under the influence of fatigue (before, 5.45 m/s; after, 5.26 m/s). The difference in the reduction of the speed of the running start was significant ( $F = 5.66$ ;  $p = 0.02$ ). In the ball shooting phase, the angular velocities of the segments (shoulder, upper arm, forearm, and hand) were, on average, slower after the fatigue protocol. The highest maximum velocity in the ball shooting phase was reached in the wrist before the fatigue protocol (16.05 m/s). At this stage, a significant difference was obtained in the maximum velocity of the forearm ( $F = 5.85$ ;  $p = 0.02$ ) and the hand ( $F = 4.01$ ;  $p = 0.04$ ). Moreover, the angular velocities (shoulder, elbow, and wrist) were higher prior to performing the



fatigue test. The highest angular velocity was reached at the shoulder joint (2424.74 °/s). Significant differences were obtained in the shoulder joint ( $F = 5.39$ ;  $p = 0.02$ ) and wrist ( $F = 4.06$ ;  $p = 0.04$ ), while in the elbow joint, there was no difference between the observed groups. During the take-off phase, the mean values of angular velocities in the hip and knee joint were higher before the fatigue protocol, while in the ankle joint, they were higher after the protocol. The maximum angular velocity was reached in the ankle joint (1010.68 °/s). No significant differences between groups were obtained in the take-off phase. The speed of the ball differed significantly under the influence of fatigue ( $F = 5.42$ ;  $p = 0.02$ ). After the fatigue protocol, the speed decreased by an average of 2.76 km/h. The maximum achieved shot speed was 97.70 km/h, while the lowest was 68.60 km/h. The shot accuracy was, on average, lower (36.20 vs. 33.17 cm) after the fatigue protocol, but no significant difference was obtained.

**Table 2.** Basic descriptive parameters of the selected variables.

Variable	Valid N	Mean	Minimum	Maximum	Std. Dev.
R_HR	10	83.70	68.00	92.00	7.81
HR_max_1st	10	163.00	150.00	184.00	10.91
HR_max_2nd	10	171.40	158.00	179.00	7.04
30–15_HR_max	10	195.40	181.00	204.00	8.30
R_Lac	10	1.30	0.70	2.20	0.52
Lac_1st	10	2.07	0.90	4.50	1.16
30–15_Lac	10	11.88	7.20	18.40	3.33
30–15_V	10	18.20	16.50	20.00	1.01
30–15_BORG	10	8.20	7.00	9.00	0.63

Legend: RHR, resting heart rate; HR\_max\_1st, maximum heart rate after first set of 5 shots; HR\_max\_2nd, maximum heart rate after second set of 5 shots; 30–15\_HR\_max, maximum heart rate during the 30–15 test; R\_Lac, resting blood lactate concentration; Lac\_1st, blood lactate concentration after first 5 sets of shots; 30–15\_Lac, blood lactate concentration after the 30–15 test protocol; 30–15\_V, maximum speed reach in the 30–15 test; 30–15\_BORG, subjective evaluation of exhaustion after the 30–15 test.

**Table 3.** Basic descriptive indicators and ANOVA results for repeated measurements before and after the fatigue protocol for the observed variables.

Variable	N	Mean	Min	Max	Std. Dev.	–95%CI	+95%CI	F	p
Hand_H_pre	46	246.32	222.93	262.70	11.37				
Hand_H_after	46	244.14	216.38	267.05	13.05	–2.89	7.25	0.73	0.39
Pelvis_V_pre	46	5.45	4.32	6.35	0.39				
Pelvis_V_after	46	5.26	4.60	6.15	0.35	0.03	0.34	5.66	0.02 *
Shoulder_V_pre	46	4.58	3.43	6.19	0.63				
Shoulder_V_after	46	4.41	3.05	6.43	0.68	–0.09	0.45	1.69	0.20
Forearm_V_pre	46	11.38	10.08	12.75	0.66				
Forearm_V_after	46	11.06	9.82	12.47	0.61	0.06	0.59	5.85	0.02 *
Upper_arm_V_pre	46	6.12	5.02	7.10	0.51				
Upper_arm_V_after	46	6.03	5.12	7.11	0.51	–0.13	0.30	0.65	0.42
Hand_V_pre	46	12.49	10.33	16.05	1.28				
Hand_V_after	46	12.03	10.05	14.64	0.92	0.00	0.93	4.01	0.04 *
Shoulder_AV_pre	46	1378.09	635.38	2424.74	489.06				
Shoulder_AV_after	46	1150.72	323.98	2212.20	449.01	32.89	421.83	5.39	0.02 *
Elbow_AV_pre	46	1163.05	458.06	1846.28	318.73				
Elbow_AV_after	46	1159.14	483.28	2092.63	396.56	–145.12	152.94	0.00	0.96
Wrist_AV_pre	46	754.91	334.31	1452.54	256.92				
Wrist_AV_after	46	643.62	279.73	1484.94	272.33	1.63	220.96	4.06	0.04 *
Ankle_AV_pre	46	687.34	373.93	965.48	147.88				
Ankle_AV_after	46	700.11	393.48	1010.68	145.75	–73.58	48.05	0.17	0.68
Hip_AV_pre	46	420.97	226.30	548.86	53.21				
Hip_AV_after	46	415.71	337.20	525.70	38.93	–14.05	24.58	0.29	0.59
Knee_AV_pre	46	550.06	381.10	696.09	72.60				
Knee_AV_after	46	545.60	401.37	723.48	62.01	–23.50	32.44	0.10	0.75
Ball_V_pre	46	87.28	69.60	97.70	5.47				
Ball_V_after	46	84.52	68.60	93.70	5.89	0.41	5.12	5.42	0.02 *
Accuracy_pre	46	36.20	–5.65	106.62	25.71				
Accuracy_after	46	33.17	0.94	118.89	27.30	–7.96	14.02	0.30	0.59

\* Hand\_H, highest position of the shooter's hand; Pelvis\_V, maximum pelvis speed; Scheme 0.  $F = 2.38$ ;  $p = 0.00$ .

#### 4. Discussion

The aim of this study was to determine whether the dynamic motor stereotype of movement (shooting technique), i.e., the ball shooting speed and the accuracy of shooting, is violated during jump shots on the goal under conditions of an increased lactate concentration in the player's blood after the test. Significant differences in the kinematic parameters of the shot on the goal before and after the fatigue protocol prove a change in the complete kinematic chain of technique, i.e., motor knowledge of shooting on the goal. This is particularly true for the dominant upper extremities, where a significant difference was visible (at maximum forearm and hand speed), which also influences the speed of a shot.

The speed of running and the realization of the spatial-temporal advantage of the start and the start acceleration, i.e., the first step, is extremely important for the situational success in handball, both individual and team. Counterattack is the main determining factor of success in teams of the same rank in handball [30] and the most efficient method of scoring a goal with 88.23% of success rate. Accordingly, the change of rules in handball resulted in the concentration of the best teams on counterattack tactics (and also speeding up the game during the attack). The reaction time and the players' speed are the factors that allow them to gain an advantage over an opponent in the counterattack stage of the game, which has an impact on the final score. Consequently, the players' speed leads to easier realization, a better position to shoot on the goal, and a better position to pass to a player who is in a better position. Meanwhile, reducing speed during the fatigue phase reduces the situational efficiency of the individual player and the team. After the 30–15 test, aimed at fatiguing the players and achieving high blood lactates levels, a significant difference in the running approach speed (best performance speed for jump shot) was obtained ( $p = 0.02$ ). Only one player had a blood lactate level below 8 mmol/L, which was the reference value used in a similar study [21], where the authors tested if skeletal muscle fatigue affects shooting accuracy and ball speed in handball. By observing the speed parameters, these findings indicate a reduced individual situational efficiency and player abilities. Achieving the spatial-temporal advantage with regard to the running start speed and running was reduced in the fatigue phase. This consequently reduced the success of passing in a one-on-one game, in fast phases of the game such as fast transition, and individual breakthrough or counterattack, which is the fastest way to score a goal on an opposing team, since it does not have enough time to organize the defensive phase. Moreover, the reduced speed of the body affected the final speed of the shot, because the higher the speed of the body during the shot, the higher the speed of the shot on the goal [16,20].

The average values of the maximum velocity of the upper body segments that play a key role in the ball shooting phase (shoulder, upper arm, forearm, and hand), looking at the average values achieved in the tests, were reduced after the fatigue protocol. This was most pronounced in the significant differences before and after the protocol, where the velocities of the forearm ( $p = 0.02$ ) and the hand ( $p = 0.04$ ) were reduced after the fatigue protocol. A reduced speed of the forearm and hand has a negative effect on the speed of the shot, because they are the last parts of the body that are activated in the final phase of the shot on the goal [29]. Moreover, depending on the shooting technique (whether there is more emphasis on the wrist), a reduced speed in certain parts of the body such as the hand and forearm can vary. Herein, the influence of physiological load on changes in the observed kinematic parameters of the upper extremities was not frequently observed. However, Tripp et al. [31] observed the influence of physiological load on the positions of the joints in charge of throwing a baseball, which has similar biomechanical principles in throws to a handball shot and found that the sensorimotor system is impaired under the influence of fatigue. Uygur et al. [32] analyzed the effects of fatigue on the kinematics of free throws in basketball, where the respondents were subjected to a fatigue protocol with physiological load. They found that the maximum physiological load provoked by running sprints up to 30 m and two-foot jumps until failure in top-level seniors does not

influence the kinematic parameters of performance or accuracy. On the contrary, an impact on performance in younger basketball players was visible.

The angular velocities of the kinematic chain (shoulder, elbow, and wrist) were higher before the fatigue test, and significant differences were obtained at the maximum angular velocity in the shoulder joint ( $p = 0.02$ ) and the wrist ( $p = 0.04$ ). As a result of the stated reductions in the speed of the shoulder and wrist joints, and the reduction of the speed of the hand and forearm after the fatigue protocol, a significant difference in the speed of shooting on the goal was obtained. This is a logical sequence due to a reduction in the maximum speed in almost all parameters that affect the extension of the elbow, which is a determining factor for shooting speed [33,34]. In team handball, the ability to score largely depends on the ball speed and the accuracy of shoots. A quick shot directed toward the goal is considered an advantage to beat the goalkeeper and score a goal, which is the main objective of the game. This is the reason why coaches concentrate on training to increase shooting speed with an emphasis on optimizing shooting technique [35]. The obtained results indicate a decrease in the shot speed, which is one of the two significant abilities for scoring, while the shooting accuracy did not differ significantly before and after the fatigue protocol. The accuracy of the shot on the goal was reduced in the average values before and after the fatigue protocol, but it was not significant. The accuracy of the shot [36] is, in addition to the speed of the shot, one of the two parameters of success in scoring [4,5]. By reducing the accuracy and speed when shooting on the goal, the chances of successful team defense and individual goalkeeper defense increase. This is especially important under situational conditions or in positional shots, where the speed of the shot is not as significant for shooting success (a similar conclusion was obtained in a water polo study [37]). Examples include shooting from wing positions, shooting in a one-on-one situation with a goalkeeper, or shooting from a pivot's position. The other monitored variables such as maximum shoulder and upper arm velocity in the upper body and maximum angular velocity at the elbow joint, on average, decreased after the fatigue protocol. Segments of the lower body also indicate a decrease in the maximum angular velocity in the hip and knee joint in the average values, but the abovementioned upper and lower body parts did not experience significant deviations after the 30–15 test. Of all of the variables, only the maximum angular velocity of the ankle was increased, but not significantly.

The maximum speed in the shoulder joint, especially the flexion of the shoulder, together with the extension of the elbow and the deviation of the wrist, had the greatest influence on the speed of the shot. Ulnar deviation of the joint causes a higher speed ball rotation, which positively affects the quality of the ball flight, and thus the ball speed. Top-level handball players have an individual automated movement from the preparation to the realization of the shot, i.e., a dynamic motor stereotype of shooting on the goal, which they additionally emphasize in the final phase of the shot with their wrist [38]. Thus, top-level handball players, both with the influence of the load and the change in the kinematic chain of the entire motor knowledge, still maintain the accuracy of the shot, assuming that they avoid a foul attempt in defense [7]. Due to the reduction in shot speed from the aspect of situational efficiency, it is recommended for coaches to include, in the training process (technical or specific-situational shooting trainings), contents that are carried out with a physiological load similar to handball matches. This is an important factor in the training process in order to change and facilitate later adaptations of the entire motor knowledge (the players' movement) to fatigue that occurs during matches. Moreover, it is essential in the phases of the match when the physiological load is greatest, because it is in these phases that the winner of the match, competition, or championship is decided.

The strength of this research is reflected in the quality sample of respondents who play in the highest national rank of the competition. The research limit is manifested in the playing positions and situational conditions during the match. The speed of the shot in certain positions or situations in the game is not the most important element, but precision is, which comes to the fore (shooting from the wing position at a reduced angle or shooting

from the pivot's position under pressure or foul). However, each player used the same jump shot technique, which is used when shooting from outside positions without pressure or contact with defensive players.

## 5. Conclusions

High blood lactate levels affect changes in certain kinematic parameters during the performance of a jump shot in handball, as well as a change in the complete kinematic chain of performing the technique of shooting on the goal. This is especially pronounced in the upper extremities of the body in the form of a decrease in the speed of the forearm and hand and in the maximum angular velocities of the shoulder and wrist joints. The complete technique of shooting in handball is a sequence of successive actions of individual parts of the body. The final part of this series is the dominant arm and extension in the elbow, where the last phase ends with the forearm and hand. As a result of changes in the kinematic chain in the performance of the jump shot technique, the shot speed was reduced after the fatigue protocol. This can consequently affect situational performance, since the shot speed is one of the two significant parameters of successful scoring. Recommendations for coaches involve including technical and specific-situational shooting trainings with (high) physiological loads in their training process to simulate fatigue, which occurs in handball matches.

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

**Data Availability Statement:** Data available upon request.

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

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Article

# Sprint Variables Are Associated with the Odds Ratios of Non-Contact Injuries in Professional Soccer Players

Hadi Nobari <sup>1,2,3,4,\*</sup>, Elena Mainer-Pardos <sup>5,\*</sup>, Angel Denche Zamorano <sup>2</sup>, Thomas G. Bowman <sup>6</sup>,  
Filipe Manuel Clemente <sup>7,8</sup> and Jorge Pérez-Gómez <sup>2</sup>

<sup>1</sup> Department of Physical Education and Sports, University of Granada, 18010 Granada, Spain

<sup>2</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain; angeldenche@gmail.com (A.D.Z.); jorgepg100@gmail.com (J.P.-G.)

<sup>3</sup> Department of Exercise Physiology, Faculty of Educational Sciences and Psychology, University of Mohaghegh Ardabili, Ardabil 56199-11367, Iran

<sup>4</sup> Sports Scientist, Sepahan Football Club, Isfahan 81887-78473, Iran

<sup>5</sup> Health Sciences Faculty, Universidad San Jorge, Autov A23 km 299, 50830 Villanueva de Gállego, Spain

<sup>6</sup> Department of Athletic Training, College of Health Sciences, University of Lynchburg, Lynchburg, VA 24501, USA; bowman.t@lynchburg.edu

<sup>7</sup> Escola Superior Desporto e Lazer, Instituto Politécnico de Viana do Castelo, Rua Escola Industrial e Comercial de Nun'Álvares, 4900-347 Viana do Castelo, Portugal; Filipe.clemente5@gmail.com

<sup>8</sup> Instituto de Telecomunicações, Delegação da Covilhã, 1049-001 Lisboa, Portugal

\* Correspondence: hadi.nobari@gmail.com (H.N.); epardos@usj.es (E.M.-P.)

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**Abstract:** Significant evidence has emerged that a high volume of sprinting during training is associated with an increased risk of non-contact injuries in professional soccer players. Training load has been reported as a modifiable risk factor for successive injury in soccer. Sprint workload measures and non-contact injuries were recorded weekly in twenty-one professional soccer players over a one season period. Odds ratio (OR) and relative risk (RR) were calculated based on the weeks of high and low load of total distance (TD), high-speed distance (HSD), sprint distance (SPD), and repeated sprints (RS). The Poisson distribution estimated the interval time between the last injury and the new injury. The weeks with high-load levels increased the risk of non-contact injury associated with TD (OR: 4.1; RR: 2.4), HSD (OR: 4.6; RR: 2.6), SPD (OR: 6.9; RR: 3.7), and RS (OR: 4.3; RR: 2.7). The time between injuries was significantly longer in weeks of low-load in TD (rate ratio time (RRT) 1.5 vs. 4.2), HSD (RRT: 1.6 vs. 4.6), and SPD (RRT: 1.7 vs. 7.7) compared to weeks of high-load. The findings highlight an increased risk of non-contact injuries during high weekly sprint workloads. Possibly, TD, HSD, and SPD measured via a wearable inertial measurement unit could be modeled to track training and to reduce non-contact injuries. Finally, the interval time between the last injury and the new injury at the high-load is shorter than the low-load.

**Keywords:** football; injury risk; high load; external monitoring; performance; high-speed distance; global positioning system

## 1. Introduction

Soccer is considered an intermittent sport and it demands a wide variety of skills at high intensity with periods of rest or low intensity [1]. Professional soccer players have a congested calendar which usually requires playing successive matches with three days of recovery [2]. These players are exposed to high training load due to the poor recovery periods between years of long training and high match frequencies. These competitive demands could increase injury risk and reduce performance; therefore, they could be detrimental to team success [2–4].

In recent times, there has been an increase of high-speed distance (HSD) and running during competitive soccer matches [5]. Moreover, the ability to produce HSD seems to be a



significant quality for performance [6]. Soccer players need improved development of high speed and sprint running ability to gain advantage in attacking and defensive situations [7]. Malone et al. [8] reported that exposing players to large and rapid increases in HSD and sprint distances (SD) increased the odds of injury. Bowen et al. [9] demonstrated that three weekly accumulations of accelerations  $>9254$  were related to an elevated risk of non-contact injury. Nevertheless, from an injury perspective, more studies are warranted that allow coaches to understand the dose-response of HSD within training environments.

Micro-technology use, such as global position systems (GPS) which measure external training loads of players, has become prevalent in professional soccer [10]. The tool is used to identify the activity profile of players during training and matches [10]. Also, GPS is able to quantify the distance covered along with high acceleration efforts, short duration, high velocity sprints and repeated sprint (RS) exercise bouts [11]. Angelidis et al. [12] examined the link between GPS variables and non-contact injuries in soccer players in a recent systematic review. They found eight variables, total distance (TD), high-speed running, total load, accelerations, decelerations, new body load, meter per minute, and sprinting, that deserve particular attention when monitoring soccer players' external load for the purpose of injury prevention. At each training and competition, players are exposed to a given workload [13]. An inappropriate workload during these periods can increase injury risk. An increased acute GPS-derived workload in team sports produced a higher association with injury risk [14,15]. In professional soccer players, when the chronic loads were low with very high acute spikes, non-contact injury risk increased [16]. However, more studies are needed to consider the effect of the accumulation load to establish stronger conclusions between GPS training load measurement and non-contact injury in professional soccer players.

Time loss due to injury is one of the bigger problems during soccer players' careers [3]. Most injuries occur in the lower extremities [17]. In addition, injuries of professional soccer players have a considerable impact on the sports industry [18], the recovery and rehabilitation of players is associated with a considerable cost [19]. A non-contact injury is an injury with no physical contact with other players, and a considerable proportion of injuries in soccer are non-contact [17]. Talukder et al. [20] proposed that the most relevant characteristics for predicting injuries are the average speed, the number of past competitions played, the average distance covered, the number of minutes played to date, and the average field goals attempted. Hence, researchers and coaches are interested in reducing the likelihood of injuries to their soccer players and consequently, injury forecasting is engaging more interest within the sporting environment.

Training load has been reported as a modifiable risk factor for successive injury in soccer [21]. Gabbet and Ullah [22] observed a correlation between high intensity running and injury risk during training sessions. Within elite soccer players, maintaining heart rate (HR) above 85% HRmax during training was associated with increased risk of injury [23]. Malone et al. [8] recently showed that higher chronic loads and higher aerobic conditioning appear to offer a better protective effect against injury for professional soccer players, and they should be considered mediators of injury risk. It is important for practitioners to understand the optimal training load at which adaptation occurs without raising the risk of injury. However, there are no studies that have investigated the relationship between non-contact injury incidence and sprint variables through GPS within professional soccer players.

Given the need for coaches and practitioners to know the relationship between GPS-derived workload and non-contact injury risk in professional soccer players, the aims of the current study were to investigate, (i) the relationship between the total distance, HSD, sprint distance (SPD), and RS with non-contact injuries in professional players throughout a full soccer season; (ii) the injury risk associated between high- versus low-load level for each of the aforementioned parameters with odds ratios (OR) and relative risk (RR), respectively; and (iii) ultimately, a Poisson test to obtain lambda values (i.e., number of

injuries per week for each level of cases listed), with the predicted time between injuries to new injury to calculate the rate ratio.

## 2. Materials and Methods

### 2.1. Participants

Twenty-one soccer players [age (year),  $28.3 \pm 3.8$ ; height (cm),  $181.2 \pm 7.1$ ; weight (kg),  $74.5 \pm 7.7$ ] from a professional team in the Persian Gulf Pro League were analyzed during a full season (2018–2019 years). These players had more than 8 years of professional playing experience. The criterion for entering the participants' information into the analysis was that they had participated in at least three training sessions per week. The criterion for excluding participants' data in the analysis was that data was not available for 2 consecutive weeks or they had not participated for 2 consecutive weeks in the training. Also, goalkeepers' data were eliminated in this study. Based on a power analysis of results from previous studies, we believed a sample size of 21 was adequate to reduce the risk of a Type 2 error [8,22]. This research was conducted by the training coaches of the club after approval with the relevant authorities and the head coach in the club. Prior to commencing the study, the approval of the research ethics committee from the University of Isfahan (IR.UI.REC.1399.064) was also received. All players were informed of the purpose of the study before signing the informed consent. All stages of this study were carried out according to the human studies in the Helsinki Declaration.

### 2.2. Study Design

The design of the prospective cohort study was performed in a full season during the Persian Gulf Pro League and knockout tournament. External load monitoring was performed by GPS (GPSPORTS systems Pty Ltd, Model: SPI High Performance Unit (HPU), Canberra, Australia) at each training and match session over the whole season. All non-contact injuries (that is, occurring without contact with foreign material or athletes) were recorded during the season. During the full season, 7-weeks congested play (i.e., two or more matches within 7-days), 30-weeks non-congested play; 44 matches, 11-weeks with no competitions, 200 training sessions, and 14,126.9 minutes of time play and sessions were held. Almost all training sessions and competitions were held on a natural grass field. The team between weeks 26 to 32 (mid-season break) was at the International Camp in Turkey. The running variables recorded, during the season, for this study included: TD, HSD ( $18\text{--}23 \text{ km/h}^{-1}$ ) [24,25] SPD, and meters of RS. Afterward, each variable was divided into two levels, upper and lower, and subsequently, the relationship between the variables was measured.

### 2.3. Procedure

#### 2.3.1. Wearable Inertial Measurement Unit Receiver

All players' activities in training sessions and matches were recorded with GPSPORTS systems Pty Ltd, Canberra, Australia. The GPS-based tracking systems for professional athletes, model SPI HPU features included: 15 Hz position GPS, distance, and speed measurement; accelerometer: 100 Hz, 16 G Tri-Axial-Track impacts, accelerations, and declarations as well as data source body load (BL); Mag: 50 Hz, Tri-Axial; dimensions:  $74 \text{ mm} \times 42 \text{ mm} \times 16 \text{ mm}$ ; water resistance and data transmission: infra-red and weighed 56 g. Previous studies have shown that the GPS unit had very high accuracy and demonstrated excellent validity and inter-unit reliability [26]. All data were collected during training and match sessions with favorable weather and GPS satellite status.

#### 2.3.2. Data Collection

Data collection was completed as in previous studies [27,28]. As in during pre-session we placed upright tracking units in the pouch of the manufacturer provided belt, then ensured the green lights (GPS tracking) and red lights (heart-rate tracking) were flashing.

Post-session, tracking units were collected from players and placed on the docking station. Data from the units were automatically downloaded then deleted from the units to prepare for the next session. After 10 min, the units turned off automatically. The GPS system was tuned to the default SPI IQ Absolutes in this study.

### 2.3.3. Sprint Variables Calculated

TD was calculated as the mean number of weekly meters run by the team's players over the 48 weeks of the season.

HSD was calculated as the mean of weekly meters run at high speed by the team's players over the 48 weeks of the season.

SPD was calculated as the mean number of weekly meters run by the team's players during the 48 weeks of the season.

RS was calculated as the mean weekly meters of repeated sprints performed by the team's players throughout the 48 weeks of the season.

TD level division was the difference between "high-load" and "low-load" weeks according to the average weekly TD of the team. "High-load" was defined as  $TD \geq 21,900$  and "low-load" was defined as  $TD < 21,900$ , both in meters. The cut-off point was established in the TD value, in which a greater difference was found between the mean of injuries between weeks of higher and lower load (clusters with more than 11 weeks), having previously ordered the weeks in accordance with higher to lower TD.

- I. Mean of injuries during the 12 weeks of highest TD–Mean of injuries during the rest of the weeks.
- II. Mean of injuries during the 13 weeks of higher TD–Mean of injuries during the rest of the weeks.
- III. Mean of injuries during the 36 weeks of greater TD–Mean of injuries during the rest of the weeks.

HSD level division was the difference between "high-load" and "low-load" weeks according to the average weekly HSD of the team. "High-load" was defined as  $HSD \geq 288$  and "low-load" was defined as  $HSD < 288$ , both in meters. The cut-off point was established at the HSD value in which a greater difference was found between the mean of injuries between weeks of higher and lower load (clusters with more than 11 weeks), having previously ordered the weeks in accordance with higher to lower HSD.

- I. Mean of injuries during the 11 weeks of highest HSD–Mean of injuries during the rest of the weeks.
- II. Mean of injuries during the 12 weeks of higher HSD–Mean of injuries during the rest of the weeks.
- III. Mean of injuries during the 36 weeks of higher HSD–Mean of injuries during the rest of the weeks.

SPD level division was the difference between "high-load" and "low-load" weeks according to the average weekly SPD of the team. "High-load" was defined as  $SPD \geq 1601$  and "low-load" was defined as  $SPD < 1601$ , both in meters. The cut-off point was established at the SPD value in which a greater difference was found between the mean of injuries between weeks of higher and lower load (clusters with more than 11 weeks), having previously ordered the weeks in accordance with higher to lower SPD.

- I. Mean of injuries during the 11 weeks of highest SPD–Mean of injuries during the rest of the weeks.
- II. Mean of injuries during the 12 weeks of higher SPD–Mean of injuries during the rest of the weeks.
- III. Mean of injuries during the 36 weeks of higher SPD–Mean of injuries during the rest of the weeks.

RS level division was the difference between "high-load" and "low-load" weeks according to the average weekly RS of the team. "High-load" was defined as  $RS \geq 60$  and

“low-load” was defined as  $RS < 60$ , both in numbers. The cut-off point was established at the RS value in which a greater difference was found between the mean of injuries between weeks of higher and lower load (clusters with more than 11 weeks), having previously ordered the weeks in accordance with higher to lower RS.

- I. Mean of injuries during the 11 weeks of highest RS–Mean of injuries during the rest of the weeks.
- II. Mean of injuries during the 12 weeks of higher RS–Mean of injuries during the rest of the weeks.
- III. Mean of injuries during the 36 weeks of higher RS–Mean of injuries during the rest of the weeks.

#### 2.3.4. How to Record and calculate Injury

Information on injuries was updated daily by the team’s specialized medical staff. All injuries were recorded by type, location of the injury, and timing of injury based on previous studies. The information used for the injuries was as follows:

1. The number of registered injuries was the total number of non-contact injuries per week for the team over the 48 weeks of the season.
2. Weekly injury recorded the existence or not of a non-contact injury in each of the 48 weeks of the season.

#### 2.4. Statistical Analysis

IBM SPSS 25 and R Studio 3.6.2 (Statistical Computing, Vienna, Austria). were used for the statistical analyses. A descriptive statistical analysis was performed, indicating the mean values and SD of the “high-load” and “low-load” levels for the variables “TD”, “HSD”, “SPD”, and “RS”, as well as the total values. Non-parametric Mann–Whitney U tests were used to compare the median of the load levels of the previous variables, checking the existence of statistically significant differences between them. A test of normality, Kolmogorov–Smirnov, verified that the variable “Number of injuries”, did not follow a normal distribution. Additionally, a descriptive analysis of the number of injuries produced in the weeks of high and low load of each one of the variables was completed, as well as the calculation of the means of each one of them, both for the two levels of load, as well as for the total. In order to detect statistically significant inter-group differences between the means of injuries at the “high-load” and “low-load” levels of the “TD”, “HSD”, “SPD”, and “RS” variables, non-parametric tests were used, considering, as factors, the load levels of each variable. A contrast of proportions determined if significant differences existed between the levels of “high-load” and “low-load” of each variable and the weeks with injury. To estimate the risk of having a high-load level compared to a low-load level, respectively, of each variable, OR and RR were calculated, in addition to the respective confidence intervals (CI) 95%. Finally, the variable “Number of injuries” followed a Poisson distribution, allowing the performance of a Poisson test, obtaining the lambda values (average number of injuries per week for each level of load for each variable), and the expected time until a new injury occurred, once one had occurred. Possible significant differences between load levels were examined via calculating Rate Ratios, as well as respective CI 95%. The level of significance was set at  $p < 0.05$  and  $p < 0.001$  for all stages.

### 3. Results

A descriptive analysis of the high- and low-load levels for the TD, HSD, SPD, and RS variables are presented in Table 1. Statistically significant differences between “high-” and “low-” load levels were observed in all variables.

Table 2 details the relation between sprint variables with non-contact injuries. Mean injuries were significantly higher in the high load weeks compared to the low load weeks for TD, HSD, SD, and RS.

Significant differences were found ( $p < 0.05$ ) in the proportion of injury-free weeks between high- and low-load weeks in all variables studied. The OR and RR of producing some injury without contact was significantly higher in the weeks of high load compared to the weeks of low load in each one of the variables of interest. Similarly, significant RR were found for all variables except for the “RS” variable (Table 3).

**Table 1.** Descriptive information of sprint variables based on high- and low-load levels.

Variables	High-Load Median (IQR)	Low-Load Median (IQR)	<i>p</i>	Total Median (IQR)
TD (m)	23,757 (21,073–26,367)	20,006 (21,667–25,828)	$\leq 0.001$ ***	21,595 (19,211–25,046)
HSD (m)	360.2 (289.5–387.8)	207.7 (157.9–235.2)	$\leq 0.001$ ***	288.2 (210.4–361.7)
SPD (m)	1962 (1854–2473)	1429 (1189–1590)	$\leq 0.001$ ***	1845 (1518–2182)
RS (m)	77.6 (65.1–85.2)	48.6 (37.9–57.6)	$\leq 0.001$ ***	65.5 (55.9–81.0)

IQR: Interquartile range; m: meters; Total distance (TD): High load (weeks with TD  $\geq 21,900$  m); Low load (weeks with TD  $< 21,900$  m); High-speed distance (HSD): High load (weeks with HSD  $\geq 288$  m); Low load (weeks with HSD  $< 288$  m); Sprint distance (SPD): High load (weeks with SPD  $\geq 1601$  m); Low load (weeks with SPD  $< 1601$  m); Repeated sprints (RS): High load (RS  $\geq 60$ ). Low load (RS  $< 60$ ); *p* (*p* value); \*\*\* ( $p < 0.001$ ).

**Table 2.** Relation between TD, HSD, SPD, and RS with non-contact injuries (mean).

Variables	High-Load			Low-Load			<i>p</i>	Total		
	Injuries	Weeks	M	Injuries	Weeks	M		Injuries	Weeks	M
TD (m)	15	23	0.65	6	25	0.24	0.017 *	21	48	0.44
HSD (m)	16	25	0.64	5	23	0.22	0.013 *	21	48	0.44
SPD (m)	19	33	0.58	2	15	0.13	0.013 *	21	48	0.44
RS (m)	18	32	0.56	3	16	0.19	0.034 *	21	48	0.44

m: meters; Total distance (TD): High load (weeks with TD  $\geq 21,900$  m); Low load (weeks with TD  $< 21,900$  m); High-speed distance (HSD): High load (weeks with HSD  $\geq 288$  m); Low load (weeks with HSD  $< 288$  m); Sprint distance (SPD): High load (weeks with SPD  $\geq 1601$  m); Low load (weeks with SPD  $< 1601$  m); Repeated sprints (RS): High load (RS  $\geq 60$ ). Low load (RS  $< 60$ ); M: mean; *p* (*p* value); \* ( $p < 0.05$ ).

**Table 3.** Injury risk related to different load levels and sprint variables with OR and RR (mean).

Variables	High Load		Low Load		OR	CI 95%		RR	CI 95%	
	Injury (No Injury)	Total	Injury (No Injury)	Total		Min	Max		Min	Max
TD (m)	13 (10)	23	6 (19)	25	4.1	1.2	14.1	2.4	1.1	5.2
HSD (m)	14 (11)	25	5 (18)	23	4.6	1.3	16.3	2.6	1.1	6.0
SPD (m)	17 (16)	33	2 (13)	15	6.9	1.3	35.5	3.7	1.0	14.6
RS (m)	16 (16)	32	3 (13)	16	4.3	1.0	18.2	2.7	0.9	7.8

m: meters; n: numbers; Total distance (TD): High load (weeks with TD  $\geq 21900$  m); Low load (weeks with TD  $< 21900$  m); High-speed distance (HSD): High load (weeks with HSD  $\geq 288$  m); Low load (weeks with HSD  $< 288$  m); Sprint distance (SPD): High load (weeks with SPD  $\geq 1601$  m); Low load (weeks with SPD  $< 1601$  m); Repeated sprints (RS): High load (RS  $\geq 60$ ). Low load (RS  $< 60$ ); Injury (weeks with injuries). No injury (weeks without injuries). Total (total weeks). OR (Odds Ratio) RR (relative risk). CI95% (confidence interval); Min (minimum); Max (maximum).

Ultimately, it was observed that the time between injuries was reduced in weeks of high-load and longer in weeks of low-load in the four variables studied, although significant differences were found in all variables, except in the variable RS. Similarly, the rate ratios were significant in all variables for the expected weeks between injuries except for the variable RS (Table 4).

**Table 4.** Relationship between injuries and different levels of load to find the expected time until new injuries (mean).

Variables	High Load		Low Load		<i>p</i>	Rate Ratio	CI 95%	
	$\lambda$	Expected Time Injury	$\lambda$	Expected Time Injury			Min	Max
TD (m)	0.65	1.5	0.24	4.2	0.046 *	2.7	1.0	8.6
HSD (m)	0.64	1.6	0.22	4.6	0.029 *	2.9	1.0	10.3
SPD (m)	0.58	1.7	0.13	7.7	0.033 *	4.3	1.0	38.2
RS (m)	0.56	1.8	0.19	5.3	0.067	3.0	0.9	15.9

Poisson: m: meters; n: numbers; Total distance (TD): High load (weeks with TD  $\geq$  21,900 m); Low load (weeks with TD < 21,900 m); High-speed distance (HSD): High load (weeks with HSD  $\geq$  288 m); Low load (weeks with HSD < 288 m); Sprint distance (SPD): High load (weeks with SPD  $\geq$  1601 m); Low load (weeks with SPD < 1601 m); Repeated sprints (RS): High load (RS  $\geq$  60). Low load (RS < 60); *p* (*p* value); \* *p* < 0.05; CI 95% (Confidence interval); MIN (minimum); MAX (maximum); Rate ratio (Expected time injury low load/Expected time injury high load).

#### 4. Discussion

This study investigated the relationship between GPS-derived workload and non-contact injuries in professional soccer players. The present data highlight that the weeks of high-load levels increase the risk of non-contact injury within soccer players. Based on our results, TD, HSD, and SPD variables could potentially track training and may allow exercise prescription to reduce non-contact injuries. Finally, according to the rate ratios calculated, the interval time between the last non-contact injury and the new injury during high-load training in TD, HSD, and SPD) is shorter than during low-load training.

The relationship between non-contact injuries and different levels of load to find the expected time until a new non-contact injury was presented via ORs and RRs. All variables were significant except for the RS variable in RR (Table 3). TD and HSD were found previously to be the best rates of non-contact injuries [12]. In the current study and in previous studies, increased injury occurred when ORs and RRs were greater than 1 [12,29]. TD (OR: 4.1, RR: 2.4) and HSD (OR: 4.6, RR: 2.6) were significantly different between high- and low-load weeks. Therefore, participants had a higher odds and risk of a non-contact injury during high-load weeks compared to low-load weeks. Previous studies did not investigate SPD and RS, although these have been found to be associated with non-contact injury occurrence [12]. In the current study, when players covered SPD  $\geq$  1601 m during a week, they were at significantly higher risk of injury compared to weeks with SPD < 1601 m (OR: 6.9; RR: 3.7 m; *p* < 0.05). However, RS reported a significant OR (4.3; *p* < 0.05) but no significant RR (2.7; *p* > 0.05). Therefore, TD, HSD, and SPD variables could potentially be associated with non-contact injuries and the expected time until new injury and may help practitioners and coaches adjust training load in an effort to prevent non-contact injury.

Our study is one of the first to investigate the effects of GPS-derived workload on subsequent non-contact injury risk in an elite cohort of soccer players. We observed that the weeks in which players reached TD < 21,900 m, HSD < 288 m and SPD < 1601 m, they reduced injury risk compared to higher week loads (OR: 4.1 to 6.9). In addition, our findings suggest that weeks in which players repeated <60 sprints, they were at reduced injury risk compared to high-load weeks (OR: 4.3). The current data suggest that low load exposure to sprint variables can protect professional soccer players from subsequent non-contact injury risk. These results could provide coaches and practitioners with initial guidelines for optimal workload to reduce non-contact injury occurrence.

Previous literature has found that moderate and higher chronic training loads can offer a protective effect against lower limb injury risk for team sport players [8,28]. Malone et al. [30] observed that players with moderate exposures to maximum velocity (>6–10) were at reduced injury risk compared to players who experienced lower (<5) exposures (OR: 0.24). Also, the authors [30] found that players with higher 21-day chronic loads ( $\geq$ 2584 AU) completed increased high-speed and sprint running distances that indicated a protective effect against injury (OR: 0.24–1.22). The reason for these observations may be because the

players are usually exposed to a chronic training load period. Hence, they are used to tolerating high-speed running workload which reduces injury risk. In contrast, Malone et al. reported that elite soccer players were at increased risk of injury when they experienced high one-week cumulative training loads ( $\geq 1500$  to  $\leq 2120$  AU) [8]. Gabbet and Ullah [22] demonstrated that greater amounts of HSD are associated with an increased injury risk of lower body soft-tissue injury in elite rugby players (RR: 2.7). These findings can be explained by higher acute loads that are associated with an increase in fatigue status in players and resultant increases in injury risk [21]. In this regard, controversy still exists regarding the impact of sprints in non-contact injuries and further studies are needed in the elite soccer environment.

From a performance point of view, careful thought should be taken to understand and apply the present findings to elite performance soccer. Our data suggest that reducing the amount of sprinting in professional soccer training could offer a protective effect against non-contact injury. In this context, a fine balance exists between training load restriction (i.e., preventing injury) and increased training loads (i.e., improving performance) [4,8,22]. Consequently, keeping in mind the need for an appropriate stimulus to improve performance, we used the current data to vary sprinting loads during the soccer season. Our data suggest that players will be exposed to increased risk of non-contact injury when the amount of TD, HSD, SPD, and number of RS is higher. Planning a decrease of mean sprint variables in some high-load weeks may offer the balance between injury prevention and performance enhancement. The finding could be an important consideration to correctly manipulate workloads during the season, not only to reduce non-contact injury but also to enhance physical performance within professional soccer players.

A method of measuring internal training load, such as rating of perceived exertion (RPE) using the category ratio scale (CR10-scale) per session [31], could be used to assess how the players are coping with training loads every day. This method has been shown to be the most valid indicator of exercise intensity [32]. The collection of weekly GPS (external load) and session-RPE (internal load) variables allows the calculation of chronic training loads [2], enabling reduction of future loads based on these variables. Previously, Gallo et al. [33] demonstrated moderate to very large associations between session-RPE and both HSD ( $r = 0.51$ ) and TD ( $r = 0.81$ ) in team sport athletes. Therefore, including this parameter in future research could assist practitioners and coaches in finding the appropriate stimulus and balance between injuries and performance.

Recent studies observed that non-contact injuries had a high frequency of re-injury and persistent complaints after return to soccer competition [3,34]. According to the results of the rate ratios, the interval time between the last non-contact injury and the new injury at the high-load in TD, HSD, and SPD is shorter than the low-load (Table 4). Soccer injuries are defined by an inter-individual variability of flexor and extensor muscle performances [35]. One of the major risk factors that has been identified is strength asymmetry, and the asymmetry index has been considered as a valid and useful tool to detect players at high injury risk (e.g., 4-fold in players with  $>10\%$  asymmetry) of lower extremity injury [36]. Strength asymmetry has been considered in relation to movement speed in team-sport athletes [37]. Therefore, the major number of sprints during high load weeks may induce an increase of strength imbalances in the lower limbs, and a greater predisposition to injury may be possible. Consequently, inter-limb asymmetry tests and strength training programs may allow a preventive approach to reduce the risk of re-injury in professional soccer.

#### 4.1. Limitations

Several limitations are important to mention from the current study. Injury risk in soccer players can also be attributed to multiple factors such as previous injury, perceived muscle fatigue, nutrition and hydration, mood, sleep ratings, and physiological stressors, none of which were included in this study [18,27]. Research incorporating objective measures of GPS with RPE-values may provide additional insight into the training load–injury relationship. In addition, the present investigation was developed in one professional

soccer team during one season. Consequently, our results cannot be directly extrapolated to other sports, teams, or across multiple seasons. Therefore, we recommend further studies for better understanding of the workload–injury relationship in elite soccer.

#### 4.2. Practical Applications

The current study is the first to provide an indication of how players' weekly training load is associated with non-contact injuries in professional soccer players. Team soccer staff should measure weekly the internal and external load of players to plan and implement optimal trainings that improve their performance and reduce their injury risk. Given that these findings suggest that a high load of sprints increases the risk of sustaining non-contact injuries, attempting to adjust training load for sprint variables during high load weeks is recommended. However, this observation requires further investigations. One possibility to achieve this in practice is to restrict the amount of sprinting and prescribe stable and consistent weekly loads during the season to prevent any spikes in acute workload. Our results also suggest that TD, HSD, and SPD measured via GPS could be modelled to track training and to reduce non-contact injuries. Nevertheless, further studies are recommended to improve the accuracy of these variables in reducing non-contact injuries. Finally, the findings may promote an evidence-based approach for coaches and practitioners in planning and monitoring weekly training load thresholds to reduce fatigue during soccer participation and consequently, prevent non-contact injuries.

#### 5. Conclusions

The results of this study demonstrated that the high volume of sprinting during high-load weeks is associated with non-contact injury occurrence. In addition, TD, HSD, and SPD of GPS variables could track training and may allow exercise prescription to reduce non-contact injuries. Finally, the interval time between the previous injury and a new injury during high load training is shorter than during low load training. The current data provide an important contribution that may be valuable to support the decisions of coaches and practitioners when they have to choose the best GPS variables to observe non-contact injuries, to reduce the amount of sprinting during professional soccer seasons, and to adjust training loads for sprint variables. However, they could also contemplate the consequences of reducing training loads during the season on playing performance.

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Article

# Relationship between Sprint Capacity and Acceleration of Wrists in Wheelchair Basketball Players: Design and Reliability of a New Protocol

Amelia Ferro <sup>1,\*</sup>, Javier Pérez-Tejero <sup>2</sup>, Guadalupe Garrido <sup>2</sup> and Jorge Villacieros <sup>3</sup>

- <sup>1</sup> Department of Sports, Faculty of Physical Activity and Sport Sciences, Universidad Politécnica de Madrid, 28040 Madrid, Spain
- <sup>2</sup> Department of Health and Human Performance, Faculty of Physical Activity and Sport Sciences, Universidad Politécnica de Madrid, 28040 Madrid, Spain; j.perez@upm.es (J.P.-T.); lupe.garrido.pastor@upm.es (G.G.)
- <sup>3</sup> Physical Education and Adapted Sports, Campus La Salle, 28023 Madrid, Spain; jvrodriquez@lasallesagradocorazon.es
- \* Correspondence: amelia.ferro@upm.es; Tel.: +34-910678025

**Abstract:** The application of new technologies in wheelchair basketball (WB) is important for the advancement and improvement of athletic performance. The purposes of this study are twofold: (a) to develop a methodological design in order to assess WB players' performance, using wireless inertial measurement units (WIMU<sup>®</sup>) and a laser system (BioLaserSport<sup>®</sup> with computer vision), in a 20 m sprint test on court and (b) to assess bilateral symmetry as a performance indicator and for injury prevention purposes, the study of which in previous research is unknown. For both aims, the relation of the acceleration of the players' wrists to the speed achieved by the player in the wheelchair was explored. Ten elite WB players participated in an on-court 20 m sprint test during real training. BioLaserSport<sup>®</sup> with computer vision was used to assess the average velocity (Va) and maximum velocity (Vmax) of the WB players, and two WIMU<sup>®</sup> were used for the total acceleration (AcelT) of the players' wrists. A very high correlation was obtained in the assessment of the Va (0.97) and AcelT of both wrists (0.90 and 0.85). There was a significant relationship between the average AcelT of the dominant wrist and the Va on-court sprint velocity ( $p < 0.05$ ). Two players did not show good wrist symmetry. In conclusion, a new methodological protocol was developed, making it possible to assess the bilateral symmetries in elite WB players in on-court real training and the relation between the acceleration of players' wrists and players' wheelchair speed. Coaches can use this protocol to assess performance or for injury prevention, as it shows very good reliability, with high ICC values.

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**Keywords:** performance; kinematics; laser; computer vision; inertial device; IMU

## 1. Introduction

The application of new technologies in wheelchair basketball (WB) is important for the advancement and improvement of athletic performance. Initially, studies were carried out under laboratory conditions [1–5], but in recent years, real training conditions have been used [6–9].

In WB, the ability to sprint from a standstill condition is crucial. The sport-specific movement demands, established as starting, sprinting, braking, turning (pivoting) and blocking, are specifically related to the use of the wheelchair [10]. In addition, Mason et al. [11] have defined stability, initial acceleration, maneuverability and sprinting as critical areas for successful WB performance. Despite the importance of sprint capacity in this sport, it still has not been investigated in depth [3].

In the literature, several tests have been described for assessing sprint performance in WB players [7,9,12,13]; however, how the movements of the upper limbs can influence the propulsion cycle in gaining speed in the wheelchair requires further attention [14].

According to Rao et al. [15], the wrist participates very actively in the wheelchair propulsion cycle, from extension and radial deviation to flexion and ulnar deviation. For this reason, it is important to determine whether the acceleration produced by the wrist influences the increase in the players' speed in the wheelchair, since it is the closest joint to the handrim.

Acceleration has been studied through inertial measurement systems (IMU), where its reliability and validity have been demonstrated [16–18]. This technology makes it possible to systematically monitor the movements of the human body when applied to competitive sports [19–21] and to obtain a 3D orientation of the body segments [16]. Bergamini et al. [6] presented a method to quantify the characteristics of WB players in a 20 m sprint test using IMU. Their results confirmed the importance of adequate propulsion symmetry as an indicator value related to athlete sport performance and potential injuries. It should be noted that this indicator has also been studied by other authors [22,23]. However, an accurate description of the protocol was not made for its measurement, and the use of a stopwatch to calculate the speed was not reliable [6]. In our opinion, it is necessary to develop an innovative methodology that combines force moments of the wrists along wheelchair propulsion, measured through isokinetic dynamometers, with sport-specific demands. In this regard, Wei et al. [24] explored wrist kinematics during wheelchair propulsion in a non-sportive population, finding seat height to be a factor influencing handrim contact time (the higher the seat, the lower the handrim contact time, wrist extension angles and range of motion). However, no studies on wheeled sport, such as WB, were found. Furthermore, to our knowledge, no study has taken into account the acceleration generated by the wrist joints in WB-specific environments. This joint is the closest to the propulsion ring in the wheelchair, so it is probably the one that suffers the most impact [24], especially when performing a sprint. For this reason, it is necessary to design a protocol that places measurement systems on the wrists in order to relate the acceleration of these joints to the speed achieved by the player during the sprint and detect possible bilateral symmetries that may occur during the sprint. This should also be analyzed taking into account functional classification, as it is a factor influencing players' wheelchair configuration and seating height [10,24].

This gap in the previous literature should be addressed by a combination of technological developments that bring innovative options for WB players and coaches, as sprinting from a stand-still position is considered a key performance ability in this sport. For all of the above, and in the authors' opinion, the reliability of the published protocols to analyze sprint capacity in WB players is still limited. The purposes of this study are (a) to develop a reliable methodological design in order to assess sprint performance of WB players, using IMU and a laser system with computer vision in a 20 m sprint test, and (b) to assess bilateral symmetry as an indicator of sport performance and as an injury prevention indicator in WB players. Thus, the relation of the total acceleration of the players' wrists to the speed achieved by the WB players was explored during an on-court 20 m sprint test as an indicator of improvement in performance.

## 2. Materials and Methods

### 2.1. Participants

Ten elite WB players (8 men and 2 women) participated in the study. Throughout the study period, these players won first positions in the National League, the National King's Cup and the European Cup for clubs. In addition, seven of them were medalists in the Rio de Janeiro Paralympic games. All were classified by the International Wheelchair Basketball Federation (IWBF) Player Classification System [25] (Table 1). Their height was measured using a stadiometer in a standing situation (DKSH Switzerland Ltd., Zurich, Switzerland,  $\pm 0.1$  cm) Body weight was assessed using a calibrated scale (Kern MWS, Twister Medical, Barcelona, Spain,  $\pm 0.1$  kg). Players were first weighed in their own sport wheelchairs; then, the wheelchair was weighed without them, and the wheelchair weight was then calculated.

**Table 1.** Main characteristics of the wheelchair basketball players in the study.

Player	Age	Weight (Kg)	Height (m)	Disability	IWFB Classification	Experience (Years)
P1	33	74	1.80	Amputation	4.5	20
P2	39	75	1.86	Amputation	4.0	22
P3	28	75	1.86	Amputation	4.0	8
P4	39	90	1.82	Paraplegia	4.0	21
P5	17	60	1.84	Amputation	3.5	5
P6	40	90	1.82	Spina Bifida	3	10
P7	26	83	1.78	Spina Bifida	3	8
P8	23	72	1.75	Paraplegia	2.5	6
P9	30	55	1.50	Paraplegia	1	18
P10	33	102	1.80	Paraplegia	1	17
Sample	30.80 ± 7.54	77.60 ± 14.18	1.78 ± 0.11			13.50 ± 6.70

All of the players trained 4–5 days per week, playing one game per week. The study was undertaken during the competitive period. During data collection, no participants experienced injuries that could have potentially influenced their ability to perform training or research tasks. All participants were selected during the preparation–screening period. The study was approved by the University Ethics Committee, and it was undertaken according to the Helsinki Declaration on research in humans [26]. Information about the study aims was provided, and informed consent was obtained prior to the study and data collection. All of the subjects wore sports gear specific to the sport and used their sports wheelchair. In addition, the strapping and wheelchair configuration were chosen by every player for training and competition, as the players’ functional classifications are valid in these circumstances [2].

## 2.2. Equipment

Distances were measured using a laser sensor-type 1 LDM301 (Jenoptik, Jena, Germany) with a range of 0.5–300 m on natural surfaces, an accuracy of  $\pm 0.06$  m for 2 kHz and a resolution of 0.001 m. In addition, a laser TLM160i (Stanley, Mechelen, Belgium), calibrated according to the ISO standard, was used to measure test distances. Labview software, v. 13.0 (National Instruments, Austin, TX, USA), was used for data recording and processing to obtain velocities. All of the components were integrated into a BioLaserSport<sup>®</sup> (Ferro, A. Madrid, Spain) [27], real-time kinematic analysis system for training and sports competitions (UPM-UPO, Madrid, Spain) [28], which included a computer vision system developed by visual algorithm tracking in C++ [29], and which validity and reliability were calculated in previous studies using a 30 m sprint test [30]. High correlation coefficients of 0.962 for mean velocity ( $V_a$ ) were found with regard to the photocell systems and 0.869 for maximum velocity ( $V_{max}$ ) with regard to high-speed photogrammetry. For intrasession reliability, intraclass correlation coefficients (ICC) showed values of 0.945 for  $V_a$  and 0.866 for  $V_{max}$ , with 95% confidence intervals (CI) [30].

Two multi-sensor wireless inertial measurement units (WIMU<sup>®</sup> v. 1.6., Real Track System S.L, Almería, Spain) were used. These devices contained two three-axial accelerometers (400 g of full range scale and recording at 1000 Hz), which provided the components of the vector sum of gravitational and inertial linear accelerations. The data were processed with Qüiko software v.882 (Realtrack Systems, S.L. Almería, Spain).

## 2.3. Design and Procedures

The 20 m sprint test (Figure 1) was performed according to [7], in a real wooden facility with a training basketball court. Players were asked to inflate their tubes up to the maximum, as during a competition. The test started with a standard 15 min warm-up directed by the coach, which included continuous wheeling, joint mobility, and stretching of the upper limbs in static and dynamic situations. The athletes were positioned at the

start line with the front wheelchairs' castors over the line and the players' trunk behind the same line. Two posts were placed at the end of each line, to ensure that the players' chests or the wheels did not cross before the start of the test. Starting verbal signals used were "ready" and then "when you like", so the players were free to perform preparatory driving movements with their trunks and to start propelling the wheelchair forward when they were ready, avoiding the undesirable effects of the players' reaction times on sprint performance. The tests were performed twice, and the end score was the mean of the two trials. The laser was situated 5 m from the starting line and the laser beam hit the players' backs at a height of 0.63 m from the ground, while the laser beam was controlled its horizontality. A 2000 Hz sampling frequency was selected for data position recording. Data were then filtered at 3 Hz with a second-order Butterworth low-pass filter. Then, maximum velocity ( $V_{max}$ ) and average velocity ( $V_a$ ) were calculated in the sections: 0–3, 3–5, 5–10, 10–15 and 15–20 m.

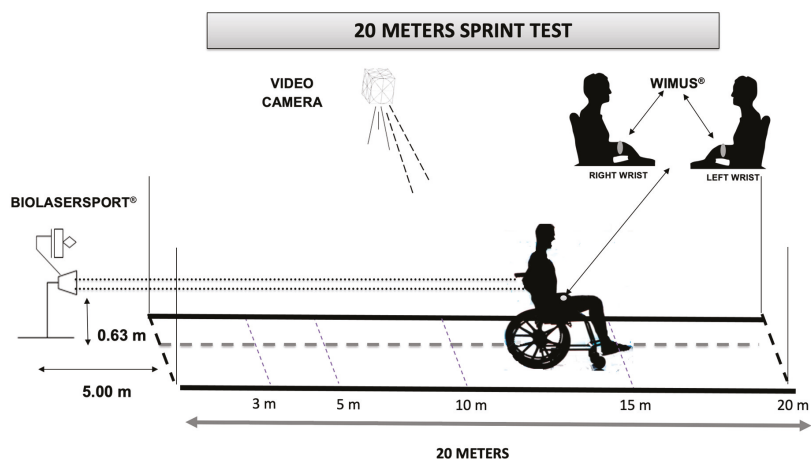


Figure 1. Diagram of field 20 m sprint test.

Both WIMU<sup>®</sup> were fastened on the players' right and left wrists using elastic bands. Players were in an upright position while devices were placed. The ulnar and radial styloid apophysis were located and, from that point on, the entire forearm was supported. This allowed the wrist total freedom of movement during the propulsion of the wheelchair. To remove random noise, the optimal cutoff frequency was established using a low-pass filter for speeds greater than 1.8 m/s [31]. In our case, the measured accelerations were low-pass-filtered with a cut-off frequency of 12 Hz using a 4th-order zero-lag Butterworth filter [6]. Accelerometer calibration was verified and checked at the beginning of the experimental session. Two video cameras (Exilim EX-ZR1000, Tokio, Japan) were placed on each side of the 20 m sprint test. All attempts were recorded at 240 Hz, and a synchronism signal was sent from 2 WIMU<sup>®</sup> and recorded to detect the propulsion cycles of each WB player. Based on the team's technical staffs' advice, for each session/player/trial, the total acceleration in each propulsion cycle ( $AcelTp$ ) and the average and maximum  $AcelT$  were recorded in the indicated five different sections. Bilateral symmetry between the dominant and non-dominant wrist was computed from  $AcelT$  in each propulsion cycle according to Bergamini et al. [6]. The calculation was carried out using the following algorithm  $sym = [AcelTp_{dominant} / (AcelTp_{dominant} + AcelTp_{non-dominant})] \times 100$ . To study if the dominant and non-dominant wrist presented similar peak accelerations [6], bilateral symmetry with values ranging between 45% and 55% indicated adequate symmetry, whereas a value lower than 45% or higher than 55% reflected greater accelerations of the non-dominant or dominant wrist, respectively [23].

#### 2.4. Statistical Analysis

Descriptive analyses were carried out for each test. The Shapiro–Wilk test was used to assess the normality of the tested variables. For intrasession reliability, the relationship between the results of AcelTp in the first and the second test series was analyzed using the ICC (2,1) as well as a CI of 95%. CI estimation provides a range of values with a specific probability, including true reliability [32]. The standard error of measurement (SEM) was calculated as  $(SD)\sqrt{1.00 - ICC}$ , where the standard deviation (SD) was the deviation of the observed scores. The relationships between the average and maximum AcelT, Va and Vmax were examined using Pearson's product–moment correlation coefficient ( $r$ ). The significance level was determined at  $p < 0.05$ . All of the calculations were performed with the SPSS software program, version 21.0 (IBM Corp., Armonk, NY, USA).

### 3. Results

#### 3.1. Twenty-Meter Sprint Test Protocol Reliability Using Two WIMU

Tables 2 and 3 show the average AcelTp (in each of the nine propulsion cycles) obtained for all the players in the 20 m sprint test, as the data allowing intrasession reliability assessment indicated no significant differences between the different series. Average ICC values were 0.90 for WIMU-1 on the non-dominant side and 0.85 for WIMU-2 on the dominant side, both with 95% CI and an average SEM of 1.15 and 1.24 g, respectively.

**Table 2.** Intrasession reliability for the average total acceleration in each of the nine propulsion cycles of the non-dominant wrists of all the players during two series of 20 m sprint tests (using WIMU-1).

Variable/ Cycle 1–9	Test 1	Test 2	ICC (95% IC)	SEM (g)
AcelTp 1 (g)	10.03 ± 2.02	9.69 ± 1.87	0.796 (0.393–0.944)	0.84
AcelTp 2 (g)	12.10 ± 2.52	11.89 ± 2.65	0.770 (0.304–0.938)	1.20
AcelTp 3 (g)	13.15 ± 3.48	12.64 ± 3.13	0.854 (0.432–0.950)	1.20
AcelTp 4 (g)	14.12 ± 3.98	13.96 ± 4.26	0.959 (0.846–0.990)	0.81
AcelTp 5 (g)	14.76 ± 4.29	14.81 ± 5.10	0.961 (0.850–0.990)	0.85
AcelTp 6 (g)	15.70 ± 5.15	15.04 ± 5.83	0.962 (0.858–0.990)	1.00
AcelTp 7 (g)	16.18 ± 5.22	15.87 ± 5.68	0.944 (0.796–0.986)	1.24
AcelTp 8 (g)	17.42 ± 6.10	16.20 ± 5.99	0.924 (0.709–0.981)	1.65
AcelTp 9 (g)	17.94 ± 6.37	16.30 ± 6.09	0.930 (0.483–0.985)	1.61

AcelTp: total acceleration in each propulsion cycle. ICC: intraclass correlation coefficient. CI: confidence interval. SEM: standard error of measurement.

**Table 3.** Intrasession reliability for the average total acceleration in each of the nine propulsion cycles of the dominant wrists of all the players during two series of 20 m sprint tests (using WIMU-2).

Variable/ Cycle 1–9	Test 1	Test 2	ICC (95% IC)	SEM (g)
AcelTp 1 (g)	9.22 ± 1.65	9.49 ± 1.95	0.772 (0.327–0.938)	0.79
AcelTp 2 (g)	10.83 ± 2.19	10.44 ± 1.88	0.772 (0.304–0.937)	0.90
AcelTp 3 (g)	11.62 ± 3.10	11.83 ± 3.02	0.855 (0.519–0.962)	1.15
AcelTp 4 (g)	12.39 ± 3.18	12.40 ± 3.08	0.869 (0.446–0.968)	1.11
AcelTp 5 (g)	13.69 ± 4.27	13.30 ± 3.63	0.866 (0.371–0.970)	1.33
AcelTp 6 (g)	13.45 ± 4.34	13.36 ± 3.76	0.884 (0.516–0.972)	1.28
AcelTp 7 (g)	14.39 ± 4.96	13.90 ± 3.84	0.855 (0.410–0.964)	1.46
AcelTp 8 (g)	14.76 ± 5.16	14.19 ± 4.61	0.863 (0.446–0.966)	1.71
AcelTp 9 (g)	14.47 ± 4.65	14.68 ± 4.76	0.904 (0.602–0.976)	1.44

AcelTp: total acceleration in each propulsion cycle. ICC: intraclass correlation coefficient. CI: confidence interval. SEM: standard error of measurement.



### 3.2. Twenty-Meter Sprint Test Protocol Reliability Using BioLaserSport® with Computer Vision System

Table 4 presents the intrasession reliability from the average Vmax assessment obtained during the 20 m sprint test for all the players, not observing significant differences between each series. Average ICC values were 0.967 for all Vmax sections with 95% CI and SEM values of <0.05 m/s.

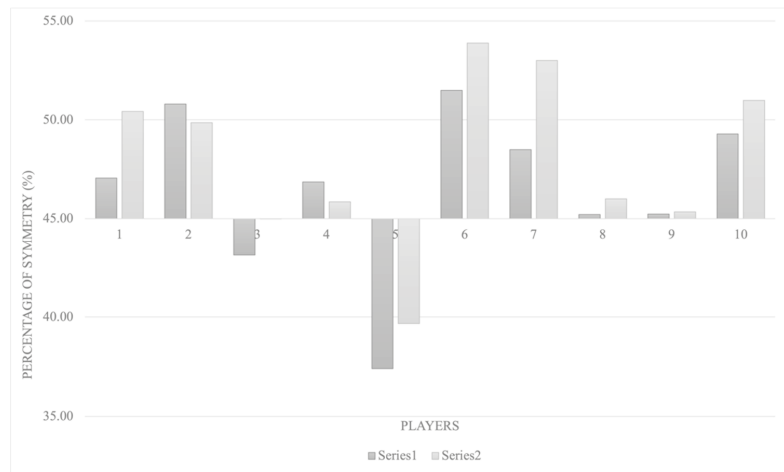
**Table 4.** Intrasession reliability for average maximum velocity for all the players during two series of 20 m sprint tests.

Variable/section	Test 1	Test 2	ICC (95% IC)	SEM (m/s)
Vmax 0–3 (m/s)	3.41 ± 0.27	3.44 ± 0.28	0.962 (0.851–0.981)	0.05
Vmax 3–5 (m/s)	3.97 ± 0.21	4.01 ± 0.16	0.962 (0.862–0.990)	0.04
Vmax 5–10 (m/s)	4.65 ± 0.22	4.75 ± 0.29	0.958 (0.851–0.989)	0.05
Vmax 10–15 (m/s)	5.05 ± 0.26	5.09 ± 0.32	0.976 (0.913–0.994)	0.05
Vmax 15–20 (m/s)	5.30 ± 0.29	5.33 ± 0.29	0.970 (0.885–0.992)	0.05

Vmax: maximum velocity. ICC: intraclass correlation coefficient. CI: confidence interval. SEM: standard error of measurement.

### 3.3. Players’ Wrist Bilateral Symmetry in Two Series of 20 M Sprint Tests

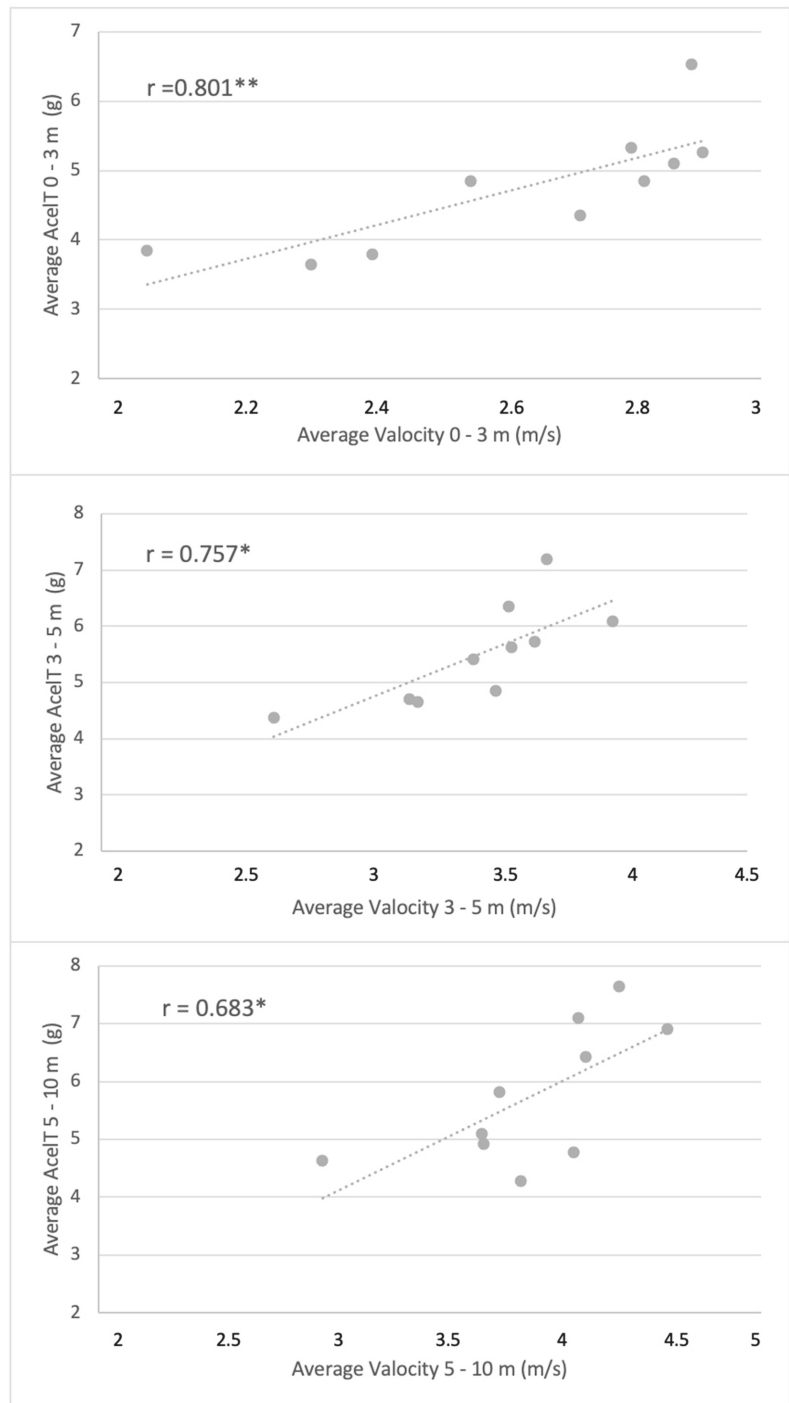
Figure 2 shows the symmetry between dominant and non-dominant wrists during the tests. Two players did not present adequate bilateral symmetry in the two 20 m sprint tests because they were not between the 45–55% range of values. The remaining players showed adequate symmetry.



**Figure 2.** Bilateral symmetry for all the WB players in two series of 20 m sprint tests.

### 3.4. Relation between Average/Maximum Total Acceleration and Average/Maximum Velocity from Dominant/Non-Dominant Players’ Wrists

Significant correlations were found for the players’ dominant wrists in 0–3 m, 3–5 m, and 5–10 m between the average velocity and average AcelT (Figure 3) ( $p < 0.05$ ). However, for the maximum AcelT and Vmax values, no significant correlations were found, with none found for the non-dominant side.



**Figure 3.** Correlations between the average velocity (Va) and the average total acceleration (AcclT) of the dominant side in the 20 m sprint test sections: 0–3 m, 3–5 m and 5–10 m (\*  $p < 0.05$ , \*\*  $p < 0.01$ ).

#### 4. Discussion

This original study presents a new and reliable methodology for WB players' sprint assessments in real sport training conditions using two new technologies, WIMU<sup>®</sup> and BioLaserSport<sup>®</sup> with a computer vision system (inertial devices and laser system). The analysis of the players' sprint capacity in relation to wrist acceleration seems important, as sprinting is a crucial skill in WB [33], with remarkable and novel significance for players and coaches. With objective data gathering from wrist acceleration using WIMU, sprinting was found in our study to be a key component in overall wheeling abilities and is also known for its potential injury incidence in this population [34]. Moreover, our analysis was conducted in a sport context, so application and transfer of these findings to a specific training assessment and screening is possible, especially with an elite-level sample. To our knowledge, this study is the first to relate the wrist triaxial acceleration of WB players to their speed achieved in an on-court 20 m sprint test.

First, there was a need to verify protocol reliability for the 20 m sprint test using the BioLaserSport<sup>®</sup> and the WIMU<sup>®</sup> systems. Using computer vision, we obtained an average ICC of 0.967 in Vmax and a lower SEM of 0.05 m/s. Comparing our tests with those performed by Ferro et al. [7], the use of the laser system with computer vision further improved the reliability of the calculation of the WB players' velocity by 7%. In relation to the use of WIMU<sup>®</sup>, following Bergamini et al. [6], this proposed protocol is not clear. The placement of their WIMU<sup>®</sup> on the wrists in that study was not well specified, so study reproducibility is not possible, and they do not provide data on measurement reliability. In our study, WIMU<sup>®</sup> placement was described, and reliability data were presented, showing an ICC of 0.90 and 0.85 for players' non-dominant and dominant wrists, and a SEM between 0.79 and 1.71 g was recorded. As a consequence, the acceleration data in our study were reliable. The combination of these two technologies provides this study with innovation and significance for future practical applications in real sport-specific WB contexts.

It is important that the technologies used in the analysis of athletic performance have certain criteria and rigor, as is the case of the WIMU<sup>®</sup> and the BioLaserSport<sup>®</sup> with computer vision used in this research. Some studies that analyze the performance velocity of athletes using a stopwatch have a limitation on the application of the results because of the less reliable system [6,12,13,35]. Similarly, and in order to assess sprint capacity in WB players, some previous studies [7,8,13,36] have used a speed test, with the 20 m sprint being the most common. However, no studies have been found in the literature observing the importance of wrist action with the speed achieved by the WB player. In our study, a significant correlation was observed between the average velocity of the WB players with an average acceleration of the dominant wrist of the players in the 0–3 m ( $r = 0.801$ ), 3–5 m ( $r = 0.757$ ) and 5–10 m sections ( $r = 0.683$ ) (Figure 3). We observed that this relationship is stronger at the beginning of the test (in the 0–3 m section versus the 5–10 m section). We could conclude that the acceleration produced by the dominant wrist is more crucial at the start of the sprint. We found studies that have analyzed the speed in the first few meters of a sprint [7,37], which suggests the importance of this ability at the start of a sprint, to enable the player to achieve very early speeds in order to intercept the opponent's trajectory during the actions of a basketball game [10].

Regarding the second aim dealing with bilateral symmetry assessment, we found articles that analyzed it but failed to identify the differences between the dominant and non-dominant sides [6,12]. In our case, two participants did show differences between the sides (Figure 2), as they showed a relationship below 45% [23]. These findings were not associated with the players' functional classification; this finding appears to be in line with the conclusions of Wai et al. [24], as no differences were found in the wrist accelerations and seating height during manual wheelchair propulsion within a non-sportive population. In this study, the differences in one of the cases were very small; however, the high precision of the devices used and the reliability of the protocol could detect them. Again, we emphasize the importance of using reliable protocols to evaluate performance in high-level athletes.

This study was carried out with 10 athletes. This could be initially considered a limitation. However, the data could be used as a reference because of the participation of high-level athletes. They achieved first place in three competitions in one season, the National League, the European Cup and the National King's Cup, and, in addition, seven of the participants were Paralympic medalists. For this reason, the results of the study are especially important. In addition, two female players participated in the study; in most clubs at national level, women participate together with men in competitive WB in order to promote their participation in this sport. If the coach allows one female player to play on court, the sum of the functional classification of the five players on court can be up to 16 points (plus 1.5 points), instead of 14.5 points when only males are on court. In our opinion, their presence gives contextual validity to the results, as we know the two female players have also reached the Paralympic level in their careers.

In future investigations, it would be interesting to carry out studies where the joint patterns during the propulsion cycle are analyzed in more detail, as well as identifying how they influence speed gains during WB sprint performance. We should consider optimizing mobility performance in court sports (wheelchair basketball, wheelchair rugby and wheelchair tennis), as this depends on an ergonomic combination of factors associated with the player, their own sport wheelchair and the interface between them [10,38].

## 5. Conclusions

In conclusion, this study presents a new, reliable and valid protocol, using WIMU<sup>®</sup> and BiolaserSport<sup>®</sup> with computer vision, to assess bilateral symmetries during sprint performance in WB players. This protocol demonstrated the relationship between the players' velocity of movement and the generated acceleration of the players' wrists. It was observed that there was a significant relationship between the average acceleration of the dominant wrist and the average velocity of the player in the wheelchair, with this relationship being stronger at the beginning (0–3 m) of the 20 m sprint test. Coaches can use this protocol to assess performance or for injury prevention, as it showed very good reliability, with very high ICC values.

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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 Universidad Politécnica de Madrid, Spain, in 4 February 2015.

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

**Data Availability Statement:** Not applicable.

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Article

# Outpatient Assessment of Mechanical Load, Heat Strain and Dehydration as Causes of Transitional Acute Kidney Injury in Endurance Trail Runners

Daniel Rojas-Valverde <sup>1,2,3,\*</sup>, Ismael Martínez-Guardado <sup>4,\*</sup>, Braulio Sánchez-Ureña <sup>5</sup>, Rafael Timón <sup>3</sup>, Volker Scheer <sup>6</sup>, José Pino-Ortega <sup>7</sup> and Guillermo Olcina <sup>3</sup>

- <sup>1</sup> Centro de Investigación y Diagnóstico en Salud y Deporte (CIDISAD), Escuela Ciencias del Movimiento Humano y Calidad de Vida (CIEMHCAVI), Universidad Nacional, Heredia 86-3000, Costa Rica
  - <sup>2</sup> Clínica de Lesiones Deportivas (Rehab & Readapt), Escuela Ciencias del Movimiento Humano y Calidad de Vida (CIEMHCAVI), Universidad Nacional de Costa Rica, Heredia 86-3000, Costa Rica
  - <sup>3</sup> Grupo Avances en Entrenamiento Deportivo y Acondicionamiento Físico (GAEDAF), Facultad Ciencias del Deporte, Universidad de Extremadura, 10003 Cáceres, Spain; rtimon@unex.es (R.T.); golcina@unex.es (G.O.)
  - <sup>4</sup> Faculty of Life and Natural Sciences, University of Nebrija, 28015 Madrid, Spain
  - <sup>5</sup> Programa de Ciencias del Ejercicio y la Salud, Escuela Ciencias del Movimiento Humano y Calidad de Vida, Universidad Nacional, Heredia 86-3000, Costa Rica; brau09@hotmail.com
  - <sup>6</sup> Ultra Sports Science Foundation, 69310 Pierre-Bénite, France; volkerscheer@yahoo.com
  - <sup>7</sup> Biovetmed & Sports Research Group, University of Murcia, 30720 San Javier, Spain; josepinoortega@um.es
- \* Correspondence: drojasv@una.cr (D.R.-V.); imartinezgu@nebrija.es (I.M.-G.); Tel.: +506-88250219 (D.R.-V.)

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**Abstract:** Background: This study aimed to globally assess heat strain, dehydration, and mechanical load as acute kidney injury (AKI) indicators in amateur endurance trail athletes during a 35.3 km run. Methods: Thirty amateur experienced trail runners completed an endurance trail run (total positive ascend 1815 m). The following assessments were performed at four measurement time points (pre-, during, immediately post [-post<sub>0h</sub>], and after 24 h of the finish of the run [-post<sub>24h</sub>]): serum test (creatinine, blood ureic nitrogen, albumin, creatine kinase, blood ureic nitrogen: creatinine ratio, creatinine clearance, and glomerular filtration rate), mechanical load (impacts and Player Load), heat strain and dehydration (hematocrit, urine solids, body weight and urine specific gravity), pain and exertion perception (rate of perceived exertion, lumbar and bipodal, and one-leg squat pain), and urinalysis (pH, protein, glucose, erythrocytes, and urine specific gravity). Results: There were pre vs. post<sub>0h</sub> changes in all serum biomarkers ( $F = 5.4\text{--}34.45$ ,  $p < 0.01$ ). The change in these biomarkers correlated with an increase in mechanical load indicators ( $r = 0.47\text{--}59$ ,  $p < 0.05$ ). A total of 40% and 23.4% of participants presented proteinuria and hematuria, respectively. Pain and perceived exertion increased significantly due to effort made during the endurance trail running ( $F = 4.2\text{--}176.4$ ,  $p < 0.01$ ). Conclusions: Endurance trail running may lead to an increase in blood and urine indicators of transitional AKI. The difference in blood and urine markers was significantly related to the mechanical load during running, suggesting potential kidney overload and cumulative mechanical load.

**Keywords:** kidney failure; AKI; health; biomarkers; strenuous exercise; mountain running; kidney function; off-road running

## 1. Introduction

There are activities of increasing popularity in endurance sports, such as running, cycling, triathlon, and open water swimming. This widespread endurance sports practice responds to the economic prosperity of some populations, relatively inexpensive travel cost, and the relatively affordable and user-friendly required equipment for its practice [1]. Endurance exercising usually requires great effort and takes the body to its physiological,



cognitive, and physical limits. The cumulative moderate to high-intensity actions over a prolonged period may lead to fatigue, temporal disfunction, injury, or even death in conjunction with other potential risks [2,3]. In this sense, trail running is one of the most physically demanding sports due to the difficulty and duration of the events, terrain characteristics, weather variation, and slope changes [3–6]. These characteristics require several concentric and eccentric muscle actions, which have proven highly tiring at the neuromuscular level [4,7].

Muscle damage is a common consequence of trail running [3]. Still, recently, there has been an increase in the concern for trail running participants' well-being due to the prevalence of some adverse health conditions at the cardiovascular, immunological, hepatic, and renal levels [3,8–10]. Specifically, at the renal level, physical stress is a factor that contributes to the transitory decrease of renal function [3,11]. This short-term kidney issue is clinically characterized by a rise of nitrogen waste products in the bloodstream in response to mechanical and thermal muscle injury and future inflammation responses that could compromise the balance of fluids in the body [2]. This condition is known as acute kidney injury (AKI), also called acute renal failure, which in most cases is reversible and asymptomatic [8,11].

There are several methods to quantify renal function. It is commonly assessed by chemical waste molecules generated from muscle metabolism. During exercise, due to muscle damage, the protein-waste products released to the bloodstream could overload the kidney. Among these biochemical markers of kidney damage are serum creatine kinase (sCK), serum creatinine (sCr), serum blood ureic nitrogen (sBUN), serum albumin (ALB), and recently through Cystatin-C (Cyst-C) [9,12]. Other indicators less frequently reported are neutrophil gelatinase-associated lipocalin (NGAL) and kidney injury molecule 1 (KIM-1) [13]. From these well-known AKI biomarkers, some other variables could be estimated, such as BUN-to-Cr ratio (BUN/Cr), glomerular filtration rate (eGFR), albumin-to-creatinine ratio (ALB/Cr), and creatinine clearance [14,15]. The clinical diagnosis of AKI has been established based mainly on sCr levels criteria [12]. Nowadays, alternative subclinical and functional measures have been proposed, but there is no scientific consensus [13], and no range values for sports have been established [3].

AKI is understood as a multi etiology condition. There are three factors proposed as enhancers of AKI development in sports: high mechanical load, heat strain, and dehydration [3,16]. Recent research has suggested that high physical load is the common denominator of AKI progress [11,17,18]. AKI has been reported among endurance athletes and other populations that usually perform high physical loads during prolonged periods under hot and humid conditions [11,19]. Heat and dehydration would enhance possible damage and exposure, and partially explain mechanical kidney damage [11].

AKI has been studied in populations with the abovementioned conditions, such as athletes, agricultural workers, firefighters, and builders [16,20]. Although this evidence exists, clarification is needed as to how these three factors contribute to AKI development under isolated conditions and in outpatient settings [2,3]. There has been no consensus on the long-term consequences of subsequent AKI events leading to chronic renal disease [10,21]. Still, some hypotheses considering renal scarring and maladaptive repair due to cumulative AKI events could better understand this link between acute and long-term renal issues [22]. For this reason, recent studies have tried to use gold-standard measurement protocols in outpatient settings that provide valuable information for a better understanding of this phenomenon [11], but more evidence is required.

The prevalence and severity of AKI episodes among endurance athletes have been debated without reaching a consensus. Some authors point out that AKI should be considered a health problem or issue [23], and others consider it a common effect of physical exercise [2,8]. Highly-demanded sports, such as running, cycling, and triathlon, have high load components understood as high volume and high intensity that may contribute to the potential development of a renal condition [11]. Due to the lack of evidence that addresses the three main causal factors of AKI in endurance sports, there is a need to clarify

specific gray points on the understanding of this condition and to determine how each of these factors influence separately and wholly the development of AKI [3]. A global assessment of the AKI phenomenon is indispensable. This study aimed to globally assess heat strain, dehydration, and mechanical load as acute kidney injury (AKI) indicators in amateur endurance trail athletes during a 35.3 km run.

## 2. Materials and Methods

### 2.1. Participants

Thirty male amateur trail runners took part in this study (age  $39.5 \pm 9.23$  years, weight  $71.26 \pm 11.17$  kg, height  $171.65 \pm 8.69$  cm). Participants were selected from different trail running clubs. Participants were required to be >18 years old, experienced ( $5.69 \pm 2.77$  years), trained ( $9.02 \pm 3.57$  h/week), and heat acclimatized (sleep and train in similar study's altitude and weather) endurance runners. Participants reported no neuromuscular or metabolic disturbance or injury at least six months before the study.

The experimental protocol was approved by the Institutional Review Board of the National University of Costa Rica (Reg. Code UNA-CECUNA-2019-P005) and the University of Extremadura (Reg. Code 139/2020). All the participants were informed of the details of the experiment procedures, the associated risks and discomforts, and their benefits and rights. Each subject gave written informed consent, according to the criteria of the Declaration of Helsinki, regarding biomedical research involving human subjects (18th Medical Assembly, 1964, revised in 2013 in Fortaleza).

### 2.2. Study Design

Participants were measured in four different time points (pre-, during, -post<sub>0h</sub>, and -post<sub>24h</sub>) (Figure 1). All participants were asked to trail run a total distance of 35.3 km (cumulative positive ascend of 1815 m, lowest and highest altitude = 906 and 1178 m.a.s.l.). The thermal stress index was registered at the start line of the trail run. The mean thermal index was  $24.31 \pm 1.6$  °C (temperature:  $25.52 \pm 1.98$  °C and humidity  $79.25 \pm 7.45\%$ ) according to the WetBulb-Globe Temperature (WBGT). The participants spent  $294.14 \pm 59.34$  min to finish the three loops.

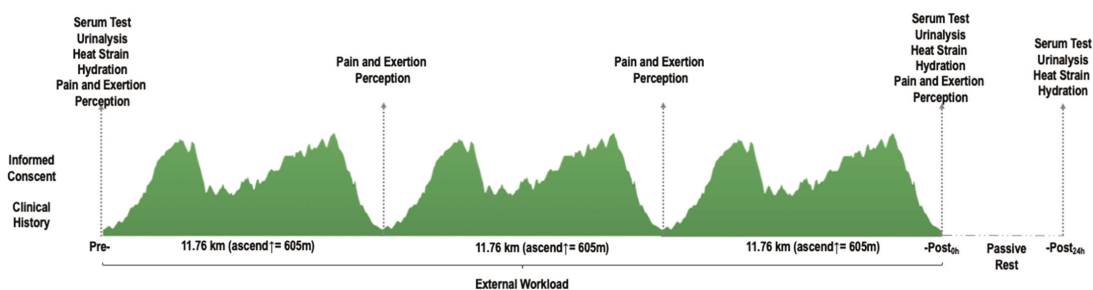


Figure 1. Schematic design of study variables with measurement time and trail altimetry.

As shown in Figure 1, serum and urine samples and heat strain and hydration variables were assessed ~15 min before and after the trail run. A follow-up was performed 24 h after (individual visits). The participants were allowed to carry their hydration throughout the event, so their liquid and food intake were established *ad libitum* as typical in trail running races. Perceived pain and exertion data was collected ~10 min before, after, and during the run. Mechanical load variables were monitored all over the trail run using wearable devices.

### 2.3. Materials and Procedures

For serum test analysis, blood was extracted *in-situ* from the antecubital vein using a 5 mL blood collection sterile tube (BD Vacutainer<sup>®</sup>, Franklin Lakes, NJ, USA) that contained spray-coated silica particles activator and gel polymer to facilitate serum separation during centrifugation (10 min at  $2000 \times g$  relative centrifugal force). Centrifugation was performed using a tube centrifuge (PLC-01, Gemmy Industrial Corp., Taipei, Taiwan).

Blood samples were stored on ice in a special cooler (45QW Elite, Pelican<sup>™</sup>, California, USA) until serum samples were frozen at  $-20\text{ }^{\circ}\text{C}$  ( $\sim 5$  h after blood extraction). Samples analyses and processing were performed 24 h after data collection in an isolated and temperature-controlled laboratory using an automatic biochemical analyzer (BS-200E, Mindray, Beijing, China) by photometry method. All procedures were executed under relevant protocols for the handling and disposal of biological materials, according to the manufacturer's instructions for the equipment and reagents used.

The variables analyzed were serum creatinine (sCr, mg/dL), creatine kinase (sCK, IU/L), ureic nitrogen (sBUN, mg/dL), and albumin (sALB, IU/L). The following variables were also estimated: eGFR ( $\text{mL}/\text{min}/1.73\text{ m}^2$ ),  $\text{Cr}_{\text{Clearance}}$  ( $\text{mL}/\text{min}$ ) and sBUN/sCr ratio.  $\text{Cr}_{\text{Clearance}}$  was predicted from sCr using the Cockcroft and Gault [14] estimation equation:

$$\text{Cr}_{\text{Clearance}} = (140 - \text{age}) * \frac{\text{body mass (kg)}}{\text{sCr (mg/dL)}} * 72 \quad (1)$$

eGFR was calculated using CKD-EPI [15] sCr-based formula for male and white population as follow:

$$\text{eGFR} = 141 * \left( \frac{\text{sCr (mg/dL)}}{0.9} - 1.209 \right) * 0.993 * \text{age} \quad (2)$$

Urine samples were collected *in-situ* in a 30 mL polypropylene sterile urine container (Nipro Medical Corp., Osaka, Japan) and analyzed with highly sensitive and accurate dipsticks for urine screening (Combur<sub>10</sub> Test M, Roche, Mannheim, Germany), previously used in distance running settings [24]. Urine dipsticks were examined immediately after collection by two different observers simultaneously using the color scale reference provided by the manufacturer. In case of disagreement between observers, a consensus was obtained with the opinion of a third observer. The following parameters were screened: pH (acidity and basicity), protein, glucose, erythrocytes, and urine specific gravity (USG). There were no reported urination problems or difficulties neither before nor after the study. Urine solids were assessed, and USG was confirmed and double-checked with a digitally valid [25] handheld refractometer (Palm Abbe<sup>™</sup>, Misco, OH, USA). USG results were classified following the hydration status ranges: well-hydrated  $<1.01$ , minimal dehydration  $1.01\text{--}1.02$ , significant dehydration  $1.02\text{--}1.03$ , and severe dehydration  $>1.03$  [26]. The refractometer was previously cleaned with distilled water and calibrated.

Additionally, a sample of capillary blood was extracted from the right index finger, using a Na-heparinized capillary tube (80 IU/mL) (Marienfeld, Lauda-Königshofen, Germany) to assess hematocrit (Htc). The capillary micro-hematocrit tubes were centrifuged (KHY-400, Gemmy Industrial Corp., Taipei, Taiwan), and hematocrit values were evaluated using a special reader (Gemmy Industrial Corp., Taipei, Taiwan).

Participant's body mass (kg) was assessed semi-nude (underwear only) immediately pre and post-event using a digital balance (BC554, Tanita, Arlington Heights, IL, USA) in an isolated tent. Percentage body weight change was categorized as follow: well-hydrated  $+1\%$  to  $-1\%$ , minimal dehydration  $-1\%$  to  $-3\%$ , significant dehydration  $-3\%$  to  $-5\%$  and serious dehydration  $>5\%$  [26]. Liquid and food intake were set ad libitum and were not controlled to maintain organic trail run conditions.

During the whole event, variations of the WetBulb-Globe Temperature (WBGT) were registered with a heat stress monitor (QUESTemp™ 36, 3M™, Saint Paul, MN, USA) every 15 min. Equipment was calibrated and mounted in a stable tripod at the start and finish of the event.

Mechanical load variables were assessed using six inertial measurement units (WIMU PRO™, RealTrack Systems, Almería, Spain) attached to different anatomical spots (one IMU at thoracic 2nd–4th [ $T_2$ – $T_4$ ], one IMU at lumbar 1st–3rd [ $L_1$ – $L_3$ ]; two IMU at right [ $VL_{right}$ ] and left [ $VL_{left}$ ] vastus lateralis muscle bellies and two IMU 3 cm cephalic to right [ $MP_{right}$ ] and left [ $MP_{left}$ ] malleolus peroneus). IMUs were mounted using special spandex dark suit to avoid non-desired movement or shaking during running, as reported in previous studies [11].

The IMUs reliability during multidevice assessment had been tested [27]. Before the study was performed, all devices were calibrated following published protocols [27]. All IMU's were turned on 30 min before the matches started to reach the optimal internal temperature of the device ( $\sim 32$  °C). The sampling frequency of accelerometry-based external load indicators was set at 100 Hz. Each participant's devices were synchronized in time before analysis, and data filtration processes were applied (at chip-level and data processing algorithms) before the "raw data" was available for researchers. Default by the manufacturer made this filtering, and data processing was performed based on the redundancy principle and fusion of the incorporated sensors (accelerometer, magnetometer, and gyroscope). Total variables' data were extracted from IMU 15 min after the participants finished using special software.

The analyzed variables were selected considering similar studies investigating mechanical load and AKI in endurance events [11]. The extracted variables were: Player Load per min (AU, PL/min) and impacts (Impacts<sub>total</sub>, n/min) per body segment. Player Load is defined as the vector sum of accelerometer data points in the three axes of movement [28], representing the sum of the magnitudes of the movement. The impacts are defined as a short duration jerk, shock, or impact, expressed in gravitational forces ( $g$ -forces) where  $1g = 9.81$  m/s [18,29].

Perceived pain and rate of perceived exertion (RPE) were measured using a 0 to 10 point visual analog scale of pain (VAS-PAIN); zero is understood as no pain, and ten is an extreme pain [30]. This variable was assessed through three different tasks as follows: lumbar pain, asking participants for lumbar pain (pain in the lower back); squat pain, perceived pain after a 2 s-90° knee squat to evaluate knee and hip flexors and extensor pain during concentric contraction; and one-leg squat pain, same as the second task (but only with reported dominant leg). The RPE during the event was assessed using the modified Borg Scale, and it was evaluated in each of the control points (each 11.46 km).

#### 2.4. Statistical Analysis

Description of variables was reported using mean, standard deviation, and lower and upper limits. The normality of the data was confirmed using the Shapiro–Wilk test. Differences between variables were explored using one-way analysis of variance (serum test, specific gravity, heat strain, and hydration); the exploration of the measurement timepoint differences was made using Bonferroni Post Hoch when required. Hematocrit and body weight data were explored using paired  $t$ -test analysis. The magnitude of the differences (effect size) was qualitatively interpreted using partial omega squared ( $\omega_p^2$ ) as follows:  $>0.01$  small;  $>0.06$  moderate and  $>0.14$  large and using Cohen  $d$  ( $d$ ) as follows:  $<0.2$  trivial;  $0.2$ – $0.49$  small;  $0.5$ – $0.79$  moderate and  $>0.8$  large [31] when corresponded.

McNemar non-parametric test was used to explore the possible change in proportion for the paired data of urinalysis. In those observed cases, the intersection frequency value was  $<5$ , and the binomial test was performed. The data of pH, glucosuria, proteinuria, hematuria, and USG were paired by measurement timepoints using a  $2 \times 2$  contingency table. A Pearson Correlation matrix was performed and graphed to explore the correlation between serum markers' change percentage ( $\Delta\%$ ) and mechanical load variables.

Alpha was prior set as  $p < 0.05$ . Data analysis was performed using the Statistical Package for the Social Sciences (SPSS, IBM, SPSS Statistics, v.22.0, Chicago, IL, USA).

### 3. Results

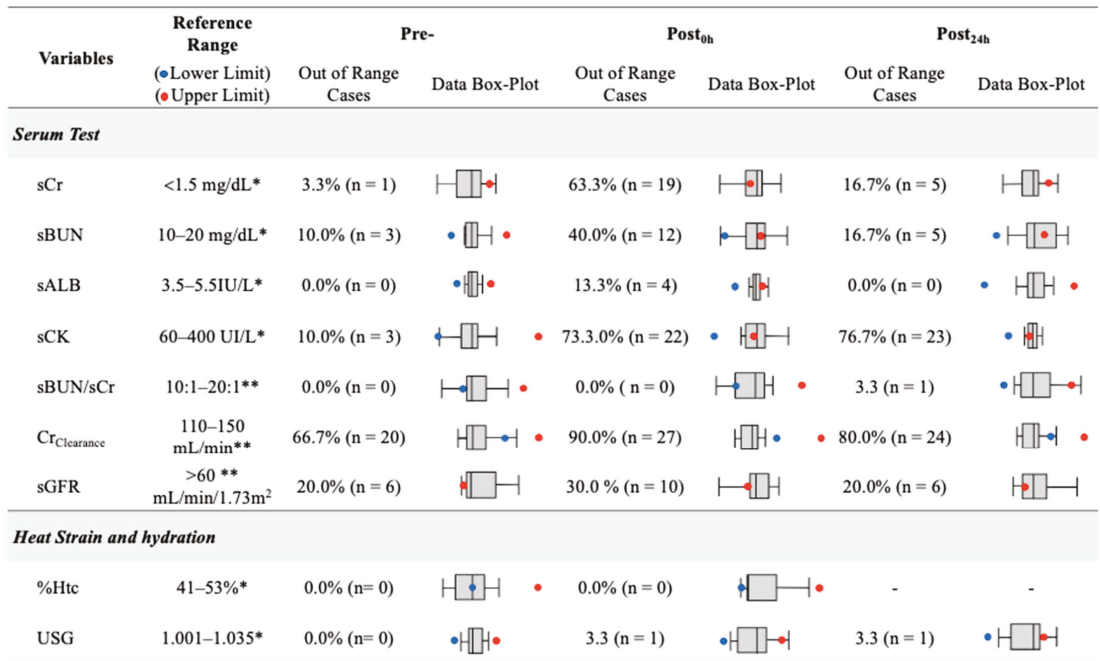
When compared pre and post<sub>0h</sub> and pre and post<sub>24h</sub> measurement time points, the levels of sCr, sCK, sBUN, sALB, USol, and sBUN/sCr ratio significantly increased ( $p < 0.01$ ). Moreover, a significant decrease in Cr<sub>Clearance</sub>, eGFR, and BW was presented after the trail run ( $p < 0.01$ ). Some variables, such as sALB, sCK, sCr, eGFR, USG, USol, Cr<sub>Clearance</sub>, and sBUN/sCr ratio also recovered the baseline values as evidence in post<sub>24h</sub> assessment (see Table 1).

**Table 1.** Pre-, -Post changes in serum, heat strain, and hydration variables.

Variable	Pre-	-Post <sub>0h</sub>	-Post <sub>24h</sub>	F/t Value	p-Value	$\omega_p^2/d$ Rating
<i>Serum test</i>						
sCr (mg/dL)	1.2 ± 0.3 (1.1 to 1.3)	1.6 ± 0.3 * (1.5 to 1.8)	1.3 ± 0.3 † (1.2 to 1.4)	34.5	<0.01	0.5 <i>large</i>
sCK (IU/L)	187.3 ± 182.2 (106.5 to 268.0)	528.0 ± 345.2 * (374.9 to 680.9)	677.0 ± 534.2 * (440.1 to 913.8)	16.3	<0.01	0.3 <i>large</i>
sBUN (mg/dL)	14.5 ± 4.1 (12.7 to 16.3)	19.4 ± 4.6 * (17.3 to 21.4)	18.9 ± 4.2 * (17.0 to 20.8)	26.9	<0.01	0.5 <i>large</i>
sALB (IU/L)	4.5 ± 0.7 (4.2 to 4.9)	5.1 ± 0.8 * (4.7 to 5.4)	4.7 ± 0.2 (4.6 to 4.8)	7.8	<0.01	0.2 <i>large</i>
eGFR (mL/min/1.73m <sup>2</sup> )	80.5 ± 24.7 (69.5 to 91.4)	69.9 ± 18.5 * (61.7 to 78.1)	74.7 ± 21.3 (65.3 to 84.1)	5.4	<0.01	0.1 <i>moderate</i>
Cr <sub>Clearance</sub> (mL/min)	87.4 ± 30.0 (74.1 to 100.7)	60.9 ± 25.9 * (49.4 to 72.4)	78.1 ± 30.8 *† (64.4 to 91.7)	43.8	<0.01	0.6 <i>large</i>
sBUN/sCr ratio	12.2 ± 3.3 (10.8 to 13.7)	12.0 ± 2.7 (10.8 to 13.3)	15.3 ± 4.0 *† (13.5 to 17.1)	15.7	<0.01	0.3 <i>large</i>
<i>Heat Strain and Hydration</i>						
USG	1.02 ± 0.02 (1.0 to 1.0)	1.02 ± 0.01 (1.0 to 1.0)	1.02 ± 0.01 (1.0 to 1.0)	0.8	0.5	0 <i>trivial</i>
USol	3.7 ± 2 (2.6 to 4.8)	5.4 ± 2 * (4.3 to 6.5)	4.7 ± 2.3 (3.5 to 6.0)	4.5	0.01	0.1 <i>moderate</i>
Htc (%)	41.2 ± 2.9 (40 to 42.4)	42.4 ± 3.7 (41 to 43.8)	-	-1.4	0.2	0.3 <i>small</i>
BW (kg)	71.7 ± 10.8 (67.6 to 75.8)	67.8 ± 15.8 * (61.7 to 73.9)	-	2.3	0.03	0.5 <i>moderate</i>

Data was presented in mean ± standard deviation and 95% upper and lower limits. Significant differences ( $p < 0.01$ ) with \* Pre- and † Post<sub>0h</sub>. sCr = creatinine, sCK = creatine kinase, sBUN = blood ureic nitrogen, sALB = albumin, eGFR, estimated glomerular filtration rate, Cr<sub>Clearance</sub> = creatinine clearance, USG = urine specific gravity, USol = urine solids, Htc = hematocrit, BW = body weight.

Figure 2 shows the number of out-of-range cases based on clinical criteria for each variable, following previous studies' references as stated in the method section. In this sense, post<sub>0h</sub> presented the higher out of range cases for AKI-related variables, such as sCr (63.0%, 19/30), sBUN (40.0%, 12/30), sALB (13.0%, 4/30), sCK (73.0%, 22/30), Cr<sub>Clearance</sub> (90.0%, 27/30), and eGFR (30.0%, 10/30).



**Figure 2.** Cases out of reference ranges in serum, heat strain and hydration variables by measurement timepoints. Reference Ranges: \* [32], \*\* [33]. sCr = creatinine, sCK = creatine kinase sBUN = blood ureic nitrogen, sALB = albumin, eGFR, estimated glomerular filtration rate, Cr<sub>Clearance</sub> = creatinine clearance, USG = urine specific gravity, Htc = hematocrit.

A significant change in proteinuria was found, presented as an increase in 40.0% of cases with 1+ or higher (pre vs. post<sub>0h</sub>). The baseline levels were recovered after 24 h in 40.0% of these cases. Hematuria cases increased by 23.4% (pre vs. post<sub>0h</sub>) and then decreased by 20.0% (post<sub>0h</sub> vs. post<sub>24h</sub>). Finally, cases of pH ≤ 5 decrease by 20.0% (pre vs. post<sub>24h</sub>) (see Table 2).

**Table 2.** Distribution and pattern of change of urine dipstick readings of pH, proteinuria, glucosuria, hematuria, and USG.

Variable	Pre-		-Post <sub>0h</sub>		χ <sup>2</sup>	p-Value	Pre-		-Post <sub>24h</sub>		χ <sup>2</sup>	p-Value	-Post <sub>0h</sub>		-Post <sub>24h</sub>		χ <sup>2</sup>	p-Value
	n	%	n	%			n	%	n	%			n	%	n	%		
pH ≤ 5	18	60.0%	13	43.3%	2.3	0.1	18	60.0%	12	40.0%	6.4	0.04	13	43.3%	12	40%	1.7	0.44
Proteinuria 1+ or higher	1	3.3%	13	43.3%	10.3	<0.01	1	3.3%	1	3.3%	0.0	1.0	13	43.3%	1	3.3%	10.3	<0.01
Glucosuria 1+ or higher	0	0.0%	0	0.0%	0.0	1.0	0	0.0%	0	0.0%	0.0	1.0	0	0.0%	0	0.0%	0.0	1.0
Hematuria 1+ or higher	1	3.3%	8	26.7%	12.2	<0.01	1	3.3%	3	6.7%	1.3	0.5	8	26.7%	3	6.7%	6.0	0.04
USG > 1.020	10	33.3%	20	66.7%	3.2	0.1	10	33.3%	14	46.7%	2.2	0.3	20	66.7%	14	46.7%	5.3	0.07

USG = urine specific gravity.

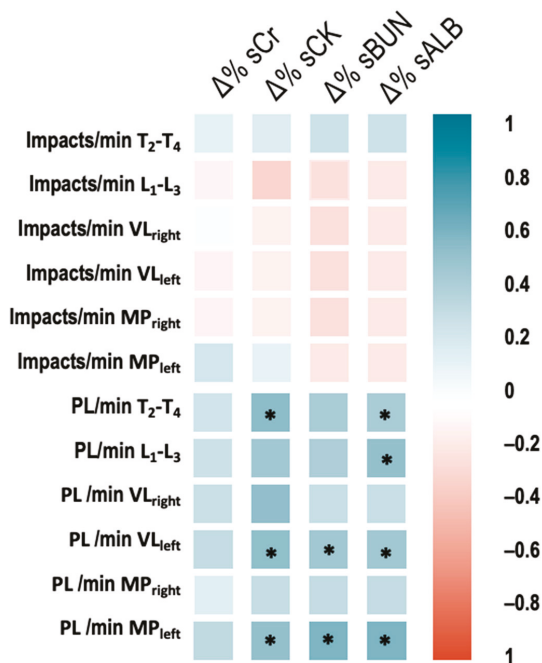
Perceived pain and physical perceived exertion changed throughout the running event. RPE, squat pain, one-leg squat pain increased ( $p < 0.01$ ) gradually between pre, 1st, and 2nd lap. Lumbar pain increased during the race, but no difference was found between the 1st and 2nd lap (see Table 3).

**Table 3.** Perceived pain and physical exertion by measurement timepoints.

Variable	Pre-	1st Lap	2nd Lap	Post <sub>0h</sub>	F Value	p-Value	$\omega_p^2$
RPE	0.0 ± 0.0 (0.0 to 0.0)	6.4 ± 2.2 * (5.5 to 7.2)	7.6 ± 2.0 * (6.8 to 8.4)	8.8 ± 1.2 *†‡ (8.3 to 9.2)	176.4	<0.01	0.9 <i>large</i>
Lumbar Pain	0.0 ± 0.0 (0.0 to 0.0)	0.0 ± 0.0 (0.0 to 0.0)	1.0 ± 1.8 *† (0.3 to 1.7)	0.9 ± 2.1 *† (0.1 to 1.8)	4.2	<0.01	0.1 <i>moderate</i>
Squat Pain	0.1 ± 0.4 (0.08 to 0.2)	0.4 ± 1.1 (0.1 to 0.8)	2.7 ± 3.4 *† (1.3 to 4.1)	2.5 ± 3.2 *† (1.2 to 3.7)	11.7	<0.01	0.3 <i>large</i>
One-leg squat Pain	0.1 ± 0.4 (0.08 to 0.2)	0.3 ± 1.0 (0.1 to 0.7)	1.7 ± 2.8 * (0.5 to 2.8)	1.9 ± 3.0 *† (0.7 to 3.2)	6.8	<0.01	0.2 <i>large</i>

Significant differences ( $p < 0.05$ ) with \* Pre-, † 1st Lap and ‡ 2nd Lap. RPE = rate or perceived exertion.

The percentage of change ( $\Delta\%$ ) of sCK correlated significantly with PL MP<sub>left</sub> ( $r = 0.59, p < 0.01$ ), PL VL<sub>left</sub> ( $r = 0.53, p = 0.02$ ) and PL T<sub>2</sub>-T<sub>4</sub> ( $r = 0.49, p = 0.04$ ). Besides, the  $\Delta\%$  of sBUN correlated with PL MP<sub>left</sub> ( $r = 0.53, p = 0.02$ ) and PL VL<sub>left</sub> ( $r = 0.47, p = 0.04$ ). Finally, the  $\Delta\%$  of sALB correlated with PL MP<sub>left</sub> ( $r = 0.6, p < 0.01$ ), PL VL<sub>left</sub> ( $r = 0.47, p = 0.04$ ), PL L<sub>1</sub>-L<sub>3</sub> ( $r = 0.52, p = 0.03$ ) and PL T<sub>2</sub>-T<sub>4</sub> ( $r = 0.49, p = 0.04$ ) (see Figure 3).



**Figure 3.** Correlation between percentage of change (pre vs. post<sub>0h</sub>) of serum values and accelerometric load (impacts and Player Load). \* significant correlation  $p < 0.05$ . PL = player load, T<sub>2</sub>-T<sub>4</sub> = thoracic 2nd-4th, L<sub>1</sub>-L<sub>3</sub> = lumbar 1st-3rd; VL<sub>right</sub> and VL<sub>left</sub> = vastus lateralis and MP<sub>right</sub> and MP<sub>left</sub> = malleolus peroneus.

#### 4. Discussion

The purpose of this study was to globally assess heat strain, dehydration, and mechanical load variables as AKI indicators in amateur endurance trail athletes during a 35.3 km run. A significant change between pre vs. post<sub>0h</sub> assessments in all serum biomarkers (sCr, sCK, sBUN, sALB, Cr<sub>clearance</sub>) was found, and its percentage of change correlated with mechanical load indicators (Player Load as accelerometry-based load index). A total of

40.0% and 23.4% of cases presented concomitant proteinuria and hematuria, respectively, due to the trail running. Pain and perceived exertion changed significantly after running. Considering selected reference criteria, there was an incidence of 63.0% (19/30) out of range cases in sCr, 40.0% (12/30) in sBUN, 13.0% (4/30) in sALB, 90.0% in Cr<sub>Clearance</sub> (27/30), and 30.0% (10/30) in sGFR after endurance running. These out-of-range cases in the AKI biomarker indicators returned to baseline after 24 h, suggesting transitional AKI. The outcomes of this study should be analyzed based on two considerations, mean change between timepoint measures and increase or decrease in cases out of range based on clinical and technical criteria. The isolated analysis of statistical differences could lead to misunderstanding the results and the effect of endurance trail running on AKI.

As found in this study ( $F = 5.4\text{--}34.5$ ,  $p < 0.01$ ), previous evidence has suggested an increase in biomarkers related to AKI after endurance running events [3]. Distance running events are considered one of the most physically demanding sports [3,22]. Consequently, relative common increases in sCK values during these kinds of events may indicate a high muscle damage rate due to the release of sarcoplasmic proteins into the bloodstream. Damage and disintegration of muscle fibers are common during strenuous physical exertion [34,35]. The structural and functional damage suffered during running could be exacerbated due to the repetitive concentric–eccentric muscle contractions when running uphill and downhill as in endurance trail events [4,6]. This kind of effort requires greater impact absorption and higher metabolic rate [4,6,11].

The physical effort and biomechanical characteristics of trail running could explain the relation found ( $r = 0.47\text{--}59$ ,  $p < 0.05$ ) between mechanical load indicators (e.g., impacts and Player Load) registered in different body segments and some AKI biomarkers (sBUN and sALB) [7,11]. Indeed, the mechanical load could increase some kidney functional and subclinical injury biomarkers as sCr, sBUN, sALB [11,36,37], as well as some other serum and urinary kidney function indicators as Cr<sub>Clearance</sub>, eGFR, hematuria, and proteinuria; similar changes to those found in this study [8,19]. In this sense, previous evidence has suggested muscle and kidney mechanical trauma in non-contact sports as endurance running, explained by the high magnitude and number of forces involved during trail running [11,38]. Running downhill, sudden changes of direction, jumps, stops, and other high-intensity actions during trail running could partially mediate the renal dysfunction, considering potential kidney shacking [11,38].

The data presented in this study reinforces the hypothesis of potential cumulative kidney mechanical trauma [11,39], by presenting relationships between the change of sBUN and sALB with the mechanical load of the lumbar region (accelerometer in L<sub>1</sub>–L<sub>3</sub>). It has been found that, due to higher intensity and higher external load presented in shorter and faster endurance events, runners usually have a more significant impact on kidney health than more extended events [8,9,11,37]. Running speed seems to be a crucial factor in developing the temporary reduction in kidney function due to strenuous exercising [9,37].

Additionally, based on selected AKI diagnosis criteria, the change in sCr may suggest a risk of AKI after running endurance trail running, but with a return to baseline values after 24 h, as evidenced in previous studies [3,37]. The increased sCr, sBUN, sCK, sALB, eGFR, Cr<sub>Clearance</sub>, and sBUN/sCr ratio, with values over the upper reference limits supports this finding [12,40]. Moreover, the increase in urine hematuria and proteinuria reinforces the evidence that AKI is a transitional condition that could be relatively common after strenuous efforts [22,24]. Endurance trail running may induce a decrease of renal perfusion and a disruption of physiological mechanisms that may maintain glomerular filtration during exercise. This effect is corrected until regular kidney function is achieved, apparently after 24 h [8,37].

In addition to the mechanical load, it has been reported that heat strain and dehydration during these kinds of events can boost the prevalence of AKI in endurance running as it has been reported in other populations [16,19,41], due to the decreased blood plasma and consequent reduction in renal blood flow [19]. In the present study, despite relatively favorable weather conditions (thermal index 24.31 °C) compared to other reported studies



during endurance running, and considering the out-of-range cases, 66.7% of total participants presented significant dehydration [26] immediately after the race. Still, a 46.7% of the participants remained dehydrated 24 h after the event. It should also be considered that there were *ad libitum* food availability and fluid intake during the race. However, although dehydration could typically contribute to AKI, it seems to have a mild impact, as previously reported [42].

Anecdotally, the change ( $F = 4.2\text{--}176.4$ ,  $p < 0.01$ ) in perceived pain and effort values during the race can be a complementary analysis to the biochemical, mechanical, and thermal load assessments to monitor endurance athletes throughout an event to prevent the appearance of AKI. Lumbar pain and its relationship with AKI biomarkers should be explored in future studies.

### Limitations

While the results of this study have provided information regarding the influence of heat strain, mechanical load and dehydration in the development of AKI, some limitations to the study must be acknowledged. One of the main limitations of this study concerns the relatively small sample; it would be interesting to extend this research to include more participants. Secondly, we must bear in mind that these findings can only be extrapolated to male amateur well-trained runners. Future exploration may analyze AKI in top-elite runners, master athletes, and women populations. Furthermore, the influence of physical fitness and experience on the prevalence and development of AKI should be explored among runners.

Some contextual factors as hydration, food intake, and supplements during running should be controlled in future studies. Moreover, despite its limited access, it may be interesting to assess some novel AKI indicators as Cyst-C, NGAL, and KIM-1 as subclinical AKI markers. It is fundamental to develop a cohort follow-up to confirm the potentiality of cumulative AKI events leading to CKD.

## 5. Conclusions

Endurance running could lead to some temporal dysfunctions, such as AKI. These complications were reflected by the increase of some serum changes as sCr, sCK, sBUN, sALB, eGFR, Cr<sub>Clearance</sub>, and sBUN/sCr ratio. This rising pattern was found in proteinuria and hematuria cases Post<sub>0h</sub>. These increased serum and urine values returned to baseline after 24 h, which may suggest transitional AKI with no additional complications. Perceptual variables as effort and pain increased gradually throughout the race may be related to muscle damage and fatigue. Higher mechanical load (Player Load) correlated significantly with the percentage of change (Pre vs. Post<sub>0h</sub>), sCK, sBUN, and sALB; this was evidence for mechanical muscle damage and strengthened the hypothesis of kidney trauma due to cumulative low to moderate contusions during running [11].

## 6. Practical Applications

Considering endurance trail running could increase some blood and urine samples related to transitory AKI, some factors such as dehydration, heat strain, and mechanical load should be monitored during training and competition to prevent potential future damage.

Managing fluid intake and restoring electrolytes before, during, and after endurance events may reduce the number or lessen the severity of AKI cases. Avoiding repeated endurance events without the required rest and recovery between exhaustive efforts could be protective against AKI.

Although there is insufficient evidence to relate AKI events with chronic kidney diseases, medical staff and trainers may monitor renal health of long-distance runners frequently to detect any change in renal function that could affect runners' health.

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Article

# Effect of Compensatory Mechanisms on Postural Disturbances and Musculoskeletal Pain in Elite Sitting Volleyball Players: Preparation of a Compensatory Intervention

Eliza Gawel\* and Anna Zwierzchowska

Institute of Sport Sciences, The Jerzy Kukuczka Academy of Physical Education in Katowice, 40-065 Katowice, Poland; a.zwierzchowska@awf.katowice.pl

\* Correspondence: elizagawel77@gmail.com

**Abstract:** The aim of the study was to identify the effect of compensatory mechanisms on the prevalence of sagittal spinal curvature deformity and musculoskeletal pain and to assess the interrelationships between those components in sitting volleyball players. Twenty-one elite Polish sitting volleyball players (age =  $34.1 \pm 7.5$ , BM =  $77.9 \pm 16.0$ ) participated in the study in which direct participatory systematic observation and a non-invasive method were used. Both objective (anthropometric, spinal curvature—Idiag M360) and subjective (musculoskeletal ailments—NMQ = 7) measurements were performed. The Statistica 13.3 software package was used for statistical analyses. The neck, lower back (43%), and upper back (38%) were the most frequently reported painful areas. Of all participants, 76% reported sagittal spinal deformities. In the habitual position, the results indicated moderate correlations ( $r = 0.5, p < 0.05$ ) between the lumbar concavity of the back and low back pain (LBP) and between thoracic convexity and LBP ( $r = 0.4, p < 0.05$ ). Internal and external compensation have an effect on the prevalence of spinal curvature deformities in the sagittal plane, with thoracic hyperkyphosis (38%) and lumbar hyperlordosis (33%) being the most common. More severe lower and upper back pain were correlated with greater angles of thoracic kyphosis and lumbar lordosis in the habitual position.

**Keywords:** spinal curvature; Paralympic volleyball; compensation strategy; thoracic hyperkyphosis; adapted training; low back pain

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## 1. Introduction

Body posture is affected by many factors. However, it is mostly determined by the shape of the spine [1], which comprises the opposing curves, i.e., kyphosis and lordosis. In a balanced spine, thoracic kyphosis and lumbar lordosis are intrinsically related, and therefore, one curvature responds to the development or disturbance of the other. Furthermore, the pelvic position strongly interacts with the spinal shape by controlling the sagittal balance between the aforementioned curvatures [2,3].

Since physical activity has been acknowledged to impact spinal curvature, athlete body posture has become an area of interest for numerous scientists [1,4,5]. According to Grabara [4], sport-specific training causes multiple changes in an athlete's body build and posture, which leads to the use of adaptative strategies, even if they are not necessarily beneficial. According to Paralympic athletes, two interdependent (internal and external) mechanisms are important. Internal compensation is a necessary yet only partly beneficial compensation strategy due to a congenital or acquired impairment. However, it mostly disturbs the proper function of movement in the human body, such as trunk rotation or pelvic flattening. On the other hand, external compensatory mechanisms are developed due to the specificity of the sport practiced, which is known as the body's adaptation to the sport-specific movements. Despite the fact that the abovementioned compensatory

mechanisms are essential for Paralympic athletes to keep the upright position and sagittal balance of the spine (internal strategy) and to meet the requirements of the sport-specific technique (external strategy), there are several disadvantages that need to be addressed. With the focus on sport-specific functional and structural movements and high training loads, athletes develop muscular dystonia and structural changes in the skeletal system; as a consequence, athletes are prone to musculoskeletal ailments [6].

As pain is known to be one of the most common problems in professional sport [7,8], there is a need for studies that address the possibilities to avoid or reduce musculoskeletal pain and the negative effects of body compensation strategies, especially in Paralympic sport. These problems are especially important in sitting volleyball players since the vast majority of them have lower body impairments [9], especially amputations or limb deficiencies. These types of disabilities activate several internal compensatory mechanisms because of the changed position of the center of gravity of the body [10]. Furthermore, in sitting volleyball players, the upper limbs are constantly overloaded because of sport-specific movements, e.g., services or attacks [11], and the necessity of playing in a sitting position. Therefore, external compensatory mechanisms such as muscle imbalance are often observed in this group [11].

It should be noted that a disabled athlete cannot control his or her physiological limitations caused by a congenital or acquired disability. However, the athlete can choose to avoid or manage musculoskeletal pain and attempt to minimize the negative effects of internal and external adaptative strategies.

To the best of our knowledge, no studies in the currently available scientific literature have examined musculoskeletal pain in relation to spinal curvatures. Therefore, the aim of our study was to evaluate the effect of internal and external compensatory mechanisms on the prevalence of spinal curvature deformities in the sagittal plane and musculoskeletal pain and to assess the interrelationships between the aforementioned components. We hypothesized that lower limb disability and sitting volleyball training impact the spinal curvature in the sagittal plane. Furthermore, it was established that spinal deformities are interrelated with the prevalence of musculoskeletal pain. We assumed that the findings of our study would indicate the need for developing an adapted training program with compensatory proprioceptive exercises that could be implemented in the future as an intervention for sitting volleyball players.

## 2. Materials and Methods

### 2.1. Participants

The study examined twenty-one elite Polish sitting volleyball players ( $n = 6$  women;  $n = 15$  men) from the Polish national team. The inclusion criteria were (a) at least a minimal disability (MD) according to the World ParaVolley classification and (b) no neuromuscular or musculoskeletal disorders. Table 1 shows a description of the participants.

The amputee group used prostheses ( $n = 11$ ) or orthopedic crutches ( $n = 2$ ) in the activities of daily living and locomotion. Only one athlete had a bilateral amputation above the knees and used a wheelchair in everyday life. The athletes from the Les Autres group used prostheses ( $n = 3$ ), orthopedic crutches ( $n = 1$ ), or no supportive equipment ( $n = 3$ ).

The measurements were carried out during a five-day national team training camp in the Jerzy Kukuczka Academy of Physical Education in Katowice, Poland. The participants were informed about the advantages and disadvantages of the study and provided written informed consent. The research protocol was approved by the Bioethics Committee for Scientific Research at the Academy of Physical Education in Katowice, Poland (No. 9/2012) and met the ethical standards of the Declaration of Helsinki, 2013. The participants were allowed to withdraw from the study at any moment. Furthermore, they were instructed to keep their normal dietary and sleeping habits for 24 h before the study.

**Table 1.** Characterization of the sitting volleyball players.

Characteristics ( <i>n</i> = 21; <i>n</i> W = 6, <i>n</i> M = 15)	Mean ± SD or Percentage
Age (years)	34.1 ± 7.5
Body mass (kg)	77.9 ± 16.0
Body height * (cm)	178.6 ± 0.1
Hip circumference (cm)	103.3 ± 10.0
Waist circumference (cm)	89.3 ± 11.1
BMI with a limb deficiency ( <i>n</i> = 16)	23.7 ± 4.9
BMI without a limb deficiency ( <i>n</i> = 5)	24.9 ± 1.9
BAI* (%)	24.8 ± 3.8
Disability time (years)	20.2 ± 11.1
Experience in sitting volleyball training (years)	8.1 ± 7.6
<b>Medical Classification</b>	
Amputees in general	62%
Amputees–A1	5%
Amputees–A2	28.5%
Amputees–A4	28.5%
Les Autres in general	38%
Les Autres–LA5	33%
Les Autres–LA6	5%

*n*—total number of participants; *n*W—number of women; *n*M—number of men; SD—standard deviation; \* excluded bilateral amputation; BMI—body mass index; BAI—body adiposity index; A1—bilateral thigh amputation; A2—lateral thigh amputation; A4—lateral shank amputation; LA5—limited efficiency in one lower limb; LA6—incapacity in one upper limb.

## 2.2. Methods and Measurements

A direct participatory systematic observation method was used in the study, which requires the direct participation of the studied group and the researcher, who directly assesses the participants. The Nordic Musculoskeletal Questionnaire [12] was employed to assess the prevalence and locations of musculoskeletal pain from the last seven days (NMQ = 7) and included the following nine body parts: neck, shoulders, upper back, elbows, wrists, low back, hips/thighs, knees, and ankles/feet. Before completing the questionnaire, the athletes were instructed not to report phantom pain. Next, anthropometric measurements were taken (Figure 1). A wall-mounted stadiometer with a centimeter scale was used for body height (BH) measurements, including the wheelchair user who was able to stand on amputation stumps. Body mass (BM) was evaluated with a chair weight. Hip (HC) and waist (WC) circumferences were measured with the use of anthropometric tape on bare skin, in a lying position and according to the recommended anthropometric techniques, i.e., HC, around the greatest convexity of the gluteal muscles below the iliac ala and WC, at the midpoint between the superior iliac crest and the lowest rib [13]. Spinal curvatures were evaluated using a non-invasive method with a Medi Mouse (Idiag M360) (Figure 1), which ensures producibility, even if two different researchers perform the measurement. The examinations were conducted in three different trunk positions, i.e., sagittal standing (arms in the habitual position), sagittal standing flexion (arms in free stance), and extension (arms crossed at the shoulders, elbows up). Before the measurements, all procedures were demonstrated and explained. The measurements started by putting the Medi Mouse at the C7 level. Next, the device was moved with constant speed up to the S5 level. All measurements were automatically recorded on a computer with Idiag M360 software, which indicates the values from anteroposterior spinal curvatures, physiological values, the differences between them, and the type of sagittal spinal deviation (thoracic hypo/hyperkyphosis, lumbar hypo/hyperlordosis) based on individual BH, BM, gender, age, and the values from anteroposterior spinal curvatures in a habitual position.



**Figure 1.** Examples of anthropometric measurements and spinal curvature measurements.

### 2.3. Statistical Analysis

Statistical analyses were performed with Statistica 13.3 software package (TIBCO Software Inc., Tulsa, OK, USA). Results are presented as means  $\pm$  SD for normally distributed data and as geometric means with a 95% confidence interval. The prevalence of faulty body posture in the sagittal plane and its relation to symptoms in different parts of the musculoskeletal system in the group of Paralympic athletes was compared using statistic structure index (SSI).

Pearson's correlation coefficients were computed for the characteristics of NMQ = 7 and the parameters from Medi Mouse, recorded in different positions (sagittal standing upright, sagittal standing flexion, and extension) for the group of Paralympic athletes. The normality of thoracic kyphosis and lumbar lordosis distributions was verified with the Chi-square test. The level of statistical significance was set at 5%.

### 3. Results

Table 2 presents objective results obtained from the Medi Mouse (thoracic kyphosis and lumbar lordosis angles, physiological values, and differences between actual and physiological values of the aforementioned curvatures in three positions in the sagittal plane) and subjective results of the prevalence and location of musculoskeletal pain based on the NMQ = 7. Table 3 shows the results of statistical correlations between NMQ = 7 and angles of anteroposterior spinal curvatures and differences between physiological norms of thoracic kyphosis and lumbar lordosis (sagittal standing, sagittal standing flexion, sagittal standing extension) based on the Medi Mouse.

The neck (43%), lower back (43%), and upper back (38%) were the most often reported painful areas, whereas the lowest prevalence of pain was found for shoulders, elbows, and ankles/feet (19%). Furthermore, based on the individual reports obtained from the Idiag M360 software, sagittal spinal deviations were found in the vast majority of sitting volleyball players (76%), i.e., thoracic hyperkyphosis (38%), lumbar hypolordosis (33%), thoracic hypokyphosis (19%), and lumbar hyperlordosis (14%).

In the habitual position, the results indicate moderate correlations ( $r = 0.5, p < 0.05$ ) between the deepening of lumbar lordosis and low back pain (LBP) and between deepening thoracic kyphosis and LBP ( $r = 0.4, p < 0.05$ ). Similar moderate relationships ( $r = 0.4, p < 0.05$ ) were found for the sagittal standing extension. Moreover, a correlation between neck pain and the thoracic kyphosis angle was found in both sagittal standing flexion and extension ( $r = 0.4, p < 0.05$ ). Furthermore, the statistical analysis showed a moderate relationship between the prevalence of upper back pain and physiological norms of thoracic kyphosis ( $r = 0.4, p < 0.05$ ).

**Table 2.** Means and standard deviations (SD) of angles of anteroposterior spinal curvatures (°) in three sagittal positions and the prevalence (%) and locations of musculoskeletal pain based on NMQ = 7.

Spinal Curvature Measurements: Sagittal Plane (n = 21; nW = 6, nM = 15)		Mean ± SD (°)	Body Parts (NMQ = 7)	(n = %)
TK–sagittal standing		37.1 ± 18.8	Neck	43%
Physiological values		38.8 ± 18.9		
Difference		1.7 ± 2.6		
TK–sagittal standing flexion		49.6 ± 23.7	Shoulders	19%
Physiological values		51.2 ± 24.9		
Difference		3.7 ± 5.0		
TK–sagittal standing extension		30.4 ± 15.3	Upper back	38%
Physiological values		30.2 ± 16.2		
Difference		2.5 ± 3.3		
LL–sagittal standing		18.9 ± 13.5	Elbows	19%
Physiological values		20.5 ± 14.2		
Difference		1.6 ± 2.7		
LL–sagittal standing flexion		21.8 ± 13.5	Wrists	24%
Physiological values		20.0 ± 12.2		
Difference		3.7 ± 5.0		
LL–sagittal standing extension		29.8 ± 16.4	Lower back	43%
Physiological values		29.6 ± 15.5		
Difference		2.6 ± 3.3		
			Hips/ties	24%
			Knees *	29%
			Ankles/feet *	19%

TH—thoracic kyphosis; LL—lumbar lordosis; n—total number of participants; nW = number of women; nM = number of men; SD—standard deviation; NMQ = 7—Nordic Musculoskeletal Questionnaire from last seven days; \*—one participant did not respond due to bilateral amputation above the knees.

**Table 3.** The results of statistical correlations between the prevalence and location of musculoskeletal pain (NMQ = 7) and angles of anteroposterior spinal curvatures and differences between physiological norms of thoracic kyphosis and lumbar lordosis angles (Medi Mouse).

Body Parts (NMQ = 7)	Sagittal Standing				Sagittal Standing Flexion				Sagittal Standing Extension			
	TH	±	LL	±	TH	±	LL	±	TH	±	LL	±
Neck	0.3	0.3	SI	SI	0.4	SI	−0.09	SI	0.4	0.2	0.2	SI
Arms	0.3	SI	SI	SI	0.3	SI	SI	SI	SI	SI	SI	SI
Upper back	SI	0.4	0.3	0.3	0.16	SI	0.15	−0.1	0.15	SI	0.2	SI
Low back	0.4	−0.2	0.5	−0.2	0.2	SI	SI	−0.1	0.4	−0.2	0.4	−0.2

TH—thoracic kyphosis; ±—the difference between physiological norm and TH or LL angle; SI—statistically insignificant; LL—lumbar lordosis.

#### 4. Discussion

This study aimed to evaluate the effect of internal and external compensation on the prevalence of musculoskeletal pain and postural defects in elite Polish sitting volleyball players and to assess the interrelation between the aforementioned components. A major finding of this study was that the deeper the thoracic kyphosis and lumbar lordosis angles were, the higher was the prevalence of LBP reported in sagittal standing and sagittal standing extension. Furthermore, neck pain occurred more frequently in athletes with a deeper angle of thoracic kyphosis in both sagittal standing flexion and extension. Moreover, the statistical analysis showed direct proportional associations between upper back pain and physiological norms of the thoracic kyphosis angle.

The results of this study fully support our initial hypothesis and confirm that both lower limb deficiency or disability and sitting volleyball training impact the prevalence of



spinal deviations in the sagittal plane. Furthermore, our results point out that anteroposterior spinal curvature deviations are interrelated with musculoskeletal pain, especially in the lower back (43%), neck (43%), and upper back (38%).

Many studies have been carried out to assess the prevalence of musculoskeletal pain in elite able-bodied volleyball players, in whom the lower back was found to be the most common location of pain [14–18]. Moreover, studies by Movahed et al. [19] showed that a greater angle of lumbar lordosis in a habitual position is associated with a higher prevalence of LBP in volleyball athletes. This result corresponds with our findings; however, LBP also contributed to a deepening of lumbar lordosis in sagittal extension and a deepening of thoracic kyphosis in both sagittal standing and extension. These findings may be related to both internal and external compensatory mechanisms that might have impacted muscle imbalance and caused spinal deviations in the sagittal plane, observed in 76% of Paralympic athletes.

It needs to be noted that sagittal balance depends on the angles of thoracic kyphosis and lumbar lordosis, whereas pelvic position strongly interacts with spinal shape by regulating the sagittal balance between the curves [2]. Moreover, the available scientific studies indicate that lower limb/limbs amputation disturbs body biomechanics [10,20]; thus, to maintain balance and upright posture, the human system must activate internal compensatory mechanisms, even if this is not fully beneficial. Unilateral limb amputation/impairment affects the spinal curvature mostly by deepening thoracic kyphosis and flattening lumbar lordosis, as reported in the vast majority of sitting volleyball players.

Furthermore, adaptation to sitting volleyball training, i.e., external compensation strategy, should also be mentioned. In the currently available scientific literature, several studies have analyzed the impact of sport-specific training on the prevalence of anteroposterior spinal curvature deviations in volleyball players [4,21,22]. However, it is difficult to find a study that confirms such effects in Paralympic athletes, especially amputees. Nevertheless, Grabara [4,21] indicated volleyball training as a factor activating the external compensation strategy by deepening thoracic kyphosis and consequently flattening or deepening lumbar lordosis, which is consistent with the results of our study and indicates lumbar hypolordosis (33%) and thoracic hyperkyphosis (38%) as the most common sagittal spinal deformities in sitting volleyball players. Additionally, upper back pain appeared mostly in Paralympic athletes with incorrect values of thoracic kyphosis (38%), which corresponds with the studies of Fett et al. [23] who found the relationships between volleyball training and the high prevalence of upper back pain.

Moreover, a specific sitting position that is taken while playing sitting volleyball should be emphasized. According to the World ParaVolley rules, players can move on the court by sliding or using their upper limbs; however, at least one part of the player's buttocks must remain on the floor while the ball is in play [24]. Because of the forced sitting position, players have a tendency to overload upper limbs and develop muscular imbalances [11] that might strongly contribute to both a deepening of thoracic kyphosis and pain in an athlete's upper body, which was found in this study.

To date, plenty of research has analyzed the prevalence of musculoskeletal pain in volleyball players, which was found mostly in the upper and lower back [25–27]. However, few studies have indicated neck pain as a significant problem in volleyball players [18,24]. Nevertheless, it is hard to find a study that demonstrates the relationships between neck pain and sagittal spinal deformities. Our results have shown a moderate correlation between neck pain and thoracic hyperkyphosis in the sagittal standing flexion and extension, which might be a consequence of the aforementioned compensation strategies.

The findings of our study may be taken into consideration by sitting volleyball players, who are characterized by the high prevalence of musculoskeletal pain and spinal curvature deformities. Therefore, we recommend, especially for Paralympic athletes, an adapted compensatory training program with proprioceptive exercises (Table A1, Appendix A), which was programmed based on the obtained subjective and objective results to prevent or reduce deformities of spinal curvature and musculoskeletal pain.

### Limitations

It should be noted that our study has several limitations that need to be acknowledged. Firstly, even though we explored the entire men's (n = 15) and women's (n = 6) Polish sitting volleyball national team, the group of participants consisted in large part of men, which leads to incomplete inference, especially regarding differences in the prevalence of spinal curvature deformities between the two genders. However, it should be noted that the female sitting volleyball national team made its debut twelve years after the male team [28], which may be associated with a smaller number of elite female athletes. Furthermore, we examined athletes only from two disability groups (amputees, Les Autres).

Secondly, the programmed compensatory exercise intervention has not yet been verified. However, it was developed according to the newest trends in kinesitherapy and corrective methodology. Simultaneously, the authors are planning its verification after extending the group of participants to those with other disabilities, e.g., spinal cord injuries. Such studies will provide important information to improve athletic performance through the prevention of musculoskeletal pain and to reduce the negative effects of internal and external compensation strategies.

### 5. Conclusions

1. Internal and external compensation have an effect on the prevalence of deformities of spinal curvature in the sagittal plane, with thoracic hyperkyphosis (38%) and lumbar hyperlordosis (33%) being the most common.
2. The neck, lower back (43%), and upper back (38%) were the most frequent painful areas in sitting volleyball players. More severe LBP and upper back pain were correlated with a greater angle of thoracic kyphosis and lumbar lordosis in the habitual position.
3. The findings of the study have inspired the programming of an adapted compensatory training program to decrease and prevent the abovementioned spinal deformities and musculoskeletal pain.

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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 Institutional Review Board at the Academy of Physical Education in Katowice, Poland (No. 9/2012).

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

**Data Availability Statement:** The data presented in the study are available on request from the corresponding author.

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

Appendix A

Table A1. Adapted training program with compensatory, proprioceptive exercises for sitting volleyball players.

Exercise Number	The Kind of Exercise	Compensatory Influence	Initial Number of Series and Repetitions	Exercise Process-Version A (Easy)	Exercise Process-Version B (Difficult)	Comments
1.	Mobilization & breathing exercise (thoracic segment)	- Strengthening breathing muscles (inspiratory/ expiratory) - Stretching chest muscles - Thoracic spine mobilization	2 × 10	I.P. 90/90 sit, arms behind the neck, elbows inside. Movement: 1-4. Deep breath through the nose with progressive backward trunk bending and side elbow abduction. 5-8. Frontal trunk bending with deep exhale through the mouth and inside elbow adduction. E.P. = I.P.	I.P. 90/90 sit, arms crossed on the chest Movement: 1-4. Deep breath through the nose while going up with progressive side arm abduction and backward trunk bending. 5-8. Frontal trunk bending with a deep exhale through the mouth and crossing the arms across the chest while going down to the initial position. E.P. = I.P.	- The inspiratory and expiratory phase should last 4 seconds.
2.	Mobilization & breathing exercise (upper limbs)	- Enhancement of the range of motion in the humeral joint - Stretching chest muscles - Strengthening breathing muscles (inspiratory/ expiratory)	The side without a disfunction 2 × 10 The side with a disfunction 2 × 15	I.P. Lying sideways (left side), legs bent at the knee joints, arms in the front, hands together. Movement: 1-4. Side move of the right arm from the front to the right side while turning the head to the right and taking a deep breath through the nose. 5-8. Side move of the right arm from the right side to the front while turning the head to the left and exhaling deeply through the mouth. E.P. = I.P.	I.P. Lying sideways (left side), legs bent at the knee joints, arms in the front, hands together. Movement: 1-4. Side move of the right arm from the front to the back with a hand rotation to the dorsal position while turning the head to the right and taking a deep breath through the nose. 5-8. Side move of the right arm from the back to the front with a hand rotation to the areal position while turning the head to the left and exhaling deeply through the mouth. E.P. = I.P.	-Versions A and B are performed on both sides. - The inspiratory and expiratory phase should last 4 seconds.

Table A1. Cont.

Exercise Number	The Kind of Exercise	Compensatory Influence	Initial Number of Series and Repetitions	Exercise Process–Version A (Easy)	Exercise Process–Version B (Difficult)	Comments
3.	Mobilization exercise (lower limbs)	- Enhancement of the range of motion in the hip joints - Stretching the ilio-lumbar and quadriceps muscles	The side without a disfunction 2 × 10 The side with a disfunction 2 × 15	I.P. Seated frontal bend, arms on the floor. Movement: 1. Right leg abduction to the floor. 2. Right leg adduction to the initial position. 3. Left leg abduction to the floor. 4. Left leg adduction to the initial position. E.P. = I.P.	I.P. Seated frontal bend, arms to the side. Movement: 1. Right and left leg abduction to the floor (movement to the right). 2. Leg adduction to the initial position. 3. Left and right leg abduction to the floor. (movement to the left) 4. Leg adduction to the initial position. E.P. = I.P.	
4.	Activation exercise (upper limbs)	- Rotator cuff activation - Balance the shoulder blade rhythm - Thoracic spine activation	Version A 3 × 10 Version B 2 × 8	I.P. Lying on the front, arms overhead, hands vertical, forehead on the floor. Movement: 1. Raising the arms upwards. 2. Lowering the arms downwards while bending the elbow joints and rotating the hands to the dorsal position. 3. Raising the arms upwards while extending the elbow joints and rotating the hands to the initial position. 4. Lowering arms downwards. E.P. = I.P.	I.P. Lying on the side, arms overhead, hands vertical, forehead on the floor. Movement: 1. Raising the arms upwards. 2. Adduction of the arms sideways. 3. Internal hand rotation to the dorsal position 4. Bending arms at the elbow joints with a side move to the thoracic spine. 5. Lowering the elbows downwards. 6. Raising the elbows upwards with an arm extension to the side (hands in internal rotation). 7. Lifting the arms upwards from the front while rotating the hands to the initial position 8. Lowering arms downwards. E.P. = I.P.	- Before each repetition–retraction and depression of the scapula.

Table A1. Cont.

Exercise Number	The Kind of Exercise	Compensatory Influence	Initial Number of Series and Repetitions	Exercise Process–Version A (Easy)	Exercise Process–Version B (Difficult)	Comments
5.	Activation exercise (lower limbs)	- Gluteus muscle activation - Central stability	The side without a disfunction 3 × 6 The side with a disfunction 3 × 10	I.P. 90/90 sit, front foot in the dorsal position, arms between the knee joint. Movement: 1. Raising the front leg (bent at the knee joint) upwards. 2. Lowering the front leg to the initial position. E.P. = I.P.	I.P. Four-point kneeling Movement: 1. Moving the left leg backward. 2. Holding. 3. Moving the left leg sideways. 4. Lowering the left leg downward. 5. Moving the left leg sideways. 6. Moving the left leg backward. 7. Holding. 8. Lowering the left leg to the initial position. E.P. = I.P.	- Before each repetition–scapula protraction. - Versions A and B are performed on both sides. -Version B–lumbar spine and pelvis without extreme rotation.
6.	Activation exercise (trunk)	- Abdominal muscles activation	Version A 2 × 8 Version B The side without a disfunction 2 × 8 The side with a disfunction 2 × 12	I.P. Lying on the front, arms crossed on the chest. Movement: 1. Raising the trunk upwards. 2–3. Holding. 4. Lowering the trunk downwards. 5. Raising and turning the trunk to the left side. 6. Lowering the trunk to the initial position. 7. Raising and turning the trunk to the right side. 8. Lowering the trunk to the initial position. E.P. = I.P.	I.P. Lying on the back with the legs bent at the knee joints (feet in a dorsal position), left arm upwards, right arm on the left knee joint– pushing slightly. Movement: 1. Lowering the left arm and the right leg downwards (to the straight body level). 2. Raising the left arm and the right leg upwards to the initial position. E.P. = I.P.	- Version A: raising and lowering the trunk, vertebra by vertebra. - Version B: the exercise is performed on both sides, and the lumbar spine should globally touch the floor during the entire motor activity.

Table A1. Cont.

Exercise Number	The Kind of Exercise	Compensatory Influence	Initial Number of Series and Repetitions	Exercise Process–Version A (Easy)	Exercise Process–Version B (Difficult)	Comments
7.	Directional exercise (thoracic segment)	<ul style="list-style-type: none"> <li>- Stretching chest muscles</li> <li>- Strengthening the latissimus dorsi muscle and teres major muscle</li> <li>- Strengthening the rotator cuff</li> <li>- Balance the shoulder blade rhythm</li> </ul>	3 × 10	<p>I.P. Kneeling sit, arms downwards, hands are holding a resistance band (hips widthways).                      Movement:                      1. Moving the right arm from the front to the back.                      2. Moving the right arm from the back to the front.                      E.P. = I.P.</p>	<p>I.P. Kneeling sit, arms downwards, hands are holding a resistance band (hips widthways).                      Movement:                      1. Moving the arms from the front to the back.                      2. Moving the arms from the back to the front.                      E.P. = I.P.</p>	<ul style="list-style-type: none"> <li>- Before each repetition–retraction and depression of the scapula.</li> <li>- During the exercise, there should not be any compensation with trunk articulation in the lumbar spine</li> <li>- Version A: the exercise is performed on both sides. During the entire movement activity, the band should be maintained in a slight tension.</li> <li>- Version B: during the entire movement activity, the band should be maintained with the same tension.</li> </ul>
8.	Directional exercise (thoracic segment)	<ul style="list-style-type: none"> <li>- Stretching chest muscles</li> <li>- Strengthening the quadratus lumborum muscle, rhomboid muscle, and latissimus dorsi muscle</li> <li>- Strengthening the rotator cuff</li> <li>- Balance the shoulder-blade rhythm</li> </ul>	3 × 10	<p>I.P. 90/90 sit, arms in the front (head level), hands are holding the resistance band (shoulders widthways).                      Movement:                      1. Pulling the band from the front to the side.                      2. Returning to the initial position.                      E.P. = I.P.</p>	<p>I.P. 90/90 sit, arms upwards, hands are holding the resistance band (shoulders widthways).                      Movement:                      1. Pulling the band downwards.                      2. Returning to the initial position.                      E.P. = I.P.</p>	<ul style="list-style-type: none"> <li>- Before each repetition–scapulas retraction and depression.</li> <li>- During the entire movement activity, the band should be kept with a slight tension.</li> </ul>

Table A1. Cont.

Exercise Number	The Kind of Exercise	Compensatory Influence	Initial Number of Series and Repetitions	Exercise Process—Version A (Easy)	Exercise Process—Version B (Difficult)	Comments
9.	Directional exercise (lumbar segment)	<ul style="list-style-type: none"> <li>- Strengthening gluteus muscles</li> <li>- Strengthening serratus anterior muscle, superior and external oblique muscles</li> <li>- Central stability</li> </ul>	<p>The side without a disfunction 3 × 10</p> <p>The side with a disfunction 3 × 15</p>	<p>I.P. Lying sideways (left side) with bent legs, the left arm bent at the elbow joint and lying on the forearm, the right arm bent at the elbow joint (on the trunk level).</p> <p>Movement:</p> <ol style="list-style-type: none"> <li>1. Raising trunk upwards while raising the right leg upwards.</li> <li>2. Holding the trunk with a right arm abduction and while turning the trunk to the right.</li> <li>3. Holding the trunk with a right arm adduction and a trunk flexion to the body level.</li> <li>4. Lowering the trunk and legs to the initial position.</li> </ol> <p>E.P. = I.P.</p>	<p>I.P. Lying sideways (left side) with bent legs, the left arm bent at the elbow joint and lying on the forearm, the right arm upwards.</p> <p>Movement:</p> <ol style="list-style-type: none"> <li>1. Raising the trunk and right leg upwards.</li> <li>2. Holding the trunk while lowering the right leg.</li> <li>3. Holding the trunk while raising the right leg upwards.</li> <li>4. Lowering the trunk and legs to the initial position.</li> </ol> <p>E.P. = I.P.</p>	<ul style="list-style-type: none"> <li>- Before each repetition—retraction and depression of the scapula.</li> <li>- During the motor activity, the trunk should be stabilized.</li> </ul>
10.	Directional exercise (lumbar segment)	<ul style="list-style-type: none"> <li>- Strengthening the rectus abdominis muscle (version A) and superior and external oblique muscles (version B)</li> <li>- Stretching the quadratus lumborum muscle</li> </ul>	3 × 30 s.	<p>I.P. Lying on the back with raised legs bent at the knee joints, arms upwards.</p> <p>Movement:</p> <ol style="list-style-type: none"> <li>1. Raising the trunk while moving the arms downwards (knee joints level, hands in areal position).</li> </ol> <p><i>Alternative 2–3 (30 s.)</i></p> <ol style="list-style-type: none"> <li>2. Trunk flexion to the left side.</li> <li>3. Lowering the legs to the initial position.</li> <li>4. Moving the arms upwards.</li> </ol> <p>E.P. = I.P.</p>	<p>I.P. Lying on the back with raised legs bent at the knee joints, arms upwards.</p> <p>Movement:</p> <ol style="list-style-type: none"> <li>1. Raising the trunk while moving the arms downwards (knee joints level, hands in areal position).</li> </ol> <p><i>Alternative 2–3 (30 s.)</i></p> <ol style="list-style-type: none"> <li>2. Trunk flexion to the left side.</li> <li>3. Trunk flexion to the left side.</li> <li>4. Lowering the legs to the initial position while moving the arms upwards.</li> </ol> <p>E.P. = I.P.</p>	<ul style="list-style-type: none"> <li>- During the exercise, the lumbar spine should globally touch the floor.</li> <li>- When a lumbar lordosis accentuation can be seen, the exercise should be stopped.</li> </ul>

I.P.—internal position; E.P.—end position.

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Communication

# Effects of Maximal Effort Running on Special Agents' Loaded and Unloaded Drop Jump Performance and Mechanics

Justin J. Merrigan

Human Performance Innovation Center, Rockefeller Neuroscience Institute, West Virginia University, Morgantown, WV 26505, USA; justin.merrigan@hsc.wvu.edu

**Abstract:** The purpose was to investigate the effect of load and fatigue on landing forces and mechanics. Thirteen Department of State special agents first completed drop jump testing, a maximal treadmill test, and another round of drop jump testing. During drop jump testing, agents performed 3 maximal effort drop jumps from 30 cm with body mass only (unloaded) or a 15 kg weight-vest (loaded). A force plate was used to collect force–time data, while two laptops were placed 3 m from the force plate from frontal and sagittal planes. Two-way analyses of variance were used to analyze the effect of load and fatigue on landing forces and Landing Error Scoring System (LESS) with alpha of  $p < 0.05$ . Dropping from 30 cm with 15 kg resulted in greater landing impulse, which was driven by increases in contact time. The loaded condition also resulted in lower jump height and reactive strength indexes. After the maximal graded treadmill test there were no further changes in drop jump ground reaction forces or performance. However, relative aerobic capacity was related to impulse changes following the treadmill test in unloaded ( $R^2 = 0.41$ ;  $p = 0.018$ ) and loaded conditions ( $R^2 = 0.32$ ;  $p = 0.044$ ). External loads of 15 kg increased impulse and contact time and resultantly decreased drop jump height and reactive strength indexes. It is encouraged that training protocols be aimed to concomitantly improve aerobic capacity and lower body power. Plyometric training with progressive overloading using external loads may be helpful, but further research is warranted.

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**Keywords:** occupational health; tactical athlete; landing error scoring system; reactive strength index; tactical personnel; force plates; military; law enforcement; neuromuscular fatigue

## 1. Introduction

Special agents endure intensive physical training to best prepare them for their demanding and unpredictable occupational tasks. During training, 35% of male and 42% of female agents experience one or more injuries, which are most likely to occur at the knees and thighs [1]. These musculoskeletal injuries lead to health consequences for the individual and undue government funding and resources [2]. Risk factors for injury occurrences include traits of physical prowess, such as aerobic capacity [1,3], neuromuscular capabilities (i.e., strength and power) [4], and movement mechanics [3,5]. Injury risk is also heightened during tasks requiring load carriage [6] and/or tasks that acutely induce high levels of fatigue [7,8]. Therefore, strategies have been employed to evaluate physical capabilities in tactical populations, particularly while under external loads or during periods of heightened fatigue [9].

Vertical jump testing is one method used to identify levels of fatigue during sustained operations training in the military [10]. To further evaluate neuromuscular performance capabilities under load and fatigue, jump testing is being conducted on force plates in tactical populations [9,11,12]. The benefit of examining a movement's (e.g., countermovement jump's) force–time characteristics, is the additional data pertaining to the forces and movement strategies required to execute the movement. For example, the individual may adopt different movement strategies (e.g., shorter contraction times, deeper or shallower countermovement depths) in attempt to attain the same maximal effort jump heights as

their last testing session [13]. Although these data are useful for identifying forces generated by or acting on the body and general movement mechanics, further data collection methods are necessary to ascertain movement patterns regarding specific joints.

To identify these biomechanical movement patterns, expensive laboratory motion capture equipment is often used [14]. However, when traditional laboratory equipment is not permitted, practitioners may consider using subjective field tests, such as the Landing Error Scoring System (LESS) [14]. The LESS is a clinical tool for assessing potential errors in movement mechanics (i.e., knee valgus) during landing and jumping tasks through visual inspection of front and side view recordings [15]. The resultant score is a summation of “errors” identified throughout various stages of the movement. Higher scores allude to poorer mechanical movement patterns, which may occur due to fatigue [16] and are associated with knee injuries in tactical populations [5]. Thus, these tools may be useful to help evaluate injury risk factors during jumping and landing tasks.

For example, external loads required of tactical personnel may lower jumping performance [14,17] and alter movement strategies and forces imposed on the individual when landing [9,14,18,19]. These biomechanical analyses informing the effects of external loading may preface the negative influence of external loading on injury risk [20] and performance of high-intensity tactically related duties (e.g., combative movements) [21–23]. Similarly, movement mechanics, such as LESS, have been linked with injury risk [3,5] and are often impaired due to acute bouts of fatiguing tasks [16,24,25]. Peak landing forces have also been increased, alongside incidences of stress fractures, during fatiguing tasks [26]. However, some have failed to find altered vertical ground reaction forces (vGRFs) from exercise induced fatigue [27]. Yet, despite no changes in jump height or peak vGRF following a fatiguing bout of running, knee valgus increased [24]. Thus, a combined analysis of the vGRF and movement mechanics may better inform the cumulative effects of running induced fatigue and external loading, which is necessary to investigate as running prior to landing tasks may impair mechanics and performance in loaded conditions [19].

Physical prowess may also reduce the negative effects of external loading or fatigue on movement patterns. For example, stronger individuals may note less reductions in drop jump performance [14]. Likewise, individual’s that are more aerobically fit require a lower working capacity to achieve the same running outcome as individual’s that are less aerobically fit [6]. Investigating the relationship between aerobic capacity and alterations in movement patterns from fatigue may, in part, help to explain the potential mediation between aerobic capacity as a risk factor for injury [1] and the common occurrence of injuries during load carriage tasks [6]. This may be particularly pertinent to investigate in the Department of State Diplomatic Mobile Security Deployment (MSD) Special Agents who endure intense training to prepare them to operate in high-threat environments with little outside support. These agents are often deployed to global hotspots to be readily available for quick responses to protect U.S. federal government officials from kidnapping and terrorist threats and to protect and evacuate U.S. citizens out of crisis areas. Despite some aspects of the day-to-day operations of MSD special agents being considered sedentary (i.e., screening visitors), their training and operations often involve advanced and precise firearm handling and tactics, close quarters combat, counter-terror tactics, off-road and/or high-speed vehicle operations, advanced navigation, first-aid, and survival capabilities under fatigued and loaded conditions. Thus, the purpose of this study was to investigate the effect of external loading and short bouts of maximal effort running on landing forces and mechanics in MSD agents and whether their aerobic fitness levels would influence the effects of running on drop jump results.

## 2. Materials and Methods

### 2.1. Subjects

According to average effects of training induced fatigue on jump performance from prior research [11], large effects were anticipated revealing an a priori minimum sample size estimate of 12 for the current study. Thirteen Department of State MSD Special

Agents (age,  $37 \pm 5$  years; body mass,  $71.67 \pm 3.81$  kg; height,  $202.11 \pm 28.71$  cm;  $VO_{2max}$ ,  $4.18 \pm 0.63$  L·min<sup>-1</sup>; relative  $VO_{2max}$ ,  $45.50 \pm 4.10$  mL·kg<sup>-1</sup>·min<sup>-1</sup>) participated. All agents had at least 2 years of consistent physical training and were considered healthy in accordance with the physical activity readiness questionnaire (PAR-Q). Agents were asked to refrain from activities that may fatigue musculature and inhibit their ability to perform the current tasks for 48 h prior to arriving to the laboratory (ex. resistance training, high volume or intensity running or occupational tasks). Participants were also asked to adhere to normal sleeping and eating habits and avoid alcohol, tobacco, caffeine, and other ergogenic aids/supplements for at least 3 h before testing.

## 2.2. Design

To determine the effects of short bursts of maximal running on neuromuscular performance and mechanics, special agents completed drop jump testing before and after a maximal treadmill test, separated by 2 min of rest. Prior to testing, agents completed a short dynamic warm-up (5 min cycle and 5 min lower body dynamic stretching) and were familiarized with the drop jump protocols. All testing procedures occurred at approximately the same time of day (1000–1300) under the supervision of certified strength and conditioning specialists (NSCA CSCS).

## 2.3. Maximal Graded Treadmill Testing

During treadmill (ELG, Woodway, Waukesha, WI, USA) protocols, inspired and expired gases were transferred through a two-way valve into a gas analyzer (ParvoMedics TrueOne 2400 Metabolic Cart) to assess aerobic capacity ( $VO_{2max}$ ). Throughout the test heart rate was assessed using a chest strap device (PolarH7, Kempele, Finland). The first and second stages were a warm-up at  $5.0$  km·hour<sup>-1</sup> with 0% grade and  $6.5$ – $8.0$  km·hour<sup>-1</sup> at a 5.2% incline, respectively. The remainder of the maximal treadmill test was performed at 5.2% incline. Speed began at their 2-mile run pace and increased after each one-minute stage by  $1.0$  km·hour<sup>-1</sup> until volitional fatigue. All tests were completed within 7–10 min and considered true maximal tests based on previous criteria, described elsewhere including: plateau of oxygen uptake despite an increase in workload, respiratory exchange ratio above 1.10, achieving 90% of their age estimated max heart rate ( $206.9 - 0.67 \times \text{age}$ ), rating of perceived exertion greater than or equal to 18 (from Borg-scale of 6–20); and a venous blood lactate  $>8$  mM [28]. The blood lactate was collected from a fingertip, cleaned by an alcohol swab, through a small incision from a lancet (Tenderlett; Accriva Diagnostics; San Diego, CA, USA). The initial blood sample was wiped away with medical gauze, and the subsequent drop was used for analysis. A collecting strip (Lactate Plus meter test strips; Nova Biomedical, Waltham, MA, USA) was inserted into a portable lactate analyzer (Lactate Plus meter; Nova Biomedical), which was calibrated with a control solution (Lactate plus control solution level 2; Nova Biomedical) according to factory guidelines.

## 2.4. Drop Jump Testing and Analysis

During drop jump testing, agents performed 3 maximal effort jumps from a 30 cm box without and with a 15 kg weight-vest, in random order using a counterbalanced design (7 participants begin with unloaded condition and 6 with loaded condition). Rest between jumps was 30-s while rest between conditions was 1 min. Agents were instructed to step off, not walk or jump off, the box and immediately perform a maximal effort countermovement jump with little ground contact.

Force–time data were collected from a portable force plate (AccuPower; AMTI, Watertown, MA, USA) via a custom-built interface box with an analog-to-digital card (NI cDAQ-9174; National Instruments, Austin, TX, USA) at 1600 Hz and analyzed using Matlab (version 7.12, MathWorks, R2011a, Natick, MA, USA). The landing phase was identified from ground contact, when forces were  $>5$  standard deviations above the one-second quite weighing phase average, to takeoff, when forces were  $<5$  standard deviations of the quite weighing phase. The following force–time metrics were calculated during the

entire landing contact duration and used in analyses: peak vGRF, maximal vertical ground reaction force; impulse, area under the curve; rate of force development, change in vGRF from contact to 20 milliseconds after contact divided by 20 milliseconds; contact time, duration from contact to takeoff; flight time, time from takeoff to second ground contact; jump height,  $0.5 \times 9.8 \times (\text{flight time}/2)^2$ ; reactive strength index, flight time divided by contact time.

Videos, for LESS, were taken from two laptops (ThinkPad, Lenovo, Morrisville, NC, USA) with the same video recording capabilities and quality (resolution, 720p; frame rate, 30 fps), placed 3 m from the participant in frontal and sagittal planes. Drop jumps were analyzed using computer software (QuickTime; Apple, Inc, Cupertino, CA, USA) from ground contact, frame immediately prior to complete foot contact, to maximal knee flexion, using a scoring sheet described elsewhere [15]. The average total LESS score for each condition was used for analysis with a higher score indicating more landing errors.

### 2.5. Statistical Analysis

Data were considered normally distributed according to Shapiro–Wilks and visual inspection of histograms. Reliability of was calculated across the three trials for each time-point and condition using the coefficient of variation with a threshold of >10% determining an unreliable metric. Two-way analyses of variance were used to analyze the effect of load and fatigue on landing vGRFs and LESS. The association between relative  $\text{VO}_{2\text{max}}$  and the decrease in drop jump performance was assessed via linear regression analyses. Cohen’s *d* effect sizes were calculated with corresponding 95% confidence intervals with the following determinants: <0.2, negligible; 0.20–0.49, small; 0.50–0.79, moderate; >0.80, large. Analyses were conducted using R, version 3.6.2 [29] with  $p < 0.05$ .

## 3. Results

Drop jump force–time metrics were considered reliable, according to coefficient of variation calculations, except for landing rate of force development (impulse,  $3 \pm 3\%$ ; peak cGRF,  $8 \pm 8\%$ ; rate of force development,  $15 \pm 10\%$ ; contact time,  $5 \pm 4\%$ ; reactive strength index,  $7 \pm 4\%$ ; jump height,  $7 \pm 7\%$ ; LESS,  $5 \pm 5\%$ ). There was no significant external load by fatigue interaction ( $p > 0.05$ ). Dropping with 15 kg resulted in greater landing impulse and increased contact time, which resulted in lower jump heights and reactive strength indexes (Table 1). The maximal treadmill test did not alter drop jump vGRFs, mechanics, or performance (Table 2). There was no load by fatigue interaction ( $p > 0.05$ ). However, 95% confidence intervals included large effects of fatigue on landing force, jump height, and LESS (Table 2). Although there were wide confidence intervals, a high percentage of individuals in the group experienced more than  $-10\%$  increases (Table 3) in landing rate of force development, jump height (Figure 1), and RSI (Figure 2), as well as >10% increases in peak vGRF (Figure 3) and LESS (Figure 4). Lastly, relative  $\text{VO}_{2\text{max}}$  was associated with impulse changes from pre- to post-fatigue in unloaded ( $R^2 = 0.41$ ;  $p = 0.018$ ) and loaded conditions ( $R^2 = 0.32$ ;  $p = 0.044$ ) (Figure 5). Relative  $\text{VO}_{2\text{max}}$  did not predict changes in any other force–time metric due to the maximal effort bout of running.

**Table 1.** The result of load on drop jump forces and performance.

	Unloaded	Loaded	Effect Size (CI 95%)
Impulse (N·s)	1043.4 ± 155.3	1242.9 ± 200.0	1.115 ± 0.298 (0.51, 1.68) *
Peak vGRF (N)	3369.4 ± 863.5	3702.8 ± 766.8	0.408 ± 0.280 (−0.15, 0.95)
RFD (N·s <sup>−1</sup> )	4191.3 ± 1384.7	4171.7 ± 1758.1	0.012 ± 0.277 (−0.53, 0.56)
Contact Time (s)	0.638 ± 0.105	0.703 ± 0.124	0.563 ± 0.283 (0.00, 1.11) *
RSI (AU)	0.858 ± 0.209	0.712 ± 0.236	0.653 ± 0.284 (0.09, 1.20) *
Jump Height (cm)	34.96 ± 8.24	28.40 ± 7.24	0.846 ± 0.289 (0.27, 1.40) *
LESS (AU)	5.05 ± 2.73	5.38 ± 2.40	0.130 ± 0.278 (−0.42, 0.67)

Values are mean ± standard deviation for unloaded and loaded (15 kg) conditions. For effect size, values are Cohen’s *D* effect size ± standard error of the effect size estimate (95% confidence intervals, CI 95%). \*, indicates statistical significance. vGRF, vertical ground reaction force; RFD, rate of force development; RSI, reactive strength index; LESS, landing error scoring system, total score; AU, arbitrary units.

**Table 2.** The result of fatigue on drop jump forces and performance.

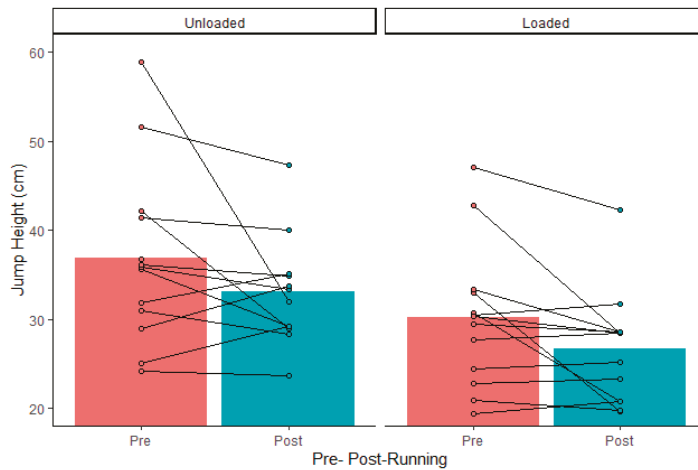
	Pre	Post	Effect Size (CI 95%)
Impulse (N·s)	1143.7 ± 198.5	1142.6 ± 213.1	0.006 ± 0.277 (−0.54, 0.55)
Peak vGRF (N)	3411.2 ± 789.5	3660.9 ± 857.7	0.303 ± 0.279 (−0.25, 0.85)
RFD (N·s <sup>−1</sup> )	4416.9 ± 1412.1	3946.2 ± 1702.7	0.301 ± 0.279 (−0.25, 0.84)
Contact Time (s)	0.675 ± 0.117	0.667 ± 0.122	0.065 ± 0.277 (−0.48, 0.61)
RSI (AU)	0.794 ± 0.238	0.775 ± 0.233	0.081 ± 0.277 (−0.46, 0.62)
Jump Height (cm)	33.53 ± 9.45	29.84 ± 6.79	0.449 ± 0.281 (−0.11, 0.99)
LESS (AU)	4.67 ± 2.65	5.77 ± 2.36	0.439 ± 0.281 (−0.12, 0.98)

Values are mean ± standard deviation for unloaded and loaded (15 kg) conditions. For effect size, values are Cohen’s D effect size ± standard error of the effect size estimate (95% confidence intervals, CI 95%). Indicates statistical significance. vGRF, vertical ground reaction force; RFD, rate of force development; RSI, reactive strength index; LESS, landing error scoring system, total score.

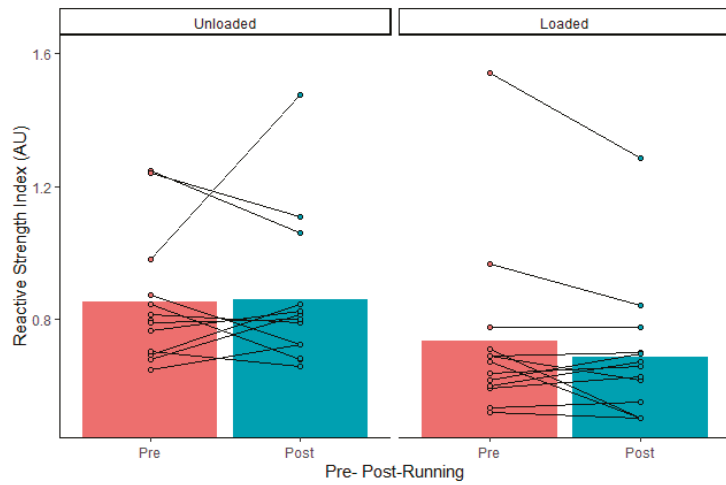
**Table 3.** Number of participants experiencing >10% change in performance due to maximal treadmill running.

Variable	Change	Unloaded # (%)	Loaded # (%)
Impulse	Decrease	1 (8)	1 (8)
Peak vGRF	Increase	6 (46)	6 (46)
RFD	Decrease	7 (54)	7 (54)
Contact Time	Increase	1 (8)	2 (15)
RSI	Decrease	4 (31)	5 (38)
Jump Height	Decrease	3 (23)	4 (31)
LESS	Increase	7 (54)	6 (46)

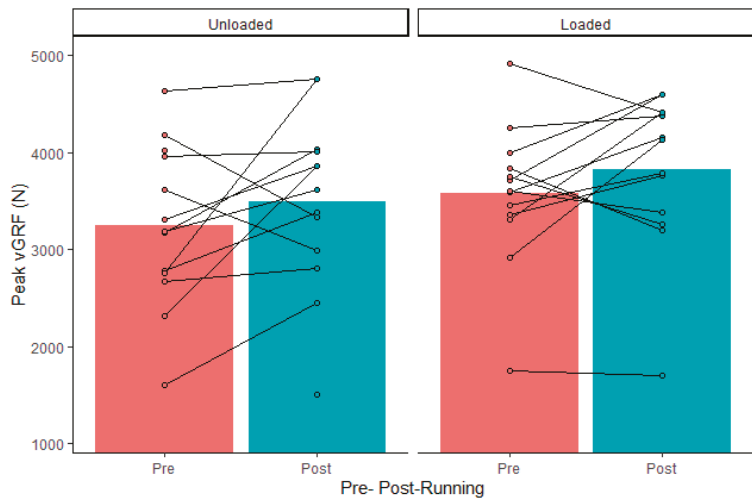
#, number of agents with corresponding percent change out of the 13 total sample. vGRF, vertical ground reaction force; RFD, rate of force development; RSI, reactive strength index; LESS, landing error scoring system, total score.



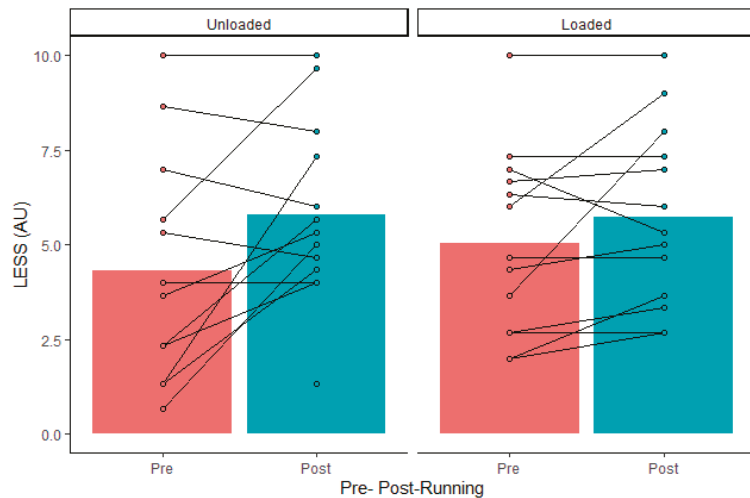
**Figure 1.** Group average (gray bars) and individual data (dots and lines) for drop jump height before (Pre) and after (Post) the maximal graded treadmill test in unloaded and loaded (15 kg) conditions.



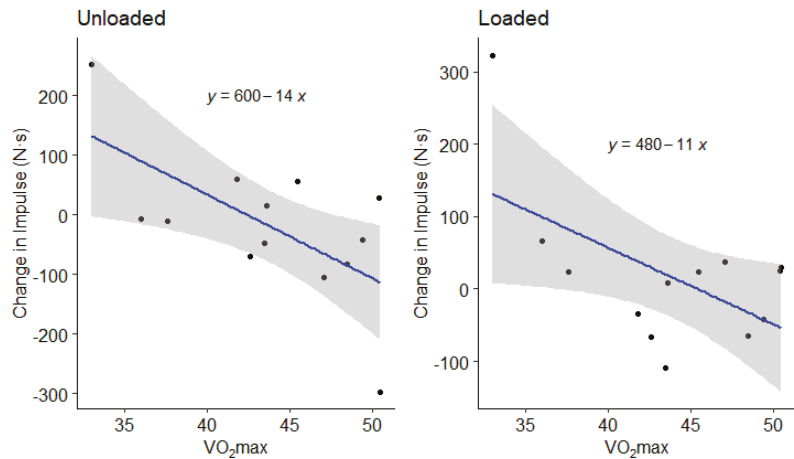
**Figure 2.** Group average (gray bars) and individual data (dots and lines) for drop jump reactive strength index before (Pre) and after (Post) the maximal graded treadmill test in unloaded and loaded (15 kg) conditions.



**Figure 3.** Group average (gray bars) and individual data (dots and lines) for drop jump landing peak vertical ground reaction forces (vGRF) before (Pre) and after (Post) the maximal graded treadmill test in unloaded and loaded (15 kg) conditions.



**Figure 4.** Group average (gray bars) and individual data (dots and lines) for drop jump Landing Error Scoring System (LESS) before (Pre) and after (Post) the maximal graded treadmill test in unloaded and loaded (15 kg) conditions.



**Figure 5.** Group average (gray bars) and individual data (dots and lines) for drop jump Landing Error Scoring System (LESS) before (Pre) and after (Post) the maximal graded treadmill test in unloaded and loaded (15 kg) conditions.

#### 4. Discussion

Body armor can protect against serious trauma, but the external loading likely reduces musculoskeletal capabilities and potentially increases injury risk [30]. The high-risk environments of MSD special agents make the body armor a necessity, subsequently creating a higher physiological demand for all movements by special agents [31]. To combat the additional physiological strain, the agents require high levels of balanced strength and aerobic capacity to withstand the external load during powerful and fatiguing movements. Furthermore, agents may require additional attention to proper movement technique, as improper movement strategies increase the effect of body armor on physiological strain [31].



Thus, it is important to consider the effects of body armor and external loading under a fatigued state, as this will likely have more real-world applications for special agents and other tactical personnel [32]. Considering the prior training and preparation of this unique group of special agents, their daily experiences may alter their responses to external loading and fatigue induced by maximal effort running, compared to previously investigated populations. Moreover, considering the uniqueness of the current group of special agents, they may require specific training needs to prepare them for future operations through improved movement competency, power output, and aerobic capacity. The current study sought to examine the effect of load and fatigue on landing forces and mechanics in Department of State MSD Special Agents.

Generally, when the mass of external loading is increased the physiological strain and decrements in movement capabilities (i.e., jump height) are exacerbated [31]. In prior research, when military personnel were equipped with heavier loads (20 and 40 kg), peak landing forces increased as the external loads became heavier [18]. To reduce the peak forces and enhance energy absorption at the knee while under load (20–40 kg), others found that military personnel relied upon more hip and knee extension when landing from 30 cm, a strategy that may reduce injury risk [33]. Yet, special agents in the current study did not alter movement mechanics according to the LESS, which is in line with prior literature examining the LESS across various loading conditions [32]. Instead, the current special agents adopted slower pacing strategies (longer total contact time) to handle the additional external load, which resulted in greater impulses but not peak vGRFs. Slower pacing strategies adopted during loaded jumping tasks have also been noted in United States Marines and led to reductions in countermovement jump heights and reactive strength indexes with a 10 kg weighted vest and a 20 kg barbell [11]. Other research in Army Reserve Officer Training Cadets also reported slower pacing strategies, but no change in joint angles (i.e., knee valgus), during drop jumps from a 30 cm box with a 15 kg weighted vest [14]. The lower jump heights and reactive strength indexes that coincided with reduced hip, knee, and ankle joint velocities and center of mass velocity, were lower in magnitude for cadets that had greater knee extensor strength [14]. Thus, the overall forces imposed on an individual during landing tasks with external loads, as well as their ability to quickly move after ground contact (i.e., jump height), may be highly influenced by contact times. Thus, tactical personnel may benefit from plyometric training aimed to explosively transition from eccentric to concentric phases to continue to improve their ability to perform landing and jumping tasks, particularly under loaded conditions.

Contrary to the original hypothesis of a greater impact of load carriage following a fatiguing protocol, there was no greater effect of external load after running to voluntary exhaustion. The aforementioned result disagrees with prior hypotheses [32] and findings of a greater impact of external loading on peak vGRFs following a bout of intense running [18]. Furthermore, the current results indicated that short bouts of running to momentary volitional fatigue did not impact landing vGRFs or movement mechanics (LESS). Others have found that an intense run prior to performing drop jumps did not influence performance under unloaded conditions, but did decrease jump height by 6% in loaded conditions ( $7.65 \pm 0.73$  kg) which were 12% lower than unloaded conditions at baseline [19]. Despite the lack of significance in the current study, the 15 kg loaded condition was similarly impacted by running induced fatigue with a 10% decline in jump height compared to 7% in the unloaded condition. In other populations, jump height and landing peak vGRFs were not impacted by running induced fatigue, but knee valgus increased [24]. Notably, and possibly driven by greater knee valgus, prior literature has found fatigued states to result in higher LESS scores [16]. Yet, the current study has demonstrated a wide range of LESS results following maximal treadmill running. This may partially be explained by various intents of jumping, despite instructions to jump as high as possible for every attempt. Resultantly, some may have moved more cautiously with lower jump height performance, while others may have maintained explosiveness at the cost of impaired movement patterns. Another potential explanation for the discrepancies in findings, is that

jumping performance may be more influenced by jumping induced fatigue than running induced fatigue [34]. Thus, more research may be warranted to investigate motor control and performance after various fatiguing events experienced by tactical populations (such as obstacle course trainings involving a combination of movements).

Additionally, individuals with a greater relative  $\text{VO}_{2\text{max}}$  had less of an increase in landing impulse due to the maximal treadmill test. Thus, those with greater aerobic capacity may be less affected by fatigue in unloaded and loaded conditions. This is an important finding as the ability to resist fatigue may be associated with lower risk of lower extremity knee injuries, according to a recent systematic review [35]. With the current findings, it is encouraged that aerobic capacity be trained in conjunction with plyometric training with 15 kg or less to reduce the impact of load carriage and fatigue on landing mechanics and vGRFs. However, despite adequate power, the sample size in the current study was still relatively small and the resultant findings should therefore be considered with caution. Still the findings expand on prior literature and should be used to direct future training studies aimed at improving the efficiency of strength and conditioning program design in tactical populations.

In summary, landing while carrying external loads of 15 kg may result in slower pacing strategies (i.e., prolonged ground contact) that increase landing vGRF impulse and reduce performance capabilities (i.e., jump height and reactive strength). Short bouts of movement to volitional fatigue may not severely impact landing forces or mechanics (LESS) for the entire group, but improvements in aerobic capacities reduces the influence of fatigue on landing impulses. These findings should be considered when implementing physical training protocols for tactical populations that perform occupational tasks under external loads or fatigued states. The current findings suggest that a concomitant training approach towards improvements on reactive strength and lower body power, as well as aerobic capacity, is necessary for tactical personnel. It is also noteworthy that force–time characteristics and jump performance, although not significantly large, may be altered if exercise is conducted prior to force plate testing. Yet, these preliminary findings should be supported by future interventions.

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**Data Availability Statement:** The data presented in this study are available on request from the corresponding author.

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Article

# The Modifications of Haemoglobin, Erythropoietin Values and Running Performance While Training at Mountain vs. Hilltop vs. Seaside

Maria Cristina Man <sup>1</sup>, Cătălin Ganera <sup>2</sup>, Gabriel Dan Bărbuleț <sup>1</sup>, Michał Krzysztofik <sup>3</sup>, Adelina Elena Panaet <sup>4</sup>, Alina Ionela Cucui <sup>5</sup>, Dragoș Ioan Tohănean <sup>6,\*</sup> and Dan Iulian Alexe <sup>7</sup>

- <sup>1</sup> Department of Physical Education, 1 Decembrie 1918 University of Alba Iulia, 510009 Alba Iulia, Romania; cristina.man@uab.ro (M.C.M.); gabriel.barbulet@gmail.com (G.D.B.)
  - <sup>2</sup> Nicolae Rotaru Sports Program High School of Constanța, 900178 Constanța, Romania; cataganera@yahoo.com
  - <sup>3</sup> Institute of Sport Sciences, The Jerzy Kukuczka Academy of Physical Education in Katowice, 40-065 Katowice, Poland; m.krzysztofik@awf.katowice.pl
  - <sup>4</sup> Doctoral School, National University of Physical Education and Sport Bucharest, 060057 Bucharest, Romania; adelina\_panaet@yahoo.com
  - <sup>5</sup> Department of Physical Education and Sports, Valahia University of Targoviste, 130024 Targoviste, Romania; haralambialina2008@yahoo.com
  - <sup>6</sup> Faculty of Physical Education and Mountain Sports, Transilvania University of Brasov, 500036 Brasov, Romania
  - <sup>7</sup> Faculty of Movement, Sports and Health Sciences, VasileAlecsandri University of Bacău, 600115 Bacău, Romania; alexedaniulian@ub.ro
- \* Correspondence: dragos.tohanean@unitbv.ro

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**Abstract:** Altitude training increases haemoglobin, erythropoietin values among athletes, but may have negative physiological consequences. An alternative, although less explored, that has the potential to positively influence performance while avoiding some of the negative physiological consequences of hypoxia is sand training. Ten endurance-trained athletes (age:  $20.8 \pm 1.4$ , body mass:  $57.7 \pm 8.2$  kg, stature:  $176 \pm 6$  cm; 5000 m  $14:55.00 \pm 0:30$  min) performed three 21-day training camps at different locations: at a high altitude (HIGH), at the sea-level (CTRL), at the sea-level on the sand (SAND). Differences in erythropoietin (EPO) and haemoglobin (Hb) concentration, body weight,  $VO_{2max}$  and maximal aerobic velocity (VMA) before and after each training cycle were compared. Data analysis has indicated that training during HIGH elicited a greater increase in  $VO_{2max}$  ( $2.4 \pm 0.2\%$ ;  $p = 0.005$  and  $1.0 \pm 0.2\%$ ;  $p < 0.001$ ) and VMA ( $2.4 \pm 0.2\%$ ,  $p < 0.001$  and  $1.2 \pm 0.2\%$ ;  $p = 0.001$ ) compared with CTRL and SAND. While increases in  $VO_{2max}$  and VMA following SAND were greater ( $1.3 \pm 0.1\%$ ;  $p < 0.001$  and  $1.2 \pm 0.1\%$ ;  $p < 0.001$ ) than those observed after CTRL. Moreover, EPO increased to a greater extent following HIGH ( $25.3 \pm 2.7\%$ ) compared with SAND ( $11.7 \pm 1.6\%$ ,  $p = 0.008$ ) and CTRL ( $0.1 \pm 0.3\%$ ,  $p < 0.001$ ) with a greater increase ( $p < 0.01$ ) following SAND compared with CTRL. Furthermore, HIGH and SAND elicited a greater increase ( $4.9 \pm 0.9\%$ ;  $p = 0.001$  and  $3.3 \pm 1.1\%$ ;  $p = 0.035$ ) in Hb compared with CTRL. There was no difference in Hb changes observed between HIGH and SAND ( $p = 1.0$ ). Finally, athletes lost  $2.1 \pm 0.4\%$  ( $p = 0.001$ ) more weight following HIGH vs. CTRL, while there were no differences in weight changes between HIGH vs. SAND ( $p = 0.742$ ) and SAND vs. CTRL ( $p = 0.719$ ). High-altitude training and sea-level training on sand resulted in significant improvements in EPO, Hb, VMA, and  $VO_{2max}$  that exceeded changes in such parameters following traditional sea-level training. While high-altitude training elicited greater relative increases in EPO, VMA, and  $VO_{2max}$ , sand training resulted in comparable increases in Hb and may prevent hypoxia-induced weight loss.

**Keywords:** altitude; haemoglobin; erythropoietin; hypoxia; endurance; sand

## 1. Introduction

High-altitude training has long been considered to improve the performance of endurance athletes [1]. As the altitude increases, the quantity of oxygen decreases, resulting in adaptations to the conditions of reduced oxygenation [2]. These changes include an increased oxygen-carrying capacity (i.e., increased haemoglobin mass) among other non-haematological changes [2]. Meta-analyses data suggests that the performance of elite endurance athletes can improve by ~4–5% following methods of live high-train low and live high-train high altitude training [3]. However, some athletes may not respond favourably to such practices potentially due to the negative physiological consequences of training in a hypoxic environment (e.g., impaired sleep quality, weight loss, decrease in muscle protein synthesis) [4,5].

An alternative, although less explored, practice that has the potential to positively influence performance while avoiding some of the negative physiological consequences of hypoxia is sand training [6,7]. Evidence suggests that the unique training adaptations for sand may have a positive influence on endurance performance [8,9]. Sand surfaces may provide a training stimulus that elicits a higher energetic cost with less ground reaction force compared to the more traditional training surfaces, such as synthetic fabrics or grass [10]. The high shock absorption capacity of sand may decrease the impact forces experienced during high-intensity activities, which may lead to a reduction in muscle lesions and pain and improve recovery time between sessions [10,11]. Considering these differences, recent evidence supports the use of sand as a training means to improve the performance of team sports athletes [12]. Binnie et al. [7] have reported that for training, the use of sand instead of grass training surfaces elicits a relatively higher training intensity, without causing any additional decrement in performance the following day (24 h post exercise). Furthermore, in a study by Yiğit and Tuncel [13], a significant improvement in predicted  $\text{VO}_{2\text{max}}$  over the 6-weeks was reported in the sand running group but not in the road running group. Therefore, sand has the potential of providing not only a unique training stimulus for athletes but also a viable option for recovery sessions.

Recently, more complex research exploring specific mechanisms that may lead to greater adaptations with sand versus more traditional training surfaces has emerged [7,12–14]. Such studies have quantified the contributions of energetic uptake (aerobic and anaerobic) during short-term exercising in the sand (namely 10 min) [14]. A study by Binnie et al. [7] investigated the effect of sand surfaces during a training session comprising running at different speeds for a longer period (60 min). Specifically, these studies compared the use of sand and grass surfaces during training sessions [7,9,11,15]. For a session functioning with standardised intervals, the use of sand instead of grass or synthetic surfaces led to a significantly higher average heart rate (sand: 172 bpm; grass: 163 bpm) and blood lactate values (sand:  $10.1 \text{ mmol} \cdot \text{L}^{-1}$ , grass:  $6.5 \text{ mmol} \cdot \text{L}^{-1}$ ) throughout the training session [13,16].

Given that the energy consumption of sand running is higher compared to training on traditional training surfaces, such as grass, this may lead to greater physiological adaptation over a specific training period. Therefore, we sought to compare differences in haemoglobin (Hb), erythropoietin (EPO) concentrations and running performance (maximal aerobic velocity [VMA] and capacity [ $\text{VO}_{2\text{max}}$ ]) in highly trained runners following 21-day high-altitude, sea-level/sand, and traditional sea-level training cycles. We hypothesized that high-altitude and sea-level training on sand will significantly increase blood parameters and physical performance in comparison to the sea-level training, while a slightly greater improvement will be noted after high-altitude training.

## 2. Materials and Methods

### 2.1. Participants and Study Design

Participants were: 10 male athletes (age:  $20.8 \pm 1.40$ , body mass:  $57.7 \pm 8.2 \text{ kg}$ , stature:  $176 \pm 6 \text{ cm}$ ), who specialised in middle-distance and long-distance events, mountain

running, with 5000 m 14:55.00  $\pm$ 0:30" at the Romanian National Championships and at International Championships. The main inclusion criteria were: a compete in the national-level event and that the participants were free from musculoskeletal injuries for at least 6 months before each training cycle of the study. The athletes went through the same training program for 21 days across three training cycles separated by one year. Standard meals were provided to the athletes at each training camp. Additionally, all athletes adhered to the same regimen of vitamin and mineral supplementation. Anthropometric measures, blood draws, and running performance tests were taken on the day before and the first day after each 21-day training program to determine changes in body weight, haemoglobin concentration, and running performance. Blood samples were taken following a 12 h fast at the same time of day ( $\pm$ 1 h) at each timepoint. Samples were stored for <48 h at 2–8 °C prior to analyses. A timeline of assessments taken before and after each 21-day training program is provided in Table 1.

**Table 1.** Graph of evaluations.

Blood Sampling		Anthropometric Measurements		Physical Tests to Determine VO <sub>2max</sub> /VMA	
Before 21-day training program	After 21-day training program	Before 21-day training program	After 21-day training program	Before 21-day training program	After 21-day training program
31 July 2017 8:00 o'clock	22 August 2017 8:00 o'clock	31 July 2017 8:30 o'clock	22 August 2017 8:30 o'clock	31 July 2017 16:00 o'clock	22 August 2017 16:00 o'clock
31 July 2018 8:00 o'clock	22 August 2018 8:00 o'clock	31 July 2018 8:30 o'clock	22 August 2018 8:30 o'clock	31 July 2018 16:00 o'clock	22 August 2018 16:00 o'clock
31 July 2019 8:00 o'clock	22 August 2019 8:00 o'clock	31 July 2019 8:30 o'clock	22 August 2019 8:30 o'clock	31 July 2019 16:00 o'clock	22 August 2019 16:00 o'clock

VMA = maximum aerobic velocity. VO<sub>2max</sub> = maximal aerobic capacity.

## 2.2. Training Programs

The athletes covered three training periods, as follows: In the first stage, the athletes carried out a training cycle for 21 days at Piatra Arsă, at an altitude of ~2000 m. In the second stage, the same group of athletes carried out the same training routine at the seaside, on the Black Sea coast, at Mamaia-Constanța (~0 m). Finally, the third training cycle took place in Blaj, at an altitude of ~600 m.

The timing and geographical locations of each training cycle were:

- 1 August 2017–22 August 2017, National Sports Complex "Piatra Arsă" of the Bucegi Mountains (altitude; HIGH G1)
- 1 August 2018–22 August 2018, on the Black Sea coast, in Mamaia-Constanța (sea-level/SAND; G2)
- 1 August 2019–22 August 2019, in Blaj, the Alba County (traditional sea-level, 600 m CTRL)

A sample training program from the "Piatra Arsă" cycle is detailed in Table 2. Similar training programs in terms of effort zones were used during the training periods on the Black Sea Coast and in the locality of Blaj, with the exception that within the training period at the seaside the accommodation period was reduced from 7 days (at high altitude) to 3 days and during the research stage at Blaj there was no accommodation period.



Table 2. Training program: Piatra Arsă (2000 m).

Day	Piatra Arsă-2000 m Altitude	Total Km-Running
1	T.S. <sub>1</sub> 8 km e.r. 50%VMA/T.S. <sub>2</sub> 8 km e.r. segment strength-65%VMA	16
2	T.S. <sub>3</sub> 10 km e.r. 50%VMA/T.S. <sub>4</sub> 6 km e.r. and 3 complete strength series-65% VMA	16
3	T.S. <sub>5</sub> 12 km r. uniform tempo, 65%VMA, segm. strength/T.S. <sub>6</sub> 10 km r. uniform tempo, 10 × 100 m a.l.-65% VMA	22
4	S.T. <sub>7</sub> 16 km r. various land, segment strength and r.l.-70%VMA	16
5	S.T. <sub>8</sub> 20 km r. various land-75% VMA/S.T. <sub>9</sub> - 8 km e.r. 3 series of ex. for strength	28
6	S.T. <sub>10</sub> 16 km r. uniform tempo. 70%VMA/S.T. <sub>11</sub> 10 km r. uniform tempo., 10 × 100 m r.l.-70% VMA	27
7	S.T. <sub>12</sub> 14 km r. uniform tempo., 75%VMA	14
8	S.T. <sub>13</sub> 16 km r. progressive various land 75–83% VMA/S.T. <sub>14</sub> 10 km r. uniform tempo., -70% VMA and 3 series of ex. for strength	26
9	S.T. <sub>15</sub> 6 km r. uniform tempo., 65%VMA, 20 × 100 m r. accelerated (100%VMA) with connection 100 m e.r. 4 km/S.T. <sub>16</sub> 10 km r. uniform tempo., stretching	24
10	S.T. <sub>17</sub> 10 km e.r., stretching 75%VMA/S.T. <sub>18</sub> 40 min r. (2 min r. tempo sustained + 1 min conn.+1 min r tempo sustained +1 min. connection) × 8 series (90%VMA)	26
11	S.T. <sub>19</sub> 15 km r. various land (75–80%VMA)	15
12	S.T. <sub>20</sub> 10 km r. various land and 10 × 100 m r.l. with 100 m e.r. 80%VMA/S.T. <sub>21</sub> 10 km r. uniform tempo., (75%VMA)	24
13	S.T. <sub>22</sub> 12 km r. tempo. progressive-88–93% VMA/S.T. <sub>23</sub> 10 km e.r. (75%VMA)	26
14	S.T. <sub>24</sub> 16 km r. various land 80% VMA/S.T. <sub>25</sub> 10 km r. uniform tempo., (75%VMA)	26
15	S.T. <sub>26</sub> 14 km r. various tempo 92–94%VMA, 1 km e.r.	27
16	S.T. <sub>27</sub> 10 km e.r. 75%VMA/S.T. <sub>28</sub> 14 km e.r. segment strength (60%VMA)	24
17	S.T. <sub>29</sub> 26 km r. various land (65%VMA)	26
18	S.T. <sub>30</sub> 6 km e.r. 15 × 100 m with 100 m (95%VMA), 3 km e.r./S.T. <sub>31</sub> 10 km r. uniform tempo, 70%VMA	22
19	S.T. <sub>32</sub> 8 km e.r.75%VMA/S.T. <sub>33</sub> 3 km e.r. 20 × 300 m with connection 100 m e.r. (30 sec) 100%	20
20	S.T. <sub>34</sub> 2 hours' walk- forest (2000 m/600 m altitude)/beach (0 m altitude)	0
21	S.T. <sub>35</sub> 15 km r. 80%VMA/S.T. <sub>36</sub> 10 km r. uniform tempo., 75%VMA	25

r. = running; e.r. = easy running; r.l. = running launched; S.T.<sub>1</sub> = training session 1; VMA = maximum aerobic velocity.

### 2.3. Anthropometrics

The bodyweight of participants was determined in the morning, using the same digital scale (ZET27288; SC Zetman Kraft SRL, Arad, Romania) and recorded to the nearest 100 g.

### 2.4. Haemoglobin Concentration

Hb was determined from venous blood collected before and after each training program using a Diagon D-Cell 60 automated haematology analyser (Diagon Ltd., Budapest, Hungary).

### 2.5. Erythropoietin Concentration

The serum concentration of EPO was determined by the commercially available Human EPO Quantikine™ IVD ELISA Kit (R&D Systems, Minneapolis, MN, USA).

### 2.6. Maximal Aerobic Velocity and Capacity

The five-minute test consisted of the athletes running at full capacity for five minutes. VMA = distance obtained [in km] × 12. The five-minute test was used to determine VMA and the formula VMA × 3.5 was used to predict VO<sub>2max</sub> because all subjects were over 18 years of age [17]. The previous study confirms it as a reliable and practical indirect method to estimate individual aerobic fitness in a trained population [18].

Chamoux et al., 1996 [19,20] mention that the VMA determined on the terrain depends on the duration of the effort and therefore on the protocol used. Examining the relationship between running speed and running time log established from world racing records foot shows a significant point at 4.97 min, proposed as the reference time for VMA. By convention, VMA could be measured on the field by a 5 min test regardless of the sport.

## 2.7. Statistical Analyses

Statistical analyses were performed using SPSS software (v.26, IBM, Armonk, NY, USA). Repeated measures ANOVA was used to determine whether characteristics (e.g., body weight, [Hb], [EPO], VMA,  $VO_{2max}$ ) remained similar prior to each 21-day training camp. Changes in these characteristics across each training camp were detected using one-way ANOVA. Repeated measures ANOVA were further used to determine differences in the relative change (% difference from pre-training) in characteristics across training camps. A Bonferroni correction was used for multiple pairwise comparisons and when sphericity was violated, the Greenhouse–Geisser corrected was used. Normality was assessed using Shapiro–Wilk’s test. Statistical significance was set a priori at  $p < 0.05$ .

## 3. Results

### 3.1. Sample Size

The sample size analysis was performed using G\*Power software (Dusseldorf, Germany). Given the study 2-way analysis of variance (ANOVA) (three conditions and two repeated measures), a moderate overall effect size (ES) = 0.58, an alpha-error < 0.05, the desired power (1- $\beta$  error) = 0.8, and correlation among repeated measures = 0.5, the total sample size resulted in nine participants. This value of effect size was chosen according to the improvements in  $VO_{2max}$  after training on the sand from Binnie et al. [7].

### 3.2. Anthropometrics

The body weight of the athletes was similar prior to each 21-day training camp ( $p = 0.133$ ). Body weight decreased by  $1.3 \pm 0.6$  kg ( $p < 0.001$ ), from  $57.7 \pm 8.2$  kg before the training period to  $56.4 \pm 8.0$  kg after HIGH. There was no change in body weight from before the training period to after SAND ( $p = 0.149$ ; pre =  $57.8 \pm 7.9$  kg vs. post =  $57.0 \pm 7.8$  kg) or from before the training period to after CTRL ( $p = 0.504$ ; pre =  $59.4 \pm 8.3$  kg vs. post =  $59.3 \pm 8.1$  kg).

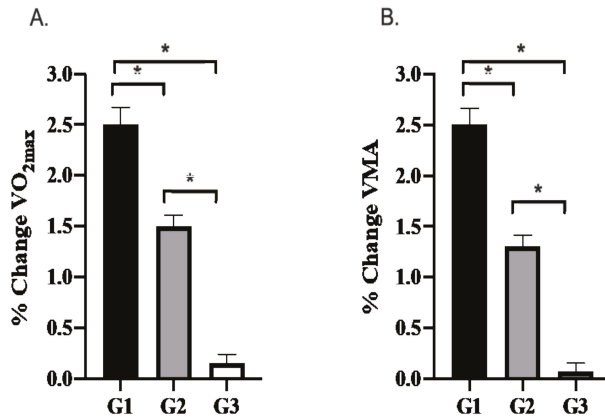
There tended to be an overall difference ( $p = 0.59$ ,  $\eta_p^2 = 0.318$ ,  $1-\beta = 0.507$ ) in the percentage of body weight change from pre- to post-training between training cycles. Athletes lost  $2.1 \pm 0.4\%$  ( $p = 0.001$ ) more weight following HIGH vs. CTRL, while there were no differences in weight changes between HIGH vs. SAND ( $p = 0.742$ ) and SAND vs. CTRL ( $p = 0.719$ ).

### 3.3. Maximal Aerobic Velocity and $VO_{2max}$

$VO_{2max}$  was higher ( $p = 0.015$ ) before the HIGH ( $66.7 \pm 0.6$  mL/kg/min) vs. CTRL ( $63.3 \pm 0.8$  mL/kg/min) training period.  $VO_{2max}$  before SAND ( $65.4 \pm 0.6$  mL/kg/min) was not different from values before HIGH ( $p = 0.142$ ) or CTRL ( $p = 0.231$ ). Similarly, MAV was higher ( $p = 0.015$ ) before HIGH ( $19.06 \pm 0.18$  km/h) vs. CTRL ( $18.10 \pm 0.23$  km/h). MAV before SAND ( $18.70 \pm 0.18$  km/h) was also not different from HIGH ( $p = 0.096$ ) or CTRL ( $p = 0.135$ ).

$VO_{2max}$  increased by  $1.7 \pm 0.4$  mL/kg/min following G1 ( $p < 0.001$ ) and by  $1.0 \pm 0.2$  mL/kg/min following G2 ( $p < 0.001$ ). VMA increased by  $0.48 \pm 0.10$  km/h following G1 ( $p < 0.001$ ) and by  $0.24$  km/h following G2 ( $p < 0.001$ ). However, training during G3 did not elicit a change in  $VO_{2max}$  ( $p = 0.146$ ) or VMA ( $p = 0.452$ ).

There was an overall effect of training cycle on the relative change in  $VO_{2max}$  ( $p < 0.001$ ,  $\eta_p^2 = 0.896$ ,  $1-\beta = 1.0$ ) and VMA ( $p < 0.001$ ,  $\eta_p^2 = 0.969$ ,  $1-\beta = 1.0$ ). Training during G1 elicited a  $2.4 \pm 0.2\%$  greater increase in  $VO_{2max}$  ( $p = 0.005$ ) and VMA ( $p < 0.001$ ) compared with G3. Training during G1 also elicited a  $1.0 \pm 0.2\%$  greater increase in  $VO_{2max}$  ( $p < 0.001$ ) and a  $1.2 \pm 0.2\%$  greater increase in VMA ( $p = 0.001$ ) compared with G2. Increases in  $VO_{2max}$  following G2 were  $1.3 \pm 0.1\%$  greater ( $p < 0.001$ ) and in VMA were  $1.2 \pm 0.1$  ( $p < 0.001$ ) than those observed after G3. Figure 1 shows the relative change in performance data following each 21-day training camp.



**Figure 1.** The relative change (pre-post) in (A) VO<sub>2</sub>max and (B) VMA following 21-days of training at altitude (G1), seaside (G2), and sea-level (G3). Values displayed are the means ± SD. Significant differences ( $p < 0.05$ ) between training camps are denoted as (\*).

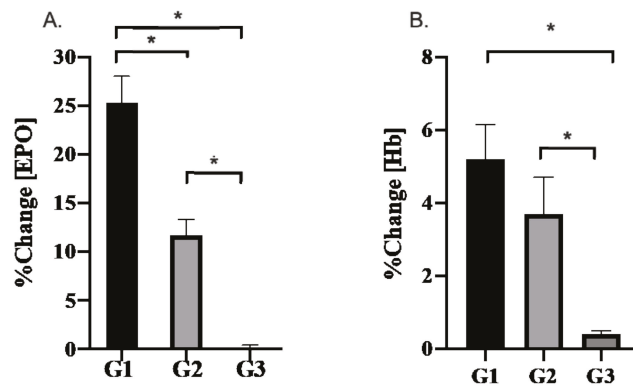
### 3.4. Erythropoietin Concentrations

EPO values differed prior to the start of each training camp ( $p = 0.003$ ) with lower values noted before the start of G1 ( $5.5 \pm 2.1$  mU/mL) compared to G2 ( $8.2 \pm 0.6$  mU/mL;  $p = 0.03$ ) and G3 ( $9.0 \pm 0.5$  mU/mL;  $p = 0.009$ ). Increases in EPO were elicited only with G1 ( $p < 0.001$ ; post =  $6.9 \pm 2.8$  mU/mL) and G2 ( $p < 0.001$ ; post =  $9.2 \pm 1.9$  mU/mL) training cycles. Whereas EPO were unchanged after G3 ( $p = 0.678$ , post =  $9.0 \pm 1.7$  mU/mL).

Training cycle had a significant effect on the relative change in EPO ( $p < 0.001$ ,  $\eta_p^2 = 0.829$ ,  $1-\beta = 1.0$ ). EPO increased to a greater extent following G1 ( $25.3 \pm 2.7\%$ ) compared with G2 ( $11.7 \pm 1.6\%$ ,  $p = 0.008$ ) and G3 ( $0.1 \pm 0.3\%$ ,  $p < 0.001$ ). Further, the relative increase in EPO was greater ( $p < 0.01$ ) following G2 compared with G3.

### 3.5. Haemoglobin Concentrations

Hb measured before the start of each training camp were similar ( $p = 0.145$ ). Each training camp provoked increases in Hb with a  $0.8 \pm 0.4$  g/dL increase observed after G1 ( $p < 0.001$ ; pre =  $14.4 \pm 0.9$  g/dL vs. post =  $15.2 \pm 1.0$  g/dL), a  $0.5 \pm 0.4$  g/dL increase after G2 ( $p < 0.001$ ; pre =  $13.8 \pm 1.1$  g/dL vs. post =  $14.3 \pm 0.8$  g/dL), and a  $0.1 \pm 0$  g/dL increase after G3 ( $p = 0.003$ ). Training cycle had a significant effect on the relative change in Hb ( $p < 0.001$ ,  $\eta_p^2 = 0.884$ ,  $1-\beta = 1.0$ ). G1 elicited a  $4.9 \pm 0.9\%$  greater increase ( $p = 0.001$ ) in Hb compared with G3. G2 elicited a  $3.3 \pm 1.1\%$  greater increase ( $p = 0.035$ ) in Hb compared with G3. There was no difference in Hb changes observed between G1 and G2 ( $p = 1.0$ ). Figure 2 displays the relative change in EPO and Hb following each training period.



**Figure 2.** The relative change (pre-post) in (A) [EPO] and (B) [Hb] following 21-days of training at altitude (G1), sea level (G2), and sea-level (G3). Values displayed are the means  $\pm$  SD. Significant differences ( $p < 0.05$ ) between training camps are denoted as (\*).

#### 4. Discussion

This study aimed to evaluate the changes in EPO, Hb concentrations and running performance (VMA and  $VO_{2max}$ ) following 21-day high-altitude, on the sand at sea level, and traditional sea-level training cycle. Our primary findings are that both, high-altitude training and sea-level training on sand resulted in significant improvements in EPO, Hb, VMA, and  $VO_{2max}$  that exceeded changes in such parameters following traditional sea-level training. While high-altitude training elicited greater relative increases in EPO, VMA, and  $VO_{2max}$ , sand training resulted in comparable increases in Hb and may prevent hypoxia-induced weight loss. The main conclusions of this study confirmed our initial hypothesis and suggest that the physiological modifications triggered by 21-days of either high-altitude training or training on the sand at sea level are greater than those attained by traditional sea-level training methods.

The effects of altitude training have been widely examined [21–26], however, there is scarce research on the training on the sand [7,9,15]. Training on soft sand led to a significant increase in heart rate ( $HR > 22$  bpm), oxygen uptake rate ( $VO_2 > 0.872$  L·min<sup>-1</sup>) compared to a hard surface, thus clearly demonstrating a higher level of physiological effects experiences while exercising on sand compared to traditional surfaces [10]. Moreover, when comparing the training on the sand and on the grass, similar levels of haemolysis (serum haptoglobin concentration) were reported when they are followed by physical exercise. [27]. Exercise-induced haemolysis, the moment when the red blood cells are destroyed, is characterised by the free increase in haemoglobin and by a decrease in blood haptoglobin provides an alternative method of measuring the degree of stress on the musculoskeletal system during exercise [28] or competitive stress [29]. Foot problems are a major cause of haemolysis, and this appears during running sessions. The variables which are involved in training sessions such as the type of terrain and the experience can influence the quantity of haemolysis. Consequently, training on sand may lead to lower levels of haemolysis suffered during exercise, due to the decrease in impact forces experienced when touching the ground. Alternatively, haemolysis may increase with a higher frequency of the contacts with the ground; it has also been proven to increase with the intensity of exercise possibly due to the compression of capillary networks, by activating lean mass to a higher degree or due to higher tissue hypoxia levels, leading to RBC oxidative stress [30]. Furthermore, the increase in EPO and haemoglobin values after training at the seaside can be the consequence of several factors: running on the sand [9,15] negative ions [31] present in large quantities on the seashore, amounts similar to altitude, wind (sea breeze) that can cause hypoxia.

Moreover, training on sand leads to an increased alteration of kinematic running (cadence, joint angles, etc), muscle activation patterns [32] and physiological responses [16].

The numerous studies on the effects of training in hypoxia show that one of the most important effects of altitude training concerns the increase in haemoglobin mass [33–38]. The mechanism of improvement seems to stem from an increase in EPO driven by stabilization of its transcription factor hypoxic inducible factor- $\alpha$  in the low PO<sub>2</sub> environment [33]. As such, a greater O<sub>2</sub> carrying capacity with chronic exposure to hypoxic environments with altitude training practices have been cited to contribute to improved submaximal and maximal exercise capacity [34]. For example, it has been shown—in a study conducted for 27 days at 2500 m—that the concentration of haemoglobin increases regularly, due to EPO concentration, featuring a 100% increase on the first day of stimulation, while the concentration of soluble receptors recorded 19% after only 19 days [39]. Wehrlin and Marti, 2006 [40] have shown—upon analysing two world-class runners in 5000 m and marathon—increases of 3.9 and 7.6% in haemoglobin mass and 5.8 and 6.3% in erythrocytes after a live high-train low training camp that lasted 28 days, 18 h/day, at an altitude of 2456 m, with training at 1800 m. This finding is interesting and practical because it shows a positive effect among world-class athletes for whom inducing even a slight gain in fitness level is a challenge. In a different study, the same authors [40] have studied well-trained triathletes and a control group: they have concluded, also using the live-high and train-low method for the altitude of 2500 m, an 18 h/day and training in 1800 and 1000 m, an increase in haemoglobin mass by 5.3% and of red cells by 5.0%, as well as improving sports performance by increasing VO<sub>2max</sub> values by 4.1%. Comparing this study with the one we conducted, we see similar changes. At 2000 m altitude, after 21 days of preparation, haemoglobin increased by 4.9% and VO<sub>2max</sub> values by 2.4%.

Although most studies indicate favourable physiological and physical changes in increasing athletes' performance, altitude training also has negative effects such as a decrease in body weight (in the present study by 2.1%) [41–44]. However, no such differences were observed in the current study after training on the sand. Therefore, this training approach induces a comparable increase in Hb concentration to altitude training and may prevent hypoxia-induced weight loss. Nevertheless, it must be mentioned that high-altitude training elicited greater relative increases in EPO, VMA, and VO<sub>2max</sub>.

Considering the training studies carried out thus far, additional research is necessary to determine a whole array of benefits associated with training on the sand. Namely, the main characteristic associated with exercising on sand is higher movement energy cost [11,15] and the capacity of reaching higher training intensities during a training session is higher compared to training on a traditional surface [45,46]. Consequently, additional research is required to investigate the implications of training on the sand, with a focus on aerobic and muscular adaptations and the causes that trigger changes in the concentration of EPO and haemoglobin.

Our study is not without limitations. Although the study was conducted on elite runners, the sample size was small. Moreover, we have measured Hb concentration rather than Hb mass. Furthermore, the EPO concentrations were significantly lower prior to the high-altitude training compared with other training—this could have possibly led to the greater increases seen in this parameter following training. In addition, the differences in weather—the higher ambient temperatures on the seaside—could have led to changes in Hb mass (w/heat training).

## 5. Conclusions

The results of this study suggest that the physiological modifications triggered by 21-days of either high-altitude training or training on sand at sea-level are greater than those attained by traditional sea-level training methods. However, it must be mentioned that high-altitude training elicited greater relative increases in EPO, VMA, and VO<sub>2max</sub>, while sand training resulted in comparable increases in Hb and may prevent hypoxia-induced weight loss. These findings are of practical value as they suggest that training on

sand at sea-level can be considered as an alternate for high-altitude training. In this regard, when the introduction of high-altitude training is impossible, the coaches and athletes could incorporate training on sand at sea-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 Commission of University Ethics and Professional Deontology within Vasile Alecsandri University of Bacău (no. 9425/2/2021).

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

**Data Availability Statement:** The datasets used and analysed during the current 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 the Result of Soccer Matches on the Heart Rate Variability of Women Soccer Players

Rosa M<sup>a</sup>. Ayuso-Moreno <sup>1</sup>, Juan Pedro Fuentes-García <sup>1,\*</sup>, Hadi Nobari <sup>2,3</sup> and Santos Villafaina <sup>4</sup>

<sup>1</sup> Faculty of Sport Science, University of Extremadura, Avda. Universidad S/N, 10003 Cáceres, Spain; roayusom@alumnos.unex.es

<sup>2</sup> Sports Scientist, Sepahan Football Club, Isfahan 81887-78473, Iran; hadi.nobari1@gmail.com

<sup>3</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain

<sup>4</sup> Physical Activity and Quality of Life Research Group (AFYCAV), University of Extremadura, 10003 Cáceres, Spain; svillafaina@unex.es

\* Correspondence: jpfuent@unex.es

**Abstract:** The present study aimed to evaluate the effects of a match lost and a match won on post-competitive heart rate variability (HRV) in semi-professional female soccer athletes. A total of 13 players, with a mean age of 23.75 (5.32), from the Cáceres Women Football Club of the Spanish Second National Division participated in our study. They were evaluated in two microcycles which correspond to a match lost and a match won. For each microcycle, baseline and post-competitive measures were collected. Results indicate that HRV was significantly reduced before a match lost and won. Significant differences in HRV variables were observed when compared the lost match, and the match won. Results highlight the importance and usefulness of analyzing the HRV as an indicator of post-competitive fatigue in semiprofessional soccer players. Therefore, a competition's results could be considered a relevant variable to consider when programming training load.

**Keywords:** female; football; autonomic modulation; fatigue; training load

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## 1. Introduction

Biological signals are used as tools for controlling and evaluating training loads and acute and chronic effects on the athlete's body [1,2]. According to the scientific literature and as technology advances, there is growing interest in monitoring training loads in athletes to control their adaptation [3]. In team sports, monitoring training load is crucial for optimizing performance and preventing injuries, enabling us to anticipate the emergence of overtraining [4,5]. Thus, biomarkers that provide us information regarding changes in athlete's fatigue are highly appreciated.

Heart rate variability (HRV) is a noninvasive index that evaluates the balance between sympathetic and parasympathetic activity due to the study of successive heartbeats variation over an interval of time [6]. A reduced HRV, induced by a predomination of sympathetic activity, has been related to a reduced regulatory capacity to adapt to different challenges such as exercise or cognitive stressors [7]. Furthermore, a decrease in HRV has been considered a marker of fatigue, poor cardiovascular adaptation to effort, and overtraining [8,9], and, consequently, it has also been correlated with low sports performance [10].

Technological advances have made it possible to improve the fatigue management of athletes through the analysis of HRV [3,9]. HRV has been used in team sports as a sport-specific indicator based on data extracted from training and competitions [11]. In this regard, the study of HRV in team sports is crucial for optimizing performance and preventing injuries, fatigue, or overtraining [12]. In soccer, previous studies have used the HRV to control and manage training load during seasons [13–18]. However, in female soccer players, studies which analyze HRV are limited. In this regard, previous studies used the HRV to control training load [19], fatigue [20], or precompetitive anxiety [21].

Fatigue induced by sport is a usual situation within training and competition, but if it is not controlled, it can lead to negative alterations [22]. It is a process that has an effect on some variables of physical performance (technique or precision), and that must be taken into account in football training and recovery [23–25]. In this regard, a previous study showed that depending on the quality of the teams against which one competes, the distance and the intensity are different, which influences the players' fatigue [26]. Furthermore, HRV is also sensitive to cognitive processes [27,28], such as emotions [29]. In this regard, previous studies have reported that athletes experienced mood and wellbeing changes after a loss [30,31]. Specifically, athletes reported higher depression, anxiety, social dysfunction, and anger after a loss, compared to a win, while lower levels of vigor were observed after a loss, compared to after a win [31]. Furthermore, previous studies have found that soccer players who lost a match significantly performed higher distance sprinting and high-speed running than those players who won the match [32]. These findings, together, could suggest that the results of a competition might have a significant impact on the players' HRV.

Nevertheless, to the best of our knowledge, no previous study has investigated the impact of the results of a soccer competition on the HRV of female soccer players. Given the preceding, the purpose of this study was to investigate if the results of competition could impact the HRV of female soccer players. Thus, a follow-up of soccer players during two weeks of the league, using an HRV recorded two days before the competition (baseline) and one recorded after the competition (post-competition), was conducted. We hypothesized that HRV would be decreased after both matches (a lost and a won match). Nevertheless, significant differences are found in a lost soccer match when compared with a won match.

## 2. Materials and Methods

### 2.1. Participants

A total of 14 players were assessed for eligibility. However, 13 female players (age = 23.76 (5.32),  $n = 13$ ) from the Spanish Second National Division soccer league team participated in this cross-sectional study (see Table 1). One participant could not complete the procedures since she was injured, so she was excluded from the statistical analyses. None of the participants manifested sickness in the match's week, and no intercurrent was registered.

**Table 1.** Descriptive data of participants.

Variable	Mean (SD)
Age (years)	23.75 (5.32)
Competition experience (years)	8.85 (2.85)
Height (cm)	163.61 (5.54)
Weight (kg)	58.30 (7.54)
Body Mass Index (kg/m <sup>2</sup> )	21.78 (2.63)

Participants who complete the procedures had three sessions per week of 1 h and a half, and they had been participating in football competitions an average of 8.85 (2.85) years. All participants agreed and gave written consent to participate in the study. In addition, procedures were approved by the University of Extremadura research ethics committee (approval number: 180/2019).

### 2.2. Procedure

Standardized procedures and recommendations for assessing and reporting HRV results were followed [33]. Due to coach requirements, in terms of time limitation, we decided to conduct a short-term record (5 min) for each HRV (baseline and post-competition). Nevertheless, five minutes of HRV is considered as the gold standard for short-term mea-

surements [34], and it has shown excellent reliability for relevant variables such as RMSSD (ICC = 0.97 (0.81–0.99)) [35].

The participants' HRV was evaluated before (baseline) and after (post-competition) two matches, in the local dressing room with controlled temperature and humidity (22.3 (1.0) °C; 46.4 (2.8)%) at rest in a sitting position. In order to avoid distractions or interactions between players, at the time of HRV assessment, the room was calmed, all the players were at their places, and they were encouraged to remain silent (without talking).

The HRV measurements were on the same day of the week and at the same time for the two matches and training sessions. Players were familiarized with the procedures and environment. All participants underwent the same training, as well as the same precompetitive routines during the two selected microcycles. Moreover, participants did not take any drug, drink, or other substance that could affect the nervous system 24 h before undergoing the protocol.

The same following procedures were carried out in each match (Figure 1):

- (1) Baseline: HRV data was recorded two days before the match (in order to avoid pre-competition anxiety response). All participants were evaluated in the same training session before the warm-up to obtain the baseline data of the players. This data was used to normalize the data obtained in the post-competition measure.
- (2) Post-competition: HRV data was recorded immediately after the match in a 5-minute register.

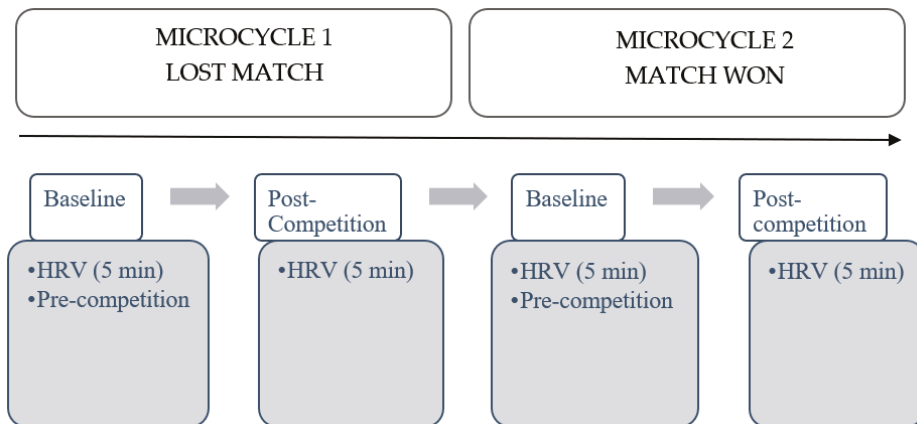


Figure 1. Study procedure timeline. HRV: Heart rate variability.

One match was lost, and the other was won, so this allowed us to evaluate if the psychophysiological impact is different between a lost or a won match.

### 2.3. Instruments and Outcomes

The HRV data was assessed using the Polar RS800CX (Polar Electro Ltd., Kempele, Finland) heart rate monitor [36] during 5 min at a sampling frequency of 1000 Hz without controlling breathing. HRV data was analyzed using the software Kubios HRV software (v. 3.3; Kubios Oy, Kuopio, Finland) [37]. A middle filter was applied to correct possible artefacts, identifying those R-to-R waves intervals shorter/longer than 0.25 s, compared to the average of the previous beats. Correction replaces the identified artefacts with cubic spline interpolation.

Time, frequency, and nonlinear domains were analyzed: (1) time domain, such as 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); (2) frequency domain, including low-frequency (LF, 0.04–0.15 Hz) and high-frequency

(HF, 0.15–0.4 Hz) ratio (LF/HF) and total power; and (3) nonlinear 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). Moreover, the stress index, the parasympathetic nervous system index (PNS index), and the sympathetic nervous system index (SNS index) were calculated. The stress index is the square root of the Baevsky's stress. The PNS index was calculated based on mean RR (ms), RMSSD (ms), and SD1 (%). The SNS index was calculated based on mean HR (bpm), Baevsky's stress index, and SD2 (%).

#### 2.4. Statistical Analysis

The IBM SPSS (Statistical Package for Social Sciences, version 25; IBM, Armonk, NY, USA) statistical package was used to analyze the data.

The Shapiro–Wilk test was conducted to explore the distribution of the HRV data since the sample size was under 50 [38]. Taking into account both the results of this test (see Supplementary Table S1) and the small sample size, nonparametric analyses were conducted. Thus, Wilcoxon signed-rank tests were conducted to explore differences between baseline and post-competitive HRV measures in both the loss and the win matches.

Subsequently, HRV data was normalized for each match (calculating the difference between match and baseline data for each HRV variable). Once HRV variables were normalized, Wilcoxon signed-rank tests were performed to explore the differences between the loss and the win matches. The effect sizes ( $r$ ) were calculated. It is classified as follows: 0.5 is a large effect, 0.3 is a medium effect, and 0.1 is a small effect [39,40].

### 3. Results

Table 2 shows the values of the HRV at baseline and after losing a soccer match. Significant differences between baseline and post-competition HRV values were found. In this regard, HRV significantly decreased all the studied variables ( $p$ -value < 0.05).

**Table 2.** HRV at baseline and after losing a soccer match for female soccer athletes.

Variable	Baseline Mean (SD)	Post-Competition Mean (SD)	$p$ -Value	Effect Size
PNS Index	0.40 (1.69)	−2.42 (0.48)	0.002 *	0.843
SNS Index	0.55 (2.98)	4.45 (1.84)	0.013 *	0.688
Stress Index	9.63 (8.31)	19.93 (8.40)	0.016 *	0.669
Mean HR	72.95 (20.06)	104.93 (10.94)	0.002 *	0.862
RR	860.26 (154.03)	578.06 (65.35)	0.002 *	0.862
pNN50	41.51 (20.72)	3.32 (4.62)	0.002 *	0.843
RMSSD	66.84 (34.23)	16.72 (9.51)	0.002 *	0.843
HF	50.17 (16.76)	13.06 (9.29)	0.002 *	0.843
LF	49.74 (16.80)	86.90 (9.32)	0.002 *	0.843
LF/HF	1.36 (1.37)	11.43 (9.24)	0.002 *	0.843
Total power	4157.06 (3956.21)	682.13 (711.64)	0.002 *	0.862
SD1	47.34 (24.25)	11.83 (6.73)	0.002 *	0.843
SD2	73.07 (33.71)	34.45 (16.47)	0.002 *	0.862

\*  $p$ -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) (ms<sup>2</sup>)/high-frequency (HF) (ms<sup>2</sup>) ratio; total power: the sum of all the spectra; PNS index: parasympathetic nervous system index, SNS index: sympathetic nervous system index and stress index; 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 comparison between baseline and post-competition HRV values after winning a soccer match. Significant differences were found in PNS index, SNS index, stress index, mean HR, RR, Pnn50, RMSSD, and SD1 ( $p$ -value < 0.05).

**Table 3.** HRV at baseline and after winning a soccer match for female soccer athletes.

Variable	Baseline Mean (SD)	Post-Competition Mean (SD)	p-Value	Effect Size
PNS Index	−0.64 (1.30)	−1.83 (0.97)	0.019 *	0.652
SNS Index	1.11 (1.60)	2.79 (1.90)	0.015 *	0.674
Stress Index	10.73 (4.23)	14.49 (6.03)	0.041 *	0.565
Mean HR	79.52 (14.63)	94.10 (14.59)	0.015 *	0.566
RR	776.95 (134.80)	653.20 (110.65)	0.023 *	0.631
pNN50	21.36 (20.10)	7.67 (10.73)	0.023 *	0.630
RMSSD	42.97 (24.91)	24.21 (15.53)	0.028 *	0.609
HF	37.45 (19.84)	16.27 (7.60)	0.006 *	0.762
LF	62.38 (19.87)	83.71 (7.58)	0.006 *	0.762
LF/HF	2.91 (3.41)	6.65 (4.02)	0.028 *	0.609
Total power	1956.09 (1698.56)	1668.89 (1608.24)	0.272	0.304
SD1	30.43 (17.65)	17.14 (11.00)	0.028 *	0.609
SD2	56.28 (23.42)	49.25 (22.56)	0.182	0.370

\*  $p$ -value < 0.005; 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) (ms<sup>2</sup>)/high-frequency (HF) (ms<sup>2</sup>) ratio; total power: the sum of all the spectra; PNS index: parasympathetic nervous system index, SNS index: sympathetic nervous system index and stress index; 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.

Comparisons between normalized HRV data in both the loss and the win matches are shown in Table 4. After losing a match, participants showed significant decrease in the PNS index, Pnn50, RMSSD, HF, total power, SD1, and SD2, and an increased SNS index and stress index ( $p$ -value < 0.05).

**Table 4.** Impact on heart rate variability of a loss and a win soccer match.

Variable	Loss Match	Win Match	p-Value	Effect Size
<b>Heart Rate Variability</b>				
PNS Index	−2.83 (1.64)	−1.06 (1.31)	0.019 *	0.549
SNS Index	3.89 (3.20)	1.47 (1.82)	0.028 *	0.610
Stress Index	10.30 (11.59)	2.64 (6.15)	0.019 *	0.649
mean HR	31.97 (17.60)	7.34 (27.14)	0.023 *	0.630
RR	−282.19 (137.09)	−173.99 (2625.27)	0.116	0.436
pNN50	−38.18 (22.40)	−14.29 (20.90)	0.023 *	0.630
RMSSD	−50.12 (34.73)	−20.62 (26.07)	0.033 *	0.591
HF	−37.11 (22.28)	−22.43 (22.31)	0.152	0.397
LF	37.16 (22.32)	14.88 (25.55)	0.055	0.533
LF/HF	10.07 (9.78)	3.22 (5.20)	0.055	0.533
Total power	−3474.93 (3524.27)	−415.57 (1706.29)	0.001 *	0.882
SD1	−35.50 (24.60)	−14.60 (18.47)	0.033 *	0.591
SD2	−38.62 (24.88)	−10.81 (27.12)	0.016 *	0.669

\*  $p$ -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) (ms<sup>2</sup>)/high-frequency (HF) (ms<sup>2</sup>) ratio; total power: the sum of all the spectra; PNS index: parasympathetic nervous system index, SNS index: sympathetic nervous system index and stress index; 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

The present article aimed to assess the effects of a lost match and a won match on the post-competitive HRV in female soccer athletes. We hypothesized that HRV would be decreased after a lost soccer match compared to a won match. Results showed that HRV significantly decreased after both a lost and a won match. In this regard, comparing the impact of the two matches (a won and a lost match), a lost match induced a significant

decrease in HRV variables (RR, pNN50, RMSSD, total power, SD1, and SD2) compared with the post-competitive HRV values obtained after the match won.

Among the tools studied to assess fatigue, HRV has emerged as a helpful tool that provides an indirect evaluation of the balance between sympathetic and parasympathetic nervous systems [34]. Thus, HRV monitoring has been used to prevent overtraining or to manage fatigue in sports such as soccer [13,14] or basketball [41]. Our results showed that a lost match induced a decrease in HRV variables such as RR, pNN50, RMSSD, total power, SD1, and SD2. In this regard, previous studies have highlighted the importance of RMSSD in the identification of fatigue [9,18]. In the same line, Proietti, di Fronso, Pereira, Bortoli, Robazza, Nakamura and Bertollo [18] showed that RMSSD is a useful HRV variable to control the training effects in professional soccer players. Therefore, these results suggest that this variable would be quite interesting in managing fatigue in female soccer players.

The physical and mental impact of losing a soccer match could explain the results obtained. Regarding the impact of physical load in the HRV, previous studies have found that increasing physical activity intensity can significantly impact the HRV [42]. A reduced HRV after a stressor itself (a soccer match) had ceased can be due to homeostatic processes. In this regard, gluconeogenesis would presumably take place, which allows muscles and the liver to refill their energy substrates. This state would require an extra cardiac output, so a higher HR, mediated by an increase in the sympathetic modulation [43,44], can be expected. In addition, the sympathetic modulation can be significantly impacted by proinflammatory cytokines [44] induced by exercises of high duration and intensity [45]. In this line, a previous study showed that soccer players who lose a match significantly performed higher distance sprinting and high-speed running than those who won the match [32]. This could explain that players showed reduced HRV after losing a match.

Regarding the impact of mental processes on HRV, previous studies have reported that athletes experienced mood and wellbeing changes after a match loss [30,31]. Athletes reported higher depression, anxiety, social dysfunction, and anger after a loss than a win, while lower levels of vigor were observed after a loss, compared to after a win [31]. Taking into account the impact of anxiety [46] or depression [47] on the HRV, as well as the role of emotions on HRV [29], decreases in HRV variables could also be justified by this reason. Therefore, the results of a competition could be considered a relevant variable to consider when programming training load. However, HRV can be considered as an index of overall fatigue, where physical and mental states can be affected. Thus, our results should be taken with caution, since the evaluation only assessed before and after two matches, and the total volume of completed load was not taken into account. Therefore, it would be necessary to independently evaluate the total volume of completed load due to the significant impact of physical load on the HRV [48]. Future studies should independently explore the role of mental and physical components in reducing HRV after a lost match.

Results indicate that five-minute pre-and post-game HRV measurements appear to be a useful way of monitoring the state of sympathetic and parasympathetic balance in female soccer players. This is relevant, since the analysis of this monitoring would be helpful to prevent fatigue by managing training loads before matches. In this regard, previous studies showed that variables such as RMSSD or the stress index are biomarkers of internal load, and therefore are sensitive enough to detect fatigue [49,50]. This is extremely useful since the player cannot voluntarily alter HRV results, unlike subjective scales such as wellness or stress questionnaires [51]. However, different protocols of HRV monitoring during sports seasons can be found in the scientific literature [13–18]. In this regard, Ravé and Fortrat [13] conducted the HRV analysis while players performed a 10 min phase in the supine position followed by a 7 min standing phase during a 5-week training period. Thorpe, Strudwick, Buchheit, Atkinson, Drust and Gregson [15] recorded the HRV during 5 min seated after 5 min cycling/5 min recovery at 130 W (85 rpm). Boullosa, Abreu, Nakamura, Muñoz, Domínguez and Leicht [14] assessed the weekly HRV, from the mean of four-daily, continuous 3 h nighttime recordings. However, there are similar protocols, but not identical to ours (5 min at rest). In this regard, Vilamitjana, Lentini, Pérez-Júnior

and Verde [16] measured the HRV during 5 min immediately after awakening on the match day. Botek, Krejčí, McKune and Klimešová [17] recorded 300 artefact-free subsequent RR intervals at rest. In the same line, Proietti, di Fronso, Pereira, Bortoli, Robazza, Nakamura and Bertollo [18] recorded 10 min seated. However, any of the protocols can be measured before and after the match. Taking into account the variability in the procedures, future studies should explore the feasibility of these protocols, trying to standardize them. This would allow the comparison with other studies.

One limitation of the present study is the relatively small sample size. Nevertheless, soccer is a sport discipline of great relevance and impact on society, and women are increasing their presence in this game. However, interfering with the dynamics of individual players on a training or match day is a drawback when it comes to obtaining a larger number of women participants. We were aware that it would be difficult for volunteers to complete the study due to, for example, injuries (a player was lost for this reason) or poor individual play. For all these reasons, we considered that, given the magnitude of the championship (second division of the Spanish female soccer league), the sample to be studied and the results obtained are of enormous interest. Furthermore, the evaluation of HRV only in relation to the results of the competition (win or lose) is very partial, and future studies should also evaluate the total volume of completed load, which affects the spectrum HRV. Moreover, it would be interesting to analyze the impact that the player's position on the field (i.e., goalkeeper, central, midfield, or forward) can have on the HRV [52–55]. It is possible that a different impact on HRV can be observed depending on the aerobic and anaerobic requirements for each position on the field. Therefore, continuous and systematic analyses throughout the season can allow for individual monitoring of each player, providing valuable information for adjusting training loads and/or detecting possible interventions on a psychological level [11,21,56]. Lastly, due to time limitation and staff's requirement, HRV assessment lasted 5 min. Although it is the gold standard for short-term measurements [34], future studies should investigate the specific reliability of this assessment in female soccer players.

## 5. Conclusions

The result of a soccer competition might significantly impact the HRV of female soccer players. In this regard, a lost match led to a decrease in HRV when compared with a match won. Therefore, researchers, coaches, or physical trainers should take into account the results of a competition when programming training load, since fatigue might be higher after a lost soccer match.

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Article

# Vision in Futsal Players: Coordination and Reaction Time

Henrique Nascimento <sup>1</sup>, Cristina Alvarez-Peregrina <sup>2</sup>, Clara Martinez-Perez <sup>1,\*</sup> and Miguel Ángel Sánchez-Tena <sup>1,3</sup>

<sup>1</sup> ISEC Lisboa, Instituto de Educação e Ciência de Lisboa, 1750-179 Lisboa, Portugal; henrique.nascimento@iseclisboa.pt (H.N.); masancheztena@ucm.es (M.Á.S.-T.)

<sup>2</sup> Department of Pharmacy, Biotechnology, Nutrition, Optics and Optometry, Faculty of Biomedical and Health Science, Universidad Europea de Madrid, 28670 Madrid, Spain; cristina.alvarez@universidadeuropea.es

<sup>3</sup> Department of Optometry and Vision, Faculty of Optics and Optometry, Universidad Complutense de Madrid, 28037 Madrid, Spain

\* Correspondence: clara.perez@iseclisboa.pt

**Abstract:** Background: Coordination and reaction time are relevant aspects of a sport's competitive performance within teams. The aim of this study was to explore if a group of futsal players, in a laboratory context, would present better results from actions where vision is prevalent compared to a control group without contact with futsal or any other sport. Methods: The digital system of the COI-SV software was used; six tests were selected, related to coordination ("Eye/hand coordination"; "Coordination and identification") and reaction time ("Anticipation Time"; "Peripheral response"; "reaction time"; "Visual memory"). Results: Of all the tests performed, only in the anticipation time test did the futsal players obtain better results than the control group. The average time of the failures was lower in relation to the control group. In the others, no differences were found between the two groups. Conclusions: The futsal players did not perform better than the control group in most of the tests carried out, except in the "anticipation time". Therefore, visual training maybe necessary to improve visual skills and sports performance.

**Keywords:** reaction time; visual coordination; visual reaction

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## 1. Introduction

The field of sports optometry is relatively new; therefore, it requires that more scientific studies be developed to prove the effectiveness of visual training in sports [1].

Futsal is a sport that was born in the 1930s (s. XX), due to the lack of spaces for the practice of soccer in Uruguay, eventually being adapted to basketball courts and other halls [2]. Futsal, in countries that have football as their national sport, is usually the second most important sport and has the largest number of practitioners. The principles of futsal are like other sports that practice in the same kind of courts like basketball, handball, and roller hockey. Therefore, when analyzing issues related to futsal, not related to vision, they may be applied to the abovementioned sports [3].

It is quite common to say that football/futsal is not taught. This is because the practice of futsal depends on a natural tendency, vocation, and skill of the practitioner. However, the development of motor abilities is essential, and learning and training techniques aim to develop the skills of the player improving its natural talents [4].

Regarding vision, there is no standard for football/futsal and the nature of the relationship between performance and vision development is unknown [2,5]. However, there are studies that conclude that visual training improves visual skills, which in turn are transmitted to sports performance [6]. As early as 1988, the visual skills associated with sports performance were analyzed. Thus, a battery of tests that approximate the visual abilities observed in sport, titled Pacific Sports Visual Performance Profile (PSVPP), was used in order to generate normative data. This study offered normative data for

non-athletes on standardized optometric tests and provided a basis for future work in vision and athletics [7].

In the study by Formenti et al. [8], they analyzed the effect of sports vision training programs for six weeks. The results showed that vision training improves cognitive skills in a non-sport-specific context (both generic and with specific motor actions), but appears to be less effective in improving sport-specific skills. This suggests that the environment where the exercises are performed plays a key role in improving perception and action on sport-specific skills, supporting the ecological approach to sports learning.

According to Welford [9], three main mechanisms govern the human sensorimotor system: perception, decision, and execution/control. The process is the perception of outside information; a decision based on what is perceived; and execution of the action according to what was decided—analysis of the result involves the sensory perception mechanism, decision-making mechanism, and execution mechanism. Later, in the study carried out by Land, it was corroborated that the visually mediated actions are based on three systems: the gaze system responsible for locating and fixing the relevant objects for the task, the motor system of the extremities to carry out the task, and the visual system to supply information to the other two. These three systems are under the supervision of a fourth system, which is the outline system, which specifies the current task and plans the general sequence of actions. These four systems have separate but interconnected cortical representations [10].

Athletic performance is highly dependent on vision [11]. There are different views on the most critical factors in athletic performance, but few disagree that vision is one of them.

The late Blanton Long Collier, a football coach who trained at the University of Kentucky between 1954 and 1961 and at the Cleveland Browns of the National Football League between 1963 and 1970, said: “eyes lead the body”. He was one of the first coaches to not only recognize that vision is critical in performance, but also that the quality of vision differs from player to player [12]. Recognizing this importance, an interdisciplinary analysis of the structure and the participants in its competitive component is necessary, so that we can do a job that improves the player’s natural abilities [13].

The objective of this study is to analyze whether futsal players have more developed visual coordination and reaction time skills compared to a control group, without contact with competitive sports.

## 2. Materials and Methods

A cross-sectional study was conducted to compare the visual skills of players from a futsal team with a control group. Inclusion criteria for both groups were aged between 18 and 30, visual acuity between 0.0 and 0.1 (LogMAR), binocular vision with vergences within normal limits and stereopsis among 40 and the 20' of arc. All participants were male, and the study took place between the months of September and November 2019 at the High-Performance Center in Sports Vision at ISEC Lisbon. Ten futsal athletes from the Portuguese senior championship the 2<sup>o</sup> division were randomly chosen. As a control group, thirty-five individuals in the same age group without contact with the sport were also chosen. Visual efficacy of all participants was evaluated with the digital system COI-Sport Vision (Centro de Optometría Internacional, Madrid, Spain), choosing the tests that measure the most relevant visual abilities for futsal [12,14]. The reliability of this software is ( $\alpha = 0.93$ ) [15]. All athletes were previously introduced to each of the tests and had a learning time, in order to ensure good repeatability of the tests and that there was no bias in the results. At the same time, during the tests all the athletes could take the tests in the time they needed, until the test ended once the tests established by the software in each test had been carried out. Peripheral vision is truly relevant in futsal practice [16]; one of the ways to train and improve it is by using eye–hand coordination exercises, which is why this system was chosen.

The order of conducting the tests was as follows:

1. Anticipation time: (reaction time): A moving object (a red ball) moves across the screen. The subject must touch the screen just as the ball passes through a rectangular area that can be in any position on the screen. The aim is that the subject could predict when the ball is going to pass through the rectangle, pressing with his/her hands or feet on the sensors to stop it. The test was performed with the dominant hand of all subjects.
2. Eye–hand coordination, coordinative reaction speed: The main purpose of this test is to train eye–hand and eye–foot coordination. The subject should press the end of the arrow with their dominant hand, which varies in orientation. The software gives information about the percentage of correct answers and how long does it take to the subject in seconds.
3. Coordination and identification: The subject should touch the red ball that moves across the screen. Along with the red ball, multiple balls of different colors appear and move on the screen as a distraction.
4. Attention and peripheral response PAT: In this test, a central circle shows a flashing light. At the ends of eight arms, other lights appear randomly. As soon as the center light and one of the peripheral lights are the same color, the subject must touch the arm light with the same color before it changes again.
5. Reaction capacity (penalty): In this test, the software measures the reaction time to the appearance of a stimulus with a certain directionality. The subject has to anticipate the movement of the ball moving across the screen and hit it through the clicker with their feet.
6. Visual memory (Tic Tac Toe): The subject has to remember the positions of various symbols that are changing position in a  $3 \times 3$  grid.

### 2.1. Statistical Analysis

Statistical analysis was conducted using the SPSS 25.0 software by (IBM Corp.: Armonk, NY, USA). Concerning the parametric distribution of the variables, the Shapiro–Wilk test ( $n < 50$ ) was used. To determine the effect size, Cohen’s  $d$  measure has been used (0.2 indicates a small effect size, 0.5 medium magnitude and 0.8 indicates a high magnitude effect). To check if there was a significant association between the continuous quantitative variables, the Pearson test was used if they had a normal distribution or the Spearman test if the distribution was not normal. The Mann–Whitney U test was used to compare the two groups. At the same time, to assess statistical significance, a cut-off point of  $p \leq 0.05$  was considered.

### 2.2. Ethical Approval

This study was approved by the ethics commission of the General Directorate for Research and Development (DGID) of Instituto Superior de Educação e Ciências (ISEC) Lisbon, Portugal. The ethical approval number is 01/27052020.

## 3. Results

The total number of participants was 45 (10 futsal players and 35 control group;  $d = 0.42$ ), hence representing small effect size, according to Cohen’s  $d$  measure. The ages were between 18 and 30 years old, the average age and standard deviation of futsal players was  $22.8 \pm 3.6$  years, and control group  $24.5 \pm 2.7$  years.

### 3.1. The Anticipation Time

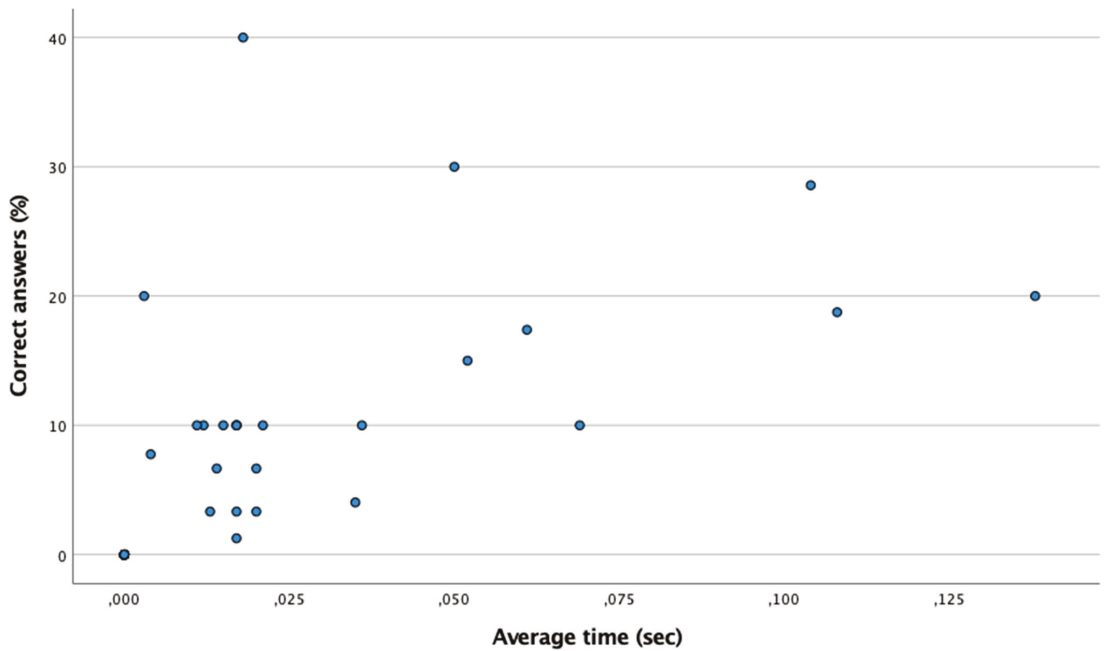
Table 1 shows the percentage and mean time of hits and misses the anticipation time. According to the Shapiro–Wilk test ( $n < 50$ ), a non-normal distribution was found ( $p < 0.05$ ). Therefore, using the Spearman test, Figure 1 shows the existence of a moderate and positive correlation between the time in reaching correct answers in the percentage of correct answers (Spearman:  $\rho = 0.877$ ;  $p \leq 0.001$ ). However, as shown in Figure 2, there is no association between mean time and percentage of mistakes (Spearman:  $\rho = 0.170$ ;

$p > 0.05$ ). Therefore, there are no differences in the mean hit time between the controls and the futsal players ( $p > 0.05$ ). However, the mean time to failures of the group of futsal players is lower than that of the control group ( $p = 0.015$ ).

**Table 1.** Results of Anticipation time test.

	Futsal Players				Control				<i>p</i> -Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	6.2	9.1	1.6	−0.3–12.7	7.7	9.7	4.0	4.2–11.1	0.620
Time to reach the correct answer (sec)	0.02	0.03	0.01	0.00–0.04	0.02	0.03	0.01	0.01–0.03	0.459
Percentage of mistakes (%)	93.8	9.1	98.3	87.3–100.3	92.5	9.4	92.5	88.9–95.7	0.600
Time to reach a mistake (sec)	0.71	1.15	0.15	−0.11–1.53	1.64	1.81	1.04	0.96–2.28	0.015

SD: standard deviation; CI: Confidence interval; sec: seconds.



**Figure 1.** Positive linear relationship between the percentage of correct answers and the mean time in the anticipation time test. The point (0,0) means that there is no correct answer.

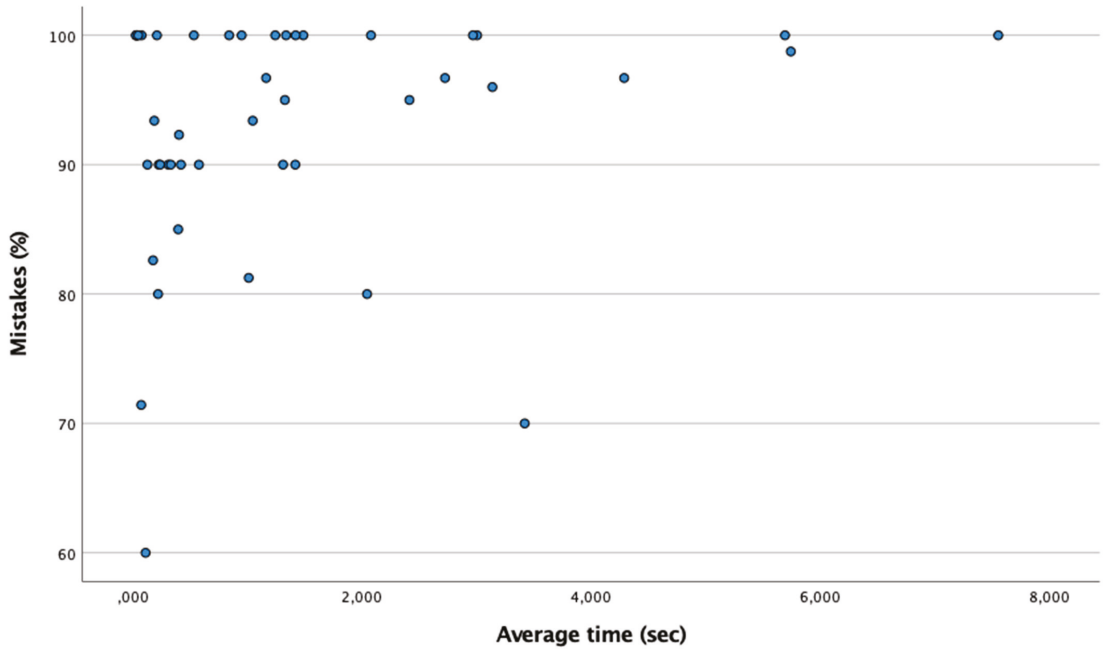


Figure 2. Absence of relationship between the percentage of failures and the mean time in the anticipation time test.

3.2. Eye-Hand Coordination Test

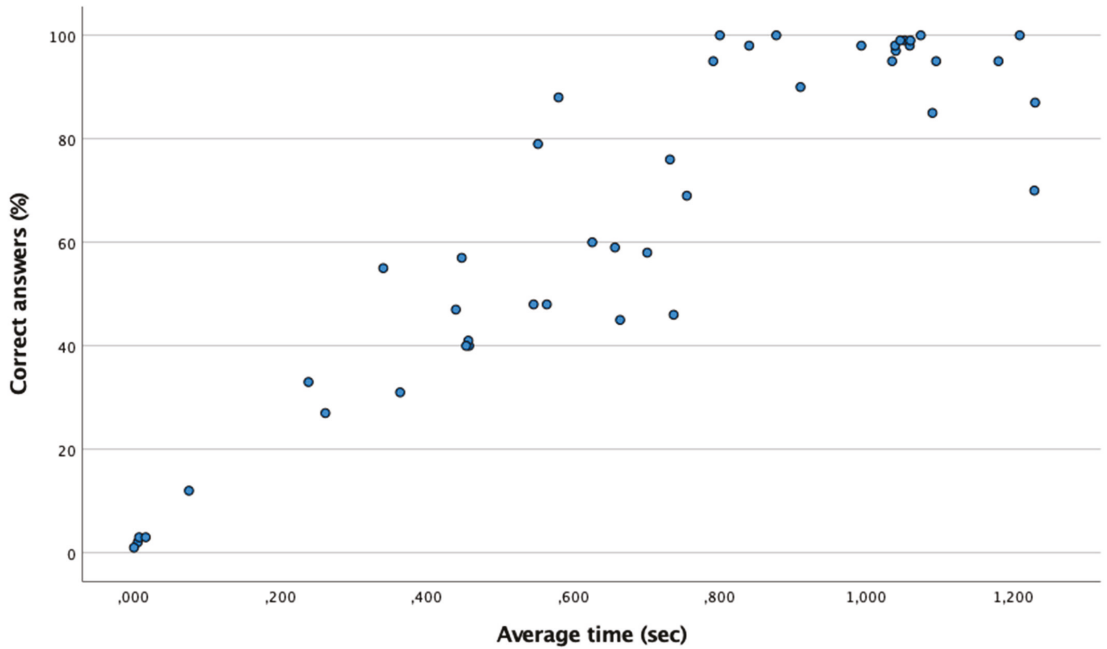
Table 2 shows the percentage of correct answers and the average time of the eye-hand coordination test. According to the Shapiro-Wilk test ( $n < 50$ ), it is a non-normal distribution ( $p < 0.05$ ), so the Spearman test has been used. Figure 3 shows the existence of a moderate and positive correlation between the time and the percentage of correct answers (Spearman:  $\rho = 0.838$ ;  $p \leq 0.001$ ). Nonetheless, the time of the futsal group is similar to that of the control group ( $p > 0.05$ ).

Table 2. Results of eye-hand coordination test.

	Futsal Players				Control				p-Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	67.0	35.2	77.0	41.8–92.2	65.6	31.8	70.0	54.7–76.5	1.000
Time to reach the correct answer (sec)	0.68	0.35	0.75	0.43–0.93	0.70	0.37	0.73	0.57–0.83	0.819

SD: standard deviation; CI: Confidence interval; sec: seconds.





**Figure 3.** Positive linear relationship between the percentage of correct answers and the average time in the eye–hand coordination test. The point (0,0) means that there is no correct answer.

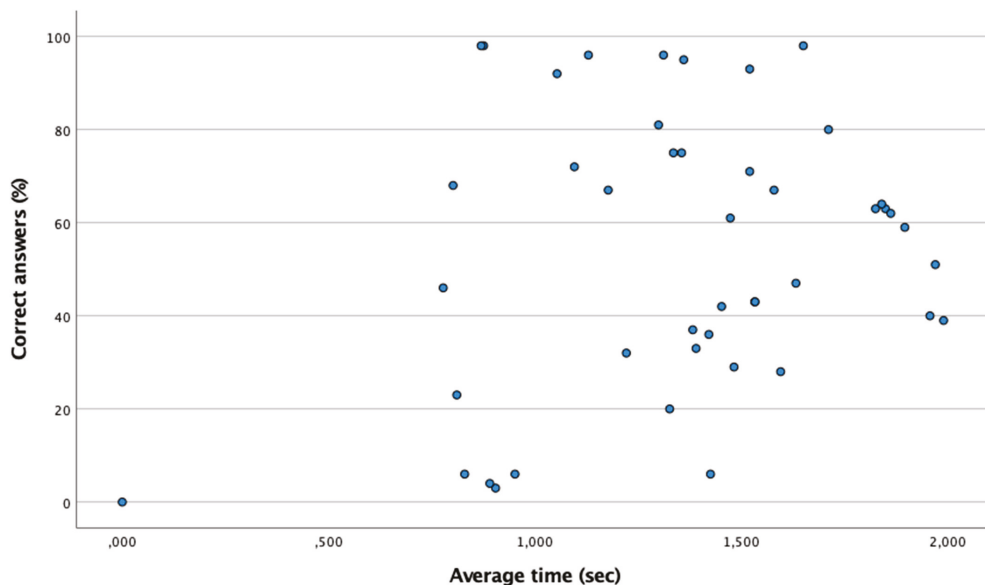
### 3.3. Coordination and Identification Test

Table 3 shows the percentage of correct answers and the average time of the coordination and identification test. According to the Shapiro–Wilk test ( $n < 50$ ), it is a non-normal distribution ( $p < 0.05$ ). Using the Spearman test, in Figure 4 no association was found between the time and the percentage of correct answers (Spearman:  $\rho = 0.086$ ;  $p > 0.05$ ). In turn, the mean time of the group of futsal players is similar to that of the control group ( $p > 0.05$ ).

**Table 3.** Results of coordination and identification test.

	Futsal Players				Control				<i>p</i> -Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	64.6	32.1	71.5	41.6–87.6	50.3	28.6	47.0	40.5–60.1	0.162
Time to reach the correct answer (sec)	1.24	0.31	1.31	1.01–1.46	1.39	0.43	1.45	1.24–1.53	0.124

SD: standard deviation; CI: Confidence interval; sec: seconds.



**Figure 4.** Absence of relationship between the percentage of correct answers and the mean time in the coordination and identification test. The point (0,0) means that there is no correct answer.

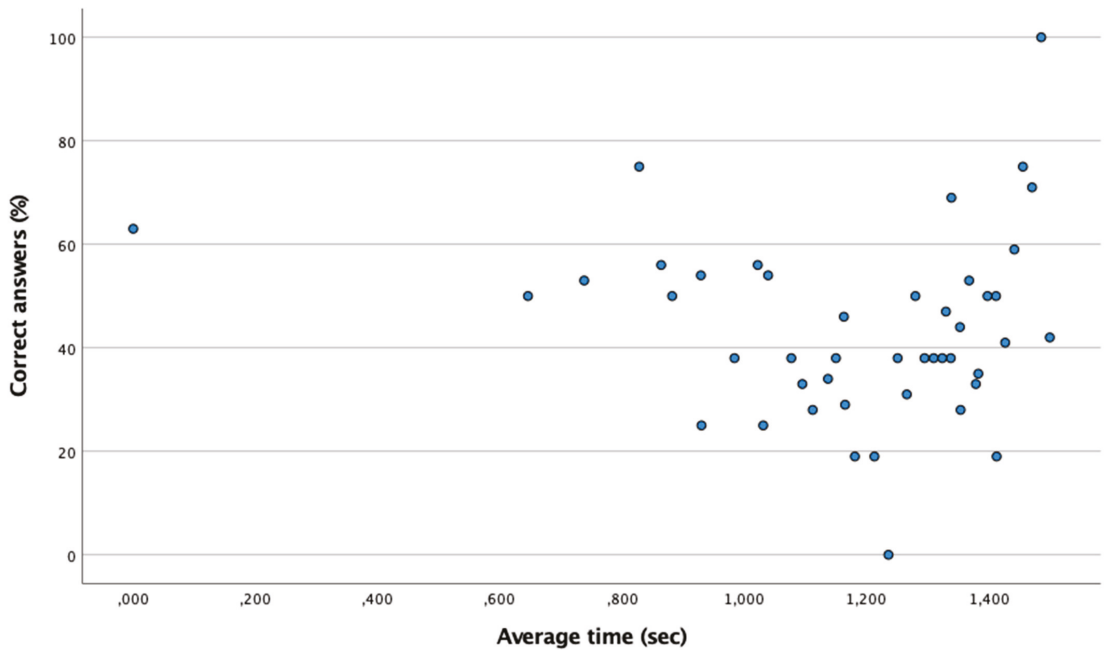
3.4. Attention and Peripheral Response (PAT Test)

Table 4 shows the percentage of correct answers, the average time and the lateral preference of the attention and Peripheral Response PAT test. According to the Shapiro–Wilk test ( $n < 50$ ), time is a variable with a non-normal distribution ( $p < 0.05$ ). The rest of the variables have normal distributions ( $p > 0.05$ ). As shown in Figure 5, there is no significant association between time of answer and the percentage of correct answers (Pearson:  $r = -0.069$ ;  $p > 0.05$  / Spearman  $\rho = 0.036$ ;  $p > 0.05$ ). Therefore, the mean time of the futsal players is similar to that of the control group ( $p > 0.05$ ).

**Table 4.** Results of Peripheral Response PAT test.

	Futsal Players				Control				p-Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	39.7	11.9	36.5	31.1–48.2	44.9	19.2	44.0	38.3–51.5	0.311
Time to reach the correct answer (sec)	1.02	0.41	1.12	0.73–1.31	1.22	0.22	1.29	1.15–1.30	0.111
Percentage of correct answer in RIGHT side (%)	28.7	20.6	29.0	13.9–43.5	46.1	25.0	50.0	37.5–54.7	0.057
Percentage of correct answer in LEFT side (%)	41.5	17.7	42.5	28.85–54.14	43.4	24.00	42.0	35.19–51.67	0.989

SD: standard deviation; CI: Confidence interval; sec: seconds.



**Figure 5.** Absence of relationship between the percentage of correct answers and the mean time in the attention and peripheral response.

When analyzing lateral preference of the visual field in the subjects, a significant association was found between the time, the percentage of correct answers, and the lateral preference left (Pearson:  $r = 0.457$ ;  $p < 0.05$  / Spearman:  $\rho = 0.364$ ;  $p < 0.05$ ). However, the lateral preference of the futsal players is similar to that of the control group ( $p > 0.05$ ).

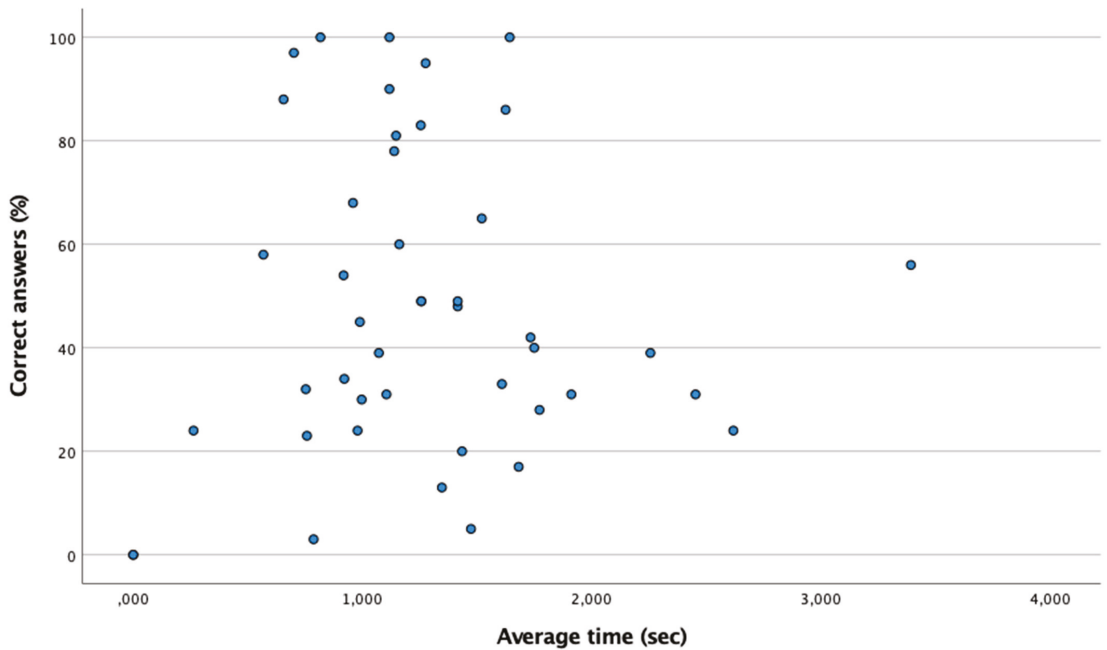
### 3.5. Reaction Capacity (Penalty)

Table 5 shows the percentage of correct answers and the average time of the reaction capacity (penalty). The Shapiro–Wilk test ( $n < 50$ ) indicates a non-normal distribution ( $p < 0.05$ ). Therefore, using the Spearman test, Figure 6 shows that there is no association between the time and the percentage of correct answers (Spearman:  $\rho = 0.002$ ;  $p > 0.05$ ). In turn, the mean time of the futsal players is similar to that of the control group ( $p > 0.05$ ).

**Table 5.** Results of the reaction capacity test.

	Futsal Players				Control				p-Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	51.4	30.2	44.5	29.8–73.0	47.1	29.6	42.0	36.9–57.2	0.697
Time to reach the correct answer (sec)	1.61	0.82	1.56	1.02–2.20	1.17	0.54	1.14	0.98–1.35	0.111

SD: standard deviation; CI: Confidence interval; sec: seconds.



**Figure 6.** Absence of relationship between the percentage of correct answers and the mean time in the reaction capacity. The point (0,0) means that there is no correct answer.

3.6. Visual Memory Test

Table 6 shows the percentage of correct answers and the average time of the visual memory test. According to the Shapiro–Wilk test ( $n < 50$ ), it is a non-normal distribution ( $p < 0.05$ ). Therefore, using the Spearman test, Figure 7 shows that there is no association between the total time and the percentage of correct answers (Spearman:  $\rho = -0.262$ ;  $p > 0.05$ ). In turn, the mean time of the futsal players is similar to that of the control group ( $p > 0.05$ ).

**Table 6.** Results of the visual memory test.

	Futsal Players				Control				p-Value
	Mean	SD	Median	CI	Mean	SD	Median	CI	
Percentage of correct answers (%)	87.9	11.7	90.8	79.5–96.2	85.7	15.9	90.9	80.3–91.1	0.717
Time to reach the correct answer (sec)	127.79	150.35	75.78	20.23–234.34	114.09	161.65	54.34	58.56–169.62	0.366

SD: standard deviation; CI: Confidence interval; sec: seconds.

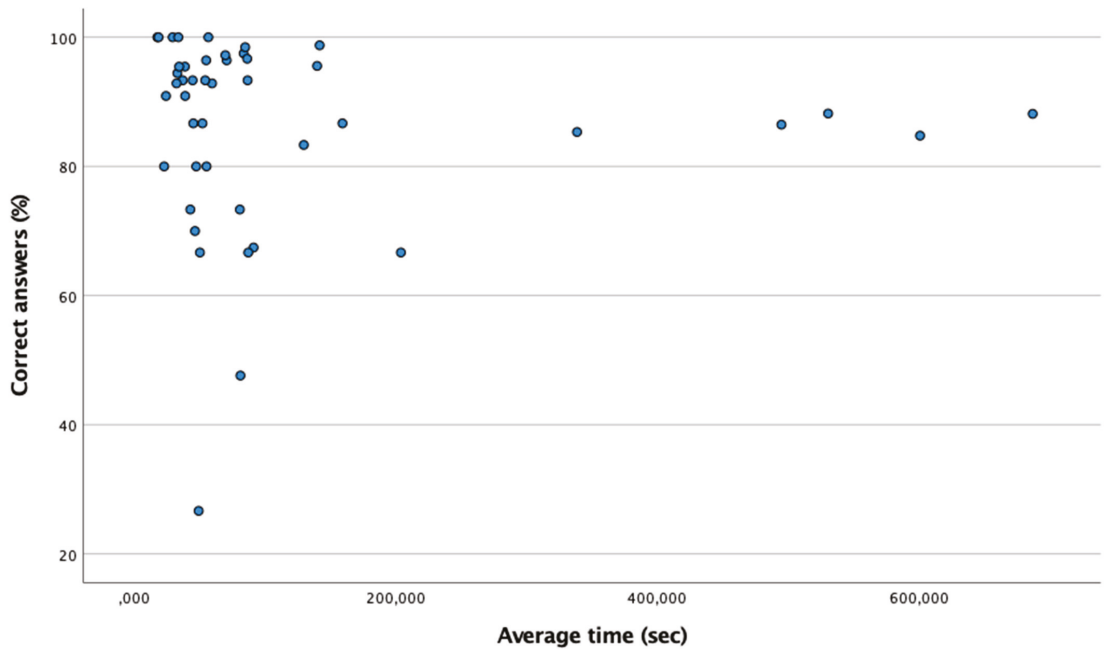


Figure 7. Absence of relationship between the percentage of correct answers and the mean time in the visual memory test.

#### 4. Discussion

It was not demonstrated in this study that by having a competitive sport activity, it is possible to develop and improve visual skills, which in turn can contribute to a better sport performance. The most important visual skills in sports are visual acuity (static and dynamic, and central and peripheral), refractive error, accommodation (amplitude; far near; relationship with convergence), eye movements, binocularity, visual perception, and visuomotor abilities [17].

In the article by Jorge et al. [14], they used the same software as in our study, COI-SV, for the analysis of dynamic visual acuity where 147 high-level futsal players were analyzed. In this article, they concluded that dynamic visual acuity varies by playing position. On the contrary, to complete what these researchers concluded, in this article it was intended to demonstrate if there was a difference between other skills such as anticipation time, eye-hand coordination, coordinative reaction speed, coordination and identification, attention and peripheral response, reaction capacity and visual memory (Tic Tac Toe). Our results agree with theirs, demonstrating the importance of vision on the field of play. In turn, both results demonstrate the validity and usefulness of sports vision software to improve performance on the playing field.

In general, eye movements, for visual information sources, precede motor actions that are supervised by the eyes. In fact, a task that requires sequential fixation requires a selection of the important in relation to the accessory [10].

However, a recent study showed that visual-motor skills play a fundamental role in sports performance suggesting that sensorimotor scans can be a useful tool in the observation of players [8]. Proprioception is a relevant aspect in this entire process, since it plays an important role in motor planning, as well as in the quick connection with the adaptation mechanisms for effecting performance changes during an execution [18]. Despite the fact that in our results, we have not found differences in the coordination between the futsal players and the control group, if we base on the studies mentioned

above, we can come to think that a training with a longer duration can improve the coordination of footballers.

There is little information about which visual skills are important for an athlete in a specific sport and about what capacity is needed to ensure consistency. As interest in sports vision is increasing, practitioners are adopting a wide diversity of methods to assess the athlete's visual skills. To improve communication between sports vision practitioners and researchers, standardized test batteries have been developed, designed to assess the different visual skills to maximized sports performance. Referred to as PSVPP (Pacific Sports Visual Performance Profile), a battery of 23 tests were designed to evaluate visual performance related to athletic competition. In addition to the classic tests of visual acuity, refractive condition, and eye health, this battery includes tests of visual performance related to the needs of tasks in different sports [19]. Thus, when comparing it with our study, we can see that since the 20th century a battery of tests was carried out to improve the vision of athletes, which has served as a guide for the development of the software that exists today.

The PSVPP includes several tests, with that on reaction time being one of the most important. Reaction time measures how fast an athlete reacts to a visual stimulus that reaches the sensory system before the motor response begins [20]. It can also be defined as the rate of preparation necessary to move, that is, the time before the action. It has an amplitude in milliseconds (ms), manifesting with different values according to the sensory system [21]. The tactile reaction time is approximately 110 (ms), the auditory is about 150 (ms) and the visual is just about 200 (ms). In the reaction time process, the stimulus via afferent reaches the primary somatosensory cortex and the posterior parietal cortex, this sensorimotor integration sends the information to area 6 of the motor cortex, where movement planning takes place. This moment of the reaction time is known as pre-motor. In the motor period, the information from area 6 is sent to area 4 of the motor cortex to generate the intention to start the movement, and the cerebellum has an important role in guiding this future action [22]. Depending on the quality of cortical excitability, the reaction time can be performed at a faster or slower speed.

Reaction time is a difficult motor skill to be trained, as it depends on the athlete's genetics. However, it can be improved by 10–20% with visual training [23]. Professional athletes have faster reaction times than beginners or untrained [23]. When the athlete is very focused on performing an activity, his reaction time is faster [24]. Other factors influence the speed of reaction time and, consequently, the speed of motor response, such as age (20 to 30 years is the peak), the type of training practiced by the athlete, their physical conditioning, the individual's level of fatigue and the cognitive level (the more intelligent, the faster reaction time) [25,26]. The reaction time is important for the practice of several sports and particularly important for futsal [27]. This agrees with the results of our study, where it has been obtained that after vision training, reaction time is better in futsal players than in the control group. Therefore, it improves the results on the playing field.

For all this, the vision must accompany the evolution of the athlete so that he can be increasingly competent in his role integrated into a team. As has been said, in practically all athletes, there is an innate part and a trainable part, which is the same as saying that it can be improved. When the athlete reaches a stage of high competition, and in exercises in which his inaptitude or his refined technique is not at stake, he is not able to rise above non-athletes, so something is not being done well in relation to his training. We verified in the results of this study, that in relation to the reaction time to different stimuli and situations outside the game environment, athletes did not clearly outperform non-athletes. On the other hand, since futsal is an activity that depends a lot on the reaction/action, it would be normal for the athletes to perform better than that seen in these results [28]. As visual training increases and develops all patterns of visual behavior and as psychologists estimate that 80% of the information we get from our external environment is through our visual pattern, we can consider this to be the most important part of the behavior pattern of the athlete [29]. Therefore, it is foreseeable that when they are part of visual training programs that improve their abilities, they can thus improve their individual and

collective sports performance. Furthermore, the development of visual skills may also benefit athletes in preventing injuries [30]. There is ample evidence that training of visual skills administered in a definitive approach and on an individual basis following particular guidelines can lead to an improved performance in various aspects of sports eventually leading to a top-level performance desired by most athletes [31].

Regarding the limitations of our study, the software used in our study has been used in another published research [14]. The decision to use this software is that COI-SV can perform both diagnostic tests and optometric treatments with a specific focus on sports vision. Without limitation of distance, it allows to work from next to the touch screen up to 15 m. In addition, all the programs have different levels of difficulty and allow us to work by quadrants of the visual field. At the same time, it is one of the few existing software that allows us to analyze various visual abilities as well as perform both visual examination and training.

## 5. Conclusions

In this study, we concluded that the futsal players did not present better visual skills than the control group, except in “eye/hand coordination”. This makes sense if we accept better coordination in an athlete as normal, in which the results were superior.

It has been proven that being a competitive player with daily sports training and with innate conditions for practicing this sport does not imply that, in terms of vision, performance is superior to a control group without contact with competitive sports.

Knowing that vision has a lot of relevance in futsal game actions and that visual skills can be developed through sports visual training programs, it is important to continue making studies that prove that improving these skills can lead to better sports performance, thus being able to motivate athletes, coaches, and managers to adopt this type of training.

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

**Data Availability Statement:** Not applicable.

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

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Article

# Effect of Virtual Reality Exercises on the Cognitive Status and Dual Motor Task Performance of the Aging Population

Hadi Nobari <sup>1,2,\*</sup>, Saeed Rezaei <sup>3</sup>, Mahmoud Sheikh <sup>3,\*</sup>, Juan Pedro Fuentes-García <sup>4</sup>  
and Jorge Pérez-Gómez <sup>2</sup>

<sup>1</sup> Department of Physical Education and Sports, University of Granada, 18010 Granada, Spain

<sup>2</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain; jorgepg100@gmail.com

<sup>3</sup> Departments of Physical Education and Sport Sciences, University of Tehran, Tehran 6619-14155, Iran; saeed\_rezayee@yahoo.com

<sup>4</sup> Didactic and Behavioral Analysis of Sports (ADICODE) Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain; jpfuent@unex.es

\* Correspondence: hadi.nobari1@gmail.com (H.N.); msheikh@ut.ac.ir (M.S.)

**Abstract:** Aging is a global phenomenon affecting numerous developed and developing countries. During this process, the functional state of the body, especially the cognitive state, declines. This research investigated the impact of virtual reality exercises on the cognitive status and dual-task performance in the elderly of Tabriz city, Iran. Forty men with a mean age of 71.5 were selected and assigned to either the experimental ( $n = 20$ ) or control groups ( $n = 20$ ). Both groups completed the Mini-Mental State Examination for cognitive status. The pre-test was performed through the Timed Up and Go test (TUG) along with a countdown of numbers. Then, the experimental group practiced virtual driving for six weeks, while the control group received no treatment. After the treatment, both groups completed the post-test. At each stage, the test was performed as a dual motor task as well. Data were analyzed using the paired  $t$ -test and the independent sample  $t$ -test to show the intra-group and inter-group differences, respectively. The results showed a significant improvement in the cognitive status and dual-task performance of the elderly men after the six-week training period, which was also significant compared to the control group. Virtual reality driving can be used to improve the cognitive status and dual task performance of elderly men.

**Keywords:** virtual driving; physical activity; behavioral status; mental state; older men

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## 1. Introduction

Aging is a global phenomenon and represents the improvement of public health. Recent studies have shown that the elderly population has increased dramatically in recent years [1]. According to the World Health Organization, the number of elderly people will exceed 780 million in 2025 [2]. However, this phenomenon is also marked by profound inequalities, such as those revealed by indicators such as life expectancy at birth. For example, while Japan has the highest life expectancy rate of 82.2 years, life expectancy in several African countries is almost half of that. Health costs increase with age [3], especially in the last two years of life, regardless of age [4]. Since people now live longer than ever before, ensuring the quality health of their remaining years is crucial.

Brucellosis in the aging population causes fundamental changes in different body systems. Morphological and biochemical changes in different parts of the brain, including signing and frontal cortex, lead to decreased cognitive features [1,5], additionally, it leads to changes in the musculoskeletal system. In this sense, different studies have shown that physical exercise positively influences different brain functions, for example, attention [6], having carried out studies to discover the effects of aerobic physical exercise on brain

neurophysiological activity during the resolution of a selective attention test [7] or working memory performance [8]. Thus, there is evidence that participation in physical activity may modify white matter integrity and the activation of regions key to cognitive processes, although additional larger hypothesis-driven studies are needed to replicate findings [9]. All of the above demonstrates that physical activity can have a positive effect on memory, executive functions, and the anti-aging resilience of the brain, by influencing certain genes associated with neuroprotective signaling [10].

The increase in spontaneous body fluctuations [11] and the high prevalence of falling down in the elderly [12] are signs of degenerative changes in the elderly. All of these changes can reduce the efficiency of dual-task performance in the elderly. In general, it can be argued that when a person grows older, the ability to simultaneously do things decreases [13]. This was demonstrated in a study of the neurophysiological differences between women with fibromyalgia (FM) and healthy controls during a dual-task, which showed women with FM had the same brain activity pattern during single-task and dual-task conditions, whereas healthy controls seemed to adapt their brain activity to the different task commitment [14]. Performing most daily activities such as dressing, brushing, and shopping, when it comes to processing external information, requires careful control and coordination in balance. Several studies have shown disorders in controlling physical force in the elderly, associated with decreased information processing when performing cognitive and motor tasks [15,16]. In recent years, a team of researchers has used a new method to evaluate balance control using the dual-task method. In this method, people are asked to perform two postural and cognitive or postural and motor tasks simultaneously [17]. The dual-task method is a useful method, as our daily activities in the standing position are dual tasks. In addition, everyday activities that require standing rarely occur individually [18]. Driving is a complex skill with the safe operation of a motor vehicle requiring good vision, high motor function, and high cognitive function. Some older drivers have difficulties when they drive in unfamiliar areas, according to an investigation carried out with five hundred and thirty-four drivers, aged  $\geq 65$  years, who completed a mail-out survey, in which 59.5% considered their abilities as poor or fair rather than good, with the most common strategies regularly used by older drivers to find their way being the use of a street directory whilst driving (61.9%) and pulling over to check a map (55.1%) [19].

Created by computer graphics and 3D screens, virtual reality (VR) offers potential solutions to the challenges posed by traditional research [20]. Recent innovations have made the technology portable, inexpensive, easily accessible, and have deleted the need for an all-inclusive driving simulator and motion tracking tools. This technology is perfectly safe to simulate and practice dangerous skills because the virtual environment is composed of graphics and users are not exposed to physical risks. Researchers have used VR as an education, practice, and rehabilitation tool [21]. Unlike the real world, the virtual world can be tailored to each individual's capacities and needs, and thus, it provides great flexibility of virtual experiences and tasks, including sequences of functional movements necessary to achieve autonomy in real-life activities. Analyzing and controlling all virtual elements of activity by the occupational therapist lead to virtual achievements that cannot be achieved in real life due to disability or environmental limitations. These achievements increase motivation, commitment, and adherence to the rehabilitation process [22]. Herrero et al. (2015) also investigated the impact of VR on the reaction time of children with autism. Twenty-seven children, 12.6 years of age on average, with high functioning autism participated in the study. The experimental group performed 15 sessions of VR training, 20 min per session. The results showed that VR exercises significantly reduced the reaction time of these children [23]. A very important feature in the technique of VR that was also considered in the present study was the presentation of activity, which is a completely activity-based approach. In this sense, the aim of this study was to analyze the effects of a 24-week exergame intervention and 24 weeks of detraining on lower-limb strength, agility, and cardiorespiratory fitness in women with FM. After the detraining period,

lower-limb strength and agility returned to their baseline level, but improvements in cardiorespiratory fitness were sustained over time, so it may have changed the lifestyle of these women, which could explain why cardiorespiratory fitness improvements remained after the detraining period [24]. This attitude is the reverse of the attitude in the pattern of behavior circuit activity, in which activities are divided into their components and each component is carried out separately until success is achieved [25]. In this study, according to the activity-based attitudes of activities, activities were presented to the elderly as a whole, and the person was busy solving in-game problems until success was achieved. In a virtual environment, all characteristics of activity such as duration, intensity, and type of feedback can be changed based on the purpose of treatment and ability of the individual [26]. People can also see their motor results and correct them if necessary [27]. With regard to cognitive status in the older ages, it can be said that aging is associated with significant changes in memory, intelligence, perception, metacognition, recalling, problem solving, and other cognitive abilities, and there is conclusive evidence of the reduction of the processing and function of memory in aging.

Liao et al., (2019) conducted a 12-week VR-based physical and cognitive training program which led to significant improvements in walking performance and dual-task performance that may be due to improved executive performance [26]. Wu et al., (2020), who reviewed 15 articles on the effects of VR exercises on the cognitive status of patients suffering from cognitive impairment, concluded that VR exercises are useful as a treatment method for people with cognitive impairments [28]. In 2014, Miller et al. conducted a systematic review on the effectiveness of VR and play systems at home in order to improve areas related to the health of the elderly. The evidence derived from 14 studies proved the effectiveness of VR and play systems by the elderly at home, which helped overcome disorders, motor limitations, and poor participation due to their bias in relation to high risks [29]. Davoodeh investigated the effect of VR games on the reaction time of elderly men, in two 15-person control and experimental groups [30]. The experimental group underwent eight weeks (three 45-min sessions a week) of VR exercise, which improved the reaction time of the elderly. VR exercises are therefore shown to be one of the safe and non-invasive methods in the literature for rehabilitation of the elderly [30]. Taking the limitations that elderly people face into consideration, one of the most attractive and non-invasive methods is the VR approach, but the study of various tools of VR and its effects on various physical and psychological factors is still in its infancy. The aim of the present study was to investigate the effect of VR exercise on the cognitive status and dual cognitive task performance in elderly men.

## 2. Materials and Methods

### 2.1. Participants

The present study is a quasi-experimental study that was conducted in a pre-test and post-test design. The statistical population consisted of all of the elderly men in the Tabriz municipality sports organization (average age of 71.5). First, a list was purposefully prepared among the elderly people of this organization, based on the inclusion criteria of the study, then, 40 eligible individuals were randomly selected, and ultimately, randomly divided into control groups with specifications ( $71.7 \pm 2.4$  yrs, confidence interval (CI) 95%, 70.6 to 72.7;  $69.3 \pm 7.9$  kg, CI 95%, 65.9 to 72.8) and experimental groups with specifications ( $71.4 \pm 2.64$  yrs, CI 95%, 70.2 to 72.6;  $68.8 \pm 7.3$  kg, CI 95%, 65.6 to 72.0). At first, all of the elderly men completed the personal information form. The selected persons, based on this form, should meet the following criteria for being included as a sample: (a) physical and mental health and the ability to perform low-intensity physical activities, (b) without a record of surgery and special illness, and (c) having a driving license and driving experience. Informed consent was given by all of the patients when collecting their clinical data. The study was approved by the Sport Science Research Institute Ethics Committee (IR.SSRI.REC.1399.851).

## 2.2. Study Design

First, a list with the names of all of the elderly members of the Tabriz municipality sports organization was prepared, and then a score or number was allocated to each of them, and finally, the required number of men was selected using a random numbers table. Among the eligible ones, 40 elderly men with an average age of 71.5 years were chosen and randomly divided into two groups: the control group or the experimental group (each group had 20 men). The control group took the pre-test and post-test with the cognitive status questionnaire and the Timed Up and Go test (TUG) at the same time as with the countdown of numbers, and performed their daily activities during the treatment period. In addition to participating in the pre-test and post-test, the experimental group participated in the six-week exercise protocol.

All participants completed the cognitive status questionnaire for the pre- and post-test, and the dual-task of sitting and standing with a countdown of numbers was used. The control group went about their daily routines, while the experimental group practiced driving for six weeks with the exercise protocol consisting of three sessions for 20 min per week.

## 2.3. Timed Up and Go Test (TUG)

The Mini-Mental State Examination [31], which is a practical method for grading cognitive patients, was used and tested five aspects of cognitive function: orientation, recording, attention, calculation, and recall. The maximum score is 30, and the scores 23 and below shows disorder (0–17 shows severe cognitive disorder, 18–23 shows mild to moderate disorder, and 24–30 shows no disorder). The mental state of all of the participants was assessed by a specialist clinician.

Dual-task assessment was performed using the Timed Up and Go test (TUG), as a quick method to determine the balance problems affecting the motor skills of the elderly person's daily life. The TUG test consists of three stages: (i) standing up from the chair, (ii) walking, (iii) turning and coming back. Participants were expected to perform this test in the least possible time. They were asked to perform the TUG test under different conditions [32]. The TUG test was performed and the TUG test was also carried out along with a cognitive task (dual cognitive task). The dual cognitive task and the TUG test were performed simultaneously with the countdown of 15 random numbers, in which the participants were evaluated using the pre-test. The experimental group participated in the six-week treatment (3 × 40-min sessions a week), and played the driving game using a computer equipped with a steering wheel and pedals. The steering wheel and pedals with specifications (Ferrari Challenge Racing Wheel) were utilized in conjunction with an ASUS laptop (G551JW) (ASUS, Taipei, Taiwan). Each participant received between 10 and 15 min extra time in the first session to provide the necessary training on how the gadget worked and to become familiar with the game.

## 2.4. Statically Analysis

To investigate the normality of data distribution and variance analysis, Shapiro–Wilk and Leven's tests were used. Due to the normality of data distribution and variance analysis, a paired *t*-test was used to investigate intra-group differences, and an independent sample *t*-test was used to investigate inter-group differences. Data were analyzed by the Statistical Package for the Social Sciences (SPSS) software, version 21 (IBM, San Diego, CA, USA), at a significance level of  $\leq 0.05$ .

## 3. Results

As Table 1 shows, there was no significant difference between the pre-test and post-test scores in the control group, but there was a statistically significant difference between the two test results in the experimental group. The results of the paired *t*-test indicated that in the experimental group, there was a significant increase in the post-test scores in cognitive

status ( $t = 4.72, p = 0.04$ ) and a significant decrease in the dual-task performance ( $t = -3.49, p = 0.02$ ).

**Table 1.** Results of paired and independent *t*-test for cognitive tests and dual-task performance.

Assessments	Groups	Pre-Intervention	Post-Intervention	Within Groups		Between Groups			
				<i>t</i>	<i>p</i>	<i>t</i>	<i>p</i>	<i>t</i>	<i>p</i>
Cognitive status	Control	22.07 ± 2.22	22.47 ± 2.10	0.67	0.162	−0.30	0.767	−2.67	0.013 §
	Experimental	22.33 ± 2.64	24.67 ± 2.41	4.72	0.04 *				
Dual-task	Control	14.40 ± 2.17	14.27 ± 2.28	−0.97	0.124	0.37	0.714	2.46	0.020 §
	Experimental	14.13 ± 1.77	12.40 ± 1.84	−3.49	0.021 *				

\* represent significant difference within groups after intervention, at a significance level of  $\leq 0.05$ . § represent significant difference between groups after intervention, at a significance level of  $\leq 0.05$ .

As Table 1 shows, the scores of cognitive status had a significant difference in the experimental group compared to the control group ( $p = 0.013$ ), in addition, there was a significant difference in the scores of the dual-task performance in the experimental group compared to the control group ( $p = 0.020$ ).

#### 4. Discussion

In this study, we investigated the effect of VR exercise on the cognitive status and dual cognitive task performance in elderly men in Tabriz. The results showed that VR exercises significantly influenced the cognitive status and dual task performance in the elderly.

There has recently been a lot of interest in using VR environments in the treatment of various psychological disorders, such as those related to FM [33]. Alternatively, creating various treatment environments with this method has motivated the patients to take a more active role in their own treatment. Since it creates a safe environment and conditions for facing different situations, patients and health care professionals have been more motivated to use VR in treatment. The decrease in sensory and motor integration is part of the aging process, which leads to elderly people facing numerous challenges while coordinating their eyes and hands, eyes and feet, and the two hands, if necessary, for many daily activities. Kinect-based VR exercise is a new technology that is increasingly used in rehabilitation for different people [34]. Findings in our study showed that the VR exercise group (i.e., the experimental group) showed a significant decrease in the cognitive status and dual-task performance compared in the intra-group (i.e., the pre-test and post-test) and inter-group in the post-test stage compared to the control group [35]. A systematic review demonstrated that video game-based interventions help promote physical health (i.e., balance, mobility, strength, physical fitness, and walking performance/gait parameters) and mental health (i.e., balance confidence, executive functions, reaction time, and processing speed) among older adults. It can also be used by researchers in this field to inform their design decisions. We have listed guidelines that can be used to frame future research in the area and enhance its quality [36].

Driving is a complex skill that involves the safe operation of a motor vehicle that requires good sight and high motor and cognitive function [37]. Driving practice as VR engages cognitive status and dual task performance ability to the maximum. Hollman et al. stated that elderly people have a slower walking speed than middle-aged and young people, and at the time of performing a dual-task, this speed decreases more. Additionally, the changes in walking throughout normal and dual task performance occur more in the elderly [38]. This issue is especially important since transport can still be an issue in later life due to physiological and cognitive challenges, with older people being generally skeptical of potential future transport, although they are welcoming of the technologies that reduce physical difficulty in mobility, and provide real-time information [39].

Another finding of our study showed VR exercises have a significant effect on the cognitive status of the elderly, and it is in line with the results of the research studies

conducted by Coyle et al. [40], Roberts [41], Chandler [42], Baker [43], and Lin [44]. The results of our study showed positive effects for VR intervention on the main dependent variables, cognitive and physical performance. These results are interesting, considering that common daily life activities are rarely carried out as a single-task but rather require the ability to perform two or more tasks simultaneously [45]. The studies show that there is a decrease in the efficacy of at least one of the tasks simultaneously performed, as a consequence of competition for attentional resources in dual-task conditions [46], resources that decrease when age is advanced [47]. The effect of VR interventions in the cognition group is consistent with the results of a systematic review by Coyle et al. [40]. Coyle et al. showed that VR improved cognitive function intervention of participants with cognitive impairment, on average. A recent VR study reported that executive functions were improved after VR intervention.

Barry investigated the effects of sports games using Xbox Kinect versus traditional exercises without virtual stimuli in gyms on postural control, technology acceptance, psychological factors, and exercise intensity in healthy adults [48]. The study, conducted on 50 healthy active adults, with an average age of 33.8 years, showed that after four weeks of exercise heart rate was equal in both groups, the Borg pressure perception scale was significantly lower in the Kinect group, and that overall, the results showed that postural control, technology acceptance, and psychological factors were improved using Kinect. Improvement of cognitive status and dual-task performance in the VR group can be attributed to the positive ability of this method to integrate the positive benefits of therapeutic techniques of repeated exercises of motion observation, motion imagination, and motor imitation, which causes the plasticity of the nervous system through mirror neurons [48].

Structural interference occurs when the physical or neural structures of the source decrease, which often occur in the implementation of two simultaneous motor tasks. In the interpretation of the present study, by performing two simultaneous motor tasks, the different senses involved in the implementation of tasks (e.g., proprioception and vision) have more practice, and also during the implementation of these tasks by moving the hands, it is likely that the change of location of the center of gravity has occurred, which has somehow led the person to use a more effective strategy to maintain balance, leading to greater balance [5]. In addition, with the continuous exercise of dual motor task ability, structural interference between the two tasks can also be reduced. Overall, it can be argued that dual-task training leads to the simultaneous involvement of postural control processes and balance with cognitive processes. It is interesting here to consider the task prioritization competition model, where older adults prioritize balance and postural stability over cognitive performance in a dual-task with dictions [49].

One of the limitations of this study was the small statistical sample. Another suggestion is to investigate this method on different genders and compare them in terms of the effect of the cognitive status and dual task performance. We hope this can pave the way for the acceptance and wide use of this method by healthcare professions in the future.

## 5. Conclusions

The results of this quasi-experimental study showed the positive benefits of the VR intervention method in improving the cognitive status and dual-task performance in the elderly population, and as a result, this treatment method can be used to improve some of the challenges our aging population is facing around the world. Given the interesting results of the present study and the effectiveness of VR exercises for the measured variables, we strongly recommend researchers conduct this study design in the future with a larger statistical population as well as between men and women separately.

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Article

# Effect of Physical Guidance on Learning a Tracking Task in Children with Cerebral Palsy

Hadi Nobari <sup>1,2,\*</sup>, Elham Azimzadeh <sup>3,\*</sup>, Hamidollah Hassanlouei <sup>3</sup>, Georgian Badicu <sup>4</sup>, Jorge Pérez-Gómez <sup>2</sup> and Luca Paolo Ardigo <sup>5</sup>

<sup>1</sup> Department of Physical Education and Sports, University of Granada, 18010 Granada, Spain

<sup>2</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain; jorgepg100@gmail.com

<sup>3</sup> Department of Behavioural, Cognitive and Technology Sciences in Sport, Faculty of Sport Sciences and Health, Shahid Beheshti University, Tehran 198396-3113, Iran; Hamidhasanlooie@gmail.com

<sup>4</sup> Department of Physical Education and Special Motricity, Transilvania University of Brasov, 500068 Brasov, Romania; georgian.badicu@unitbv.ro

<sup>5</sup> Department of Neurosciences, Biomedicine and Movement Sciences, School of Exercise and Sport Science, University of Verona, 37131 Verona, Italy; luca.ardigo@univr.it

\* Correspondence: hadi.nobari1@gmail.com (H.N.); elhamazimzadeh@gmail.com (E.A.)

**Abstract:** The purpose of this study was to investigate the effect of physical guidance (PG) frequency on learning a tracking task in children with hemiplegic spastic cerebral palsy (CP). For this purpose, 25 children, aged 7–15 years with CP affecting the left side of the body, who were classified in levels II–III of Manual Abilities Classification System (MACS) and levels III–IV of Gross Motor Function Classification System (GMFCS), were recruited from 10 clinical centers. A pre-test including two blocks of 12 trials of the tracking task without any PG was performed by all participants, after that they were assigned into five homogenous groups (with 100%, 75%, 50%, 25%, and 0% of PG) through blocked randomization according to their age. All participants involved in an intervention consisted of eight sessions (four blocks of 12 trials in each session) practicing a tracking task. The 0% PG group received no PG, the 25% PG group received PG for three trials, the 50% PG group received PG for six trials, the 75% PG group received PG for nine trials, and the 100% PG group received PG for all twelve trials. PG consisted of placing the experimenter's hand around the child's less-involved hand guiding to stay on the track and complete the task. Learning was inferred by acquisition and delayed retention tests. The results showed that the higher frequency of PG led to more accurate performance during practice phase. However, the group that received 75% PG had significantly better performance compared to the other groups in the retention phase. It is concluded that optimum level of PG, about 75% of trials, can be helpful for learning a tracking task in children with spastic hemiplegic CP, supporting the challenge point framework.

**Keywords:** physical guidance; tracking task; cerebral palsy; challenge point framework; frequency

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## 1. Introduction

Children with cerebral palsy (CP) have weak physical abilities [1]. Approximately 2–2.5/1000 children have CP with affected muscle tone, limited range of movement and motor skills, often accompanied by intellectual, communication, and behavioral impairments. These limitations may have effect on performing everyday motor activities such as putting on clothes, eating, and walking; these children also engage in less physical activity compared to their typically developing peers [2,3]. Therapeutic interventions for children with CP targeting development of gross and fine motor skills, especially upper-limb functions and eye-hand coordination, improved their functional skills and encourage them to participate in social activities. One of the most important and common debilitating factors in CP is the malfunction in the upper extremities to perform motor skills and there

is a strong evidence that task-specific training may lead to improve general upper limb function among this population [3].

In the field of motor learning, typically developing children are constantly learning new motor tasks during development. However, children with CP may use information in different way for learning motor skills depending on their stages of cognitive, sensory, and motor development [4]. The sensorimotor impairment is an important handicap in this disease which significantly impacts the quality of life in this population [5].

It has been reported that the sensorimotor impairments, visual perception, and motor planning deficits may affect motor learning in this population [6]. Previous studies showed that motor ability and performance in children with CP could be improved with physical practice [2,7].

Children with CP demonstrated more error than children with typical development in terms of accuracy and consistency during the acquisition, retention, and reacquisition phases, suggesting motor execution difficulties. These children often demonstrate different motor learning strategies due to sensory, motor execution, and cognitive impairments [8–11].

It is well documented that augmented feedback enhances acquisition and learning a motor task but there is little information available to guide practitioners in the effective use of feedback schedules to enhance acquisition and retention of motor skills in children with CP. A previous study examined the effect of augmented feedback on learning a new motor skill in adults with and without unilateral brain damage and showed that adults with unilateral brain damage exhibited more error than control participants as they practiced a rapid spatially and temporally constrained task (using their less-involved upper extremity). Differences between the groups were attributed to motor control and execution, not the cognitive learning of the motor skill [8].

One of the factors influencing learning motor skills is physical guidance (PG) that is recurrently used in education and rehabilitation. In this method, the learner is provided physical assistance during practice to facilitate the acquisition of the new skill and often involves recurrent direct guidance of the learner's moving limbs (e.g., by pulling and pushing) [12]. It has been established that PG can play a key role in acquiring a motor skill [13]. However, it has been argued that guidance must be assistive rather than limiting [14]. Often, the patient with a sensorimotor disorder, such as CP, cannot recognize the movement errors with only the therapist's verbal description, but through PG could be able to understand the spatial and temporal movement pattern and identify his/her own performance errors. Furthermore, throughout this process, the child with CP could possibly attend to appropriate wrist position, target accuracy, movement speed, and/or spatial trajectory simultaneously in a tracking task which performed by hands [15].

In order to help patients learn a skill, therapists initially provide PG, but ultimately, the primary goal is to assist the individual to develop the ability to control the movement on their own. It has been reported that PG effects in the retention phase [16]. A meta-analysis of 40 studies on learning of complex motor tasks in adults showed that continuous feedback may improve motor performance during the acquisition, but the faded feedback was more effective for the retention phase of learning [17]. Therefore, it is important to establish an optimal frequency of guidance, particularly PG, which is important for motor task learning.

According to the guidance hypothesis, practice with a high relative frequency of PG would be detrimental for learning. Although the guiding properties are beneficial for motor learning when used to reduce error, but detrimental when relied upon [18,19]. A previous study that investigated the feedback frequency in children with CP showed that continuous feedback (100%) compared to reduced (50%) or no (0%) feedback improved throwing accuracy in acquisition phase. However, only the reduced feedback group showed better performance in retention phase [2]. These results supported the guidance hypothesis [19]. However, another study indicated that the children with typical development, with 100% feedback had fewer errors than the 62% feedback group during acquisition and retention. However, there was no difference between feedback subgroups of children with CP [8].

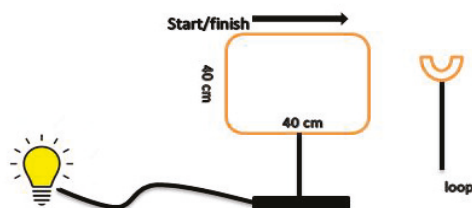
Furthermore, according to the Challenge Point Framework (CPF), there is an interaction of the information processing capabilities of the learner, the task demands, and practice conditions which influence motor learning. Practice conditions may alter the difficulty e.g., the reduced frequency of PG could be more challenging for this population to learn a new motor task [8]. Although numerous studies have considered the effect of PG, we are not aware of any research examining the effects of different frequencies of PG on learning a motor task in children with CP, and furthermore, there is little specific evidence to guide the occupational therapists to implement the more effective frequencies of PG for intervention in this population [20]. Thus, the purpose of the present study was to investigate the predictions of the challenge point hypothesis for learning a novice motor task in children with spastic hemiplegic CP, which states that too much or too little information will retard learning by manipulating the frequency of PG (i.e., 25%, 50%, 75%, or 100%).

## 2. Materials and Methods

### 2.1. Participants

Twenty-five boys with hemiplegic spastic CP, aged from 7 to 15 years, who were classified in levels II–III of Manual Abilities Classification System (MACS) and levels III–IV of Gross Motor Function Classification System (GMFCS), were recruited from ten clinical centers. Inclusion criteria for all children were (1) no severe visual deficits, (2) classifying in levels II–III of MACS and levels III–IV of GMFCS, (3) hemiplegic CP affecting the left side of the body, and (4) the less-involved (preferred) hand was the right hand. Exclusion criteria were (1) additional orthopedic or neurological disorders, (2) disability in sitting independently, and (3) pharmacological or surgical procedures in the past year. Signed informed consent was obtained from all the parents. All participants were assigned into five homogenous groups (100%, 75%, 50%, 25%, and 0% of PG) through blocked randomization according to their age.

Participants sat in front of a table while grasping a loop with their less-involved (the right) hand to perform a tracking task. The task consisted of a wire square trajectory which had 40 cm for each side, like a wire loop game. The apparatus was sensitive to touch and there was a light bulb that would turn on when the loop touched the wire. In order to complete one trial of the task, participants put the loop on the top and the left corner of the square, and after hearing the “go” signal by the trainer, they moved the loop around the wire square to reach the start place again (Figure 1).



**Figure 1.** The apparatus for training the tracking task included a wire square trajectory which had 40 cm for each side and a light bulb that would turn on when the loop touched the wire. To accomplish one trial of the task, participants had to put the loop on the top and the left corner of the square and move the loop around the wire square to reach the start place again.

### 2.2. Sample Size

We evaluated power and sample size for the design based on the statistical approach analyzed—a priori: compute required sample size; F tests; one-way, fixed effects, omnibus; number of groups = 5; power ( $1-\beta$  err prob) = 0.60—based on a prior study on the effect of physical guidance frequency and motor learning in children with hemiplegic cerebral palsy, which found a large effect size [21]. With a total of 25 subjects, there is a 62.8% (actual power)

chance of accurately rejecting the null hypothesis of no difference in variables. G-Power software was utilized for this statistical analysis (University of Dusseldorf, Dusseldorf, Germany) [22].

### 2.3. Procedure and Design

This study was an experimental design with pre-test, acquisition, and retention tests (see Figure 2). In the first session, the participants learned how to perform the task. After demonstration of the skill, a pre-test involving two blocks of 12 trials of the tracking task without any PG was performed by all participants. They were asked to make the movement as accurate as possible (i.e., fewer errors). In the acquisition phase, all participants practiced the tracking task, which consisted of eight sessions every other day (four blocks of 12 trials in each session). The participants had 10 minutes’ rest after each block. PG was implemented by placing the trainer’s hand around the child’s less-involved hand (which was the right hand) and physically guiding it to complete the trial with the least number of errors. At each session, the group with 100% PG received PG in all twelve trials; the group with 75% PG received PG in nine trials (the first three trials out of every four trials); the group with 50% PG received PG in six trials (the first trial out of every two trials); the group with 25% PG received PG in three trials (the first trial out of every four trials); and the no-PG group received no PG (0% PG) in all twelve trials. All practice sessions were conducted in the same manner. An acquisition test was performed in the last session immediately after the trials were completed, and a delayed retention test was performed four days later. The same as the pre-test, the number of total touches (errors) in 24 trials was calculated as the scores of the tests.

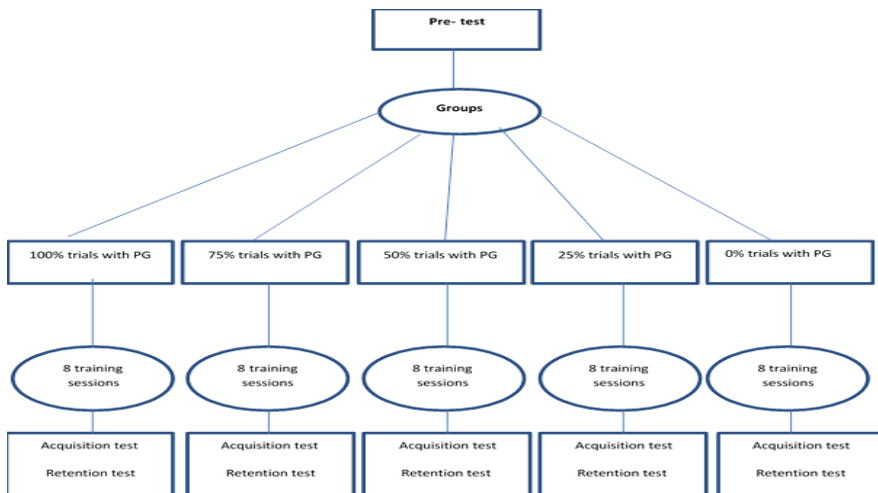


Figure 2. Research design.

### 2.4. Statistical Analysis

Shapiro–Wilk tests showed a normal distribution in all data ( $p > 0.05$ ) and Levene’s test indicated homogeneity of variances between groups ( $p > 0.05$ ) in the pre-test, acquisition, and retention phases. One-way ANOVA and Tukey’s HSD post-hoc were used to analyze acquisition and retention tests. The partial eta squared ( $\eta^2$ ) was used to evaluate the effect size of the one-way ANOVA. Data are reported as means  $\pm$  SD in the text and displayed as mean  $\pm$  SE in the figures and table. A significance level of ( $p < 0.05$ ) was used and all

the analyses were performed with IBM Statistical Package for Social Sciences (SPSS, v25.0; IBM SPSS Inc., Chicago, IL, USA). An excel file was also used to draw the figures.

### 3. Results

General characteristics of the participants and descriptive statistics of dependent variables are shown in Table 1.

**Table 1.** General characteristics of the participants by physical guidance.

Characteristics	Groups of PG										Totally (n = 5)	
	0% (n = 5)		25% (n = 5)		50% (n = 5)		75% (n = 5)		100% (n = 5)		Mean	SD
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD		
Age (years)	11.00	1.581	11.00	2.55	11.20	2.95	11.20	2.95	11.00	2.236	11.08	2.29
Height (cm)	133.2	8.468	133.80	13.9	133.8	15.9	133.0	14.5	132.6	11.69	133.2	12.04
Body mass (kg)	32.00	5.24	32.40	8.64	33.00	9.82	33.60	9.91	32.60	6.65	32.72	7.564
BMI (kg/m <sup>2</sup> )	17.91	0.7313	17.75	1.10	18.01	1.21	18.61	2.08	18.34	0.6198	18.13	1.197

BMI, body mass index; PG, physical guidance; SD, standard deviation.

Table 2 illustrates the error values (mean ± SD) obtained by each group in all tests (i.e., pre-acquisition and retention tests).

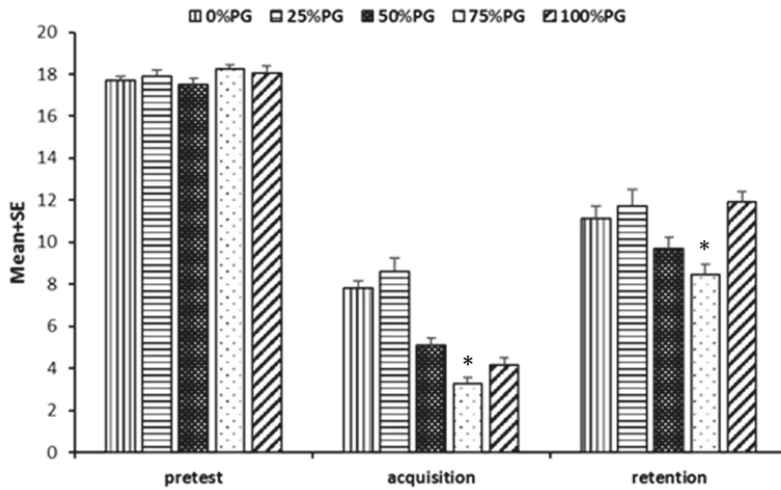
**Table 2.** Descriptive to the error values obtained in three consecutive trials.

Tests	Groups of PG										Totally (n = 5)	
	0% (n = 5)		25% (n = 5)		50% (n = 5)		75% (n = 5)		100% (n = 5)		Mean	SD
	Mean	SD	Mean	SD	Mean	Sd	Mean	SD	Mean	SD		
Pre-test	17.68	0.54	17.88	0.71	17.51	0.65	18.26	0.45	18.03	0.85	17.87	0.65
Acquisition	7.80	0.76	8.61	1.38	5.10	0.71	3.27	-0.73	4.18	0.76	5.79	2.27
Retention	11.11	1.38	11.71	1.79	9.71	1.16	8.45	1.12	11.93	1.05	10.58	1.81

PG, physical guidance; SD, standard deviation.

The results of one-way ANOVA showed that there were no significant differences between groups in the pretest ( $F_{4,20} = 1.00, p = 0.43, \eta^2 = 0.17$ ). However, in the acquisition test, there was a significant difference between groups ( $F_{4,20} = 32.73, p = 0.001, \eta^2 = 0.87$ ). Tukey’s HSD post-hoc test showed that the 75% PG group (fewer errors) had significantly better performances than 0%, 25%, and 50% PG groups (Figure 3); 100% PG was better than 0% PG and 25% PG; and the 50% PG group performed better than the 0% and 25% groups. There were no significant differences between other groups. In the retention test, one-way ANOVA was significant ( $F_{4,20} = 6.15, p = 0.002, \eta^2 = 0.55$ ). Therefore, according to Tukey’s HSD post-hoc test, the 75% PG group had significantly better performance than the 0%, 25%, and 100% PG groups, but there were no significant differences between other groups (Table 3).





**Figure 3.** Performance of the groups in the tracking task tests. \* Represents a statistically significant difference compared to the 75% PG group with the other groups (i.e., 0%, 25%, and 50% PG groups).

**Table 3.** Multiple comparisons according to Tukey post-hoc tests.

Group (I)	Comparative Group (J)	Pre-Test		Acquisition		Retention	
		MD (I-J)	p	MD (I-J)	p	MD (I-J)	p
0%	25%	-0.20	0.99	-0.82	0.57	-0.60	0.95
	50%	0.17	0.99	2.70	≤0.001 *	1.40	0.48
	75%	-0.58	0.63	4.55	≤0.001 *	2.67	0.03 *
	100%	-0.35	0.91	3.62	≤0.001 *	-0.83	0.86
25%	50%	0.37	0.90	3.52	≤0.001 *	2.00	0.16
	75%	-0.38	0.88	5.37	≤0.001 *	3.27	0.007 *
	100%	-0.15	0.99	4.34	≤0.001 *	-0.22	<0.999
50%	75%	-0.75	0.39	1.85	0.03 *	1.27	0.57
	100%	-0.52	0.72	0.92	0.51	-2.22	<0.999
75%	100%	0.23	0.98	-0.93	0.50	-3.48	0.004 *

MD—mean difference; \* represents significance at the level of  $p < 0.05$ .

#### 4. Discussion

Our study investigated the effect of different frequencies of PG in learning a tracking task in children with hemiplegic CP. The results showed that the participants who received 75% PG had significantly better performance compared to the 0%, 25%, and 50% PG groups; and the 100% PG group performed better than the 0% and 25% PG groups in the acquisition test. In the retention test, the 75% PG group had significantly fewer errors in comparison to the 0%, 25%, and 100% PG groups; however, there were no significant differences between other groups (Figure 2).

The findings were consistent with previous studies with respect to the benefits of PG, which is a simple and non-invasive method for motor learning improvement. It can also help the child to selectively attend to the model's erect posture and fluid wrist movement while reaching for the target in upper-extremity control [15]. This is more critical for children with sensorimotor dysfunction such as CP, so it may be useful for detecting the correct position of the limbs, which was the upper limb and specifically the wrist in the present study. Moreover, according to Bernstein (1967), exploration of the relationship

between a movement and the physical environment is necessary for learning motor skills, and a set of movement solutions based on the large number of joint rotations (i.e., kinematic degrees of freedom) is needed to complete the desired movement. PG likely facilitates this process [6].

Furthermore, the results of the present study confirm the guidance hypothesis and the CPF, which state that too much or too little information will retard learning a new motor skill both in children with spastic hemiplegic CP and typically developing children. Moreover, our results support the findings of [12,16,23], confirming the beneficial effect of optimal frequency of PG on learning motor skills. Furthermore, in the study of Hemayatlab, et al. [2], only the group that received reduced feedback improved accuracy in throwing in the retention phase—this was in line with our findings. One explanation for these findings is that a high level of PG (i.e., 100%) makes the individual more dependent and therefore not able to utilize his/her internal sensory information. A low frequency of guidance (i.e., 25% or 0%), on the other hand, is not helpful in identifying and developing appropriate patterns of motor skills. In fact, PG should be provided at an optimum level to decrease the child's dependency and to help them to identify appropriate patterns of skill. Continuous feedback limits the opportunity for exploration the instructions and information on each trial and may induce dependency in learner. In other words, reduced feedback or information results in a self-regulatory strategy in learning motor skills [8]. Therefore, an increased efficiency in encoding process and improved performance in the retention phase is expected [24].

Our results indicated that the optimal frequency of PG played a critical role in learning a tracking task in children with CP and likely this population may benefit from an optimal level of PG to get the appropriate amount of information as typically developing children confirming the CPF [25]. Therefore, an optimum level of PG for the children with CP would provide two important sets of resources for learning motor skill simultaneously. In this way, these children are able to identify appropriate patterns of motor skill using extrinsic information they receive through PG. Furthermore, implementing an appropriate level of PG may improve learning by activating the intrinsic feedback mechanisms [24].

Moreover, interventions involving task specificity in children with CP often relates to training of upper limb or fine motor activities. Task-specific training (TST) involves principles of motor learning with components including context, practice, and frequency of feedback. TST should involve varied components depending on the requirements of the skill, the environment, and the function of the child [3].

### *Limitations*

There were some limitations in the present study that need to be considered in future research. First is the small sample size and the ability to generalize the results to all children with hemiplegic CP. In addition, this is stated in the statistical power reported in the article method with 62%, so great caution should be taken when interpreting the results of the study and making conclusions. Second is the lack of control for additional variables influencing learning a motor task in CP children such as their cognitive abilities. Third is the comparison of the intervention on the affected hand and the less-involved one due to the application of therapeutic interventions. For this reason, we strongly recommend researchers interested in the neurophysiology of motor training in CP children to practice and experiments with the injured arm in studies to increase performance in hemiplegia, because the quality of life of these patients depends strongly on the residual capacities of the affected arm.

### **5. Conclusions**

In summary, our results suggest that children with spastic hemiplegic CP had benefited from an optimal amount of PG (about 75% of trials) when learning a new skill with their less-involved hand. The results of this study may help physiotherapists to provide effective therapeutic interventions to improve motor learning in children with CP [26].

**Author Contributions:** Conceptualization, E.A., H.H., and H.N.; methodology, H.N., E.A., H.H., and J.P.-G.; software, E.A., H.H., H.N., and G.B.; formal analysis, E.A., H.H., H.N., and L.P.A.; investigation, E.A., H.H., H.N., and L.P.A.; writing—original draft preparation, E.A., H.H., H.N., and G.B.; writing—review and editing, H.N., L.P.A., J.P.-G., and G.B. All authors have read and agreed to the published version of the manuscript.

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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 Shahid Beheshti University Research and Ethics Committee (IR.SBU.REC.1397.24, date: 15 April 2018).

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

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

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

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Article

# Early Left Ventricular Diastolic Dysfunction, Reduced Baroreflex Sensitivity, and Cardiac Autonomic Imbalance in Anabolic–Androgenic Steroid Users

Evangelia Joseph Kouidi, Antonia Kaltsatou, Maria Apostolos Anifanti and Asterios Pantazis Deligiannis \*

Sports Medicine Laboratory, Department of Physical Education and Sport Sciences, Aristotle University of Thessaloniki, 57001 Thessaloniki, Greece; kouidi@phed.auth.gr (E.J.K.); akaltsat@gmail.com (A.K.); manyfant@phed.auth.gr (M.A.A.)

\* Correspondence: adeligia@phed.auth.gr; Tel.: +30-6945151398

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**Abstract:** The effects of androgen anabolic steroids (AAS) use on athletes' cardiac autonomic activity in terms of baroreflex sensitivity (BRS), and heart rate variability (HRV) have not yet been adequately studied. Furthermore, there is no information to describe the possible relationship between the structural and functional cardiac remodeling and the cardiac autonomic nervous system changes caused by AAS abuse. Thus, we aimed to study the effects of long-term AAS abuse on cardiac autonomic efficacy and cardiac adaptations in strength-trained athletes. In total, 80 strength-trained athletes (weightlifters and bodybuilders) participated in the study. Notably, 40 of them using AAS according to their state formed group A, 40 nonuser strength-trained athletes comprised group B, and 40 healthy nonathletes (group C) were used as controls. All subjects underwent a head-up tilt test using the 30 min protocol to evaluate the baroreflex sensitivity and short HRV modulation. Furthermore, all athletes undertook standard echocardiography, a cardiac tissue Doppler imaging (TDI) study, and a maximal spirometric test on a treadmill to estimate their maximum oxygen consumption (VO<sub>2</sub>max). The tilt test results showed that group A presented a significantly lower BRS and baroreflex effectiveness index than group B by 13.8% and 10.7%, respectively ( $p < 0.05$ ). Regarding short-term HRV analysis, a significant increase was observed in sympathetic activity in AAS users. Moreover, athletes of group A showed increased left ventricular (LV) mass index (LVMI) by 8.9% ( $p < 0.05$ ), compared to group B. However, no difference was found in LV ejection fraction between the groups. TDI measurements indicated that AAS users had decreased septal and lateral peak E' by 38.0% ( $p < 0.05$ ) and 32.1% ( $p < 0.05$ ), respectively, and increased E/E' by 32.0% ( $p < 0.05$ ), compared to group B. This LV diastolic function alteration was correlated with the year of AAS abuse. A significant correlation was established between BRS depression and LV diastolic impairment in AAS users. Cardiopulmonary test results showed that AAS users had significantly higher time to exhaustion by 11.0% ( $p < 0.05$ ) and VO<sub>2</sub>max by 15.1% ( $p < 0.05$ ), compared to controls. A significant correlation was found between VO<sub>2</sub>max and LVMI in AAS users. The results of the present study indicated that long-term AAS use in strength-trained athletes led to altered cardiovascular autonomic modulations, which were associated with indices of early LV diastolic dysfunction.

**Keywords:** anabolic–androgenic steroids; athletes; baroreflex sensitivity; cardiac autonomic nervous system; cardiac function

## 1. Introduction

There are several references in the literature regarding the use of anabolic–androgenic steroids (AAS) as doping substances in sports and particularly their health effects, there are several references in the literature. Most studies on their morphological and functional adverse effects on various functional systems after acute or long-term administration are based on experimental animal results [1]. For ethical, legal, and methodological reasons,

the reports are not very well documented in athletes [1,2]. A limitation of almost all similar studies regarding the cardiovascular side-effects of AAS in athletes is the fact that the results are based on cross-sectional studies with different subject populations rather than longitudinal data. The longitudinal studies using AAS in humans would give rise to ethical problems. From the few clinical studies and case reports, the most destructive effects of AAS concern the cardiovascular system. Their administration in significant and long-term doses favors the manifestation of atherosclerosis of blood vessels, direct toxic action on myocardial cells, myocardial fibrosis, destruction of the endothelium, and dysfunction of the autonomic nervous system [2–4]. Clinical manifestations of these disorders are the occurrence of cardiomyopathy, myocardial infarction, arterial hypertension, arrhythmias, and sudden cardiac death [5–7]. Especially for the effects of AAS on athletes' cardiac autonomic nervous system, studies are minimal, and the results are controversial. It is argued that their administration increases the action of the sympathetic nervous system and reduces the vagal tone [8,9]. Thus, an imbalance of the two limbs of the autonomic nervous system occurs, which is proven by the alterations of the heart rate variability (HRV) indices. In addition, based on experimental research and recent studies in AAS users, there are indications of the decreased sensitivity of the peripheral baroreflex and the sensitivity of the Bezold–Jarisch reflex control of heart rate and blood pressure [10–13]. There is still no research that studies whether cardiac autonomic dysfunction and reduced baroreflex sensitivity (BRS) after long-term AAS abuse are independent or associated with morphological and functional cardiac remodeling. The present study aimed to investigate the function of resting peripheral BRS and HRV modulation in strength-trained athletes receiving AAS and correlate the findings with the left ventricular anatomical and functional indices, as well as their aerobic capacity levels.

## 2. Materials and Methods

There was an open call in the local training centers for strength-trained athletes, such as bodybuilders and weightlifters, to participate in the study, where the aim and the method of the study were clearly stated. Healthy males aged 18 to 45 years, with at least five years' experience in weight training, were eligible participants. Exclusion criteria were smoking, alcohol use or other drugs use besides AAS, presence of any chronic disease, atrial fibrillation, and medically prescribed testosterone therapy. Information about medical history, exercise training regime, and AAS usage was obtained from all volunteers. Based on self-reported history, athletes were allocated either to current AAS users or athletes without a history of AAS use. All AAS users reported that they were using only oral and injectable AAS substances for at least the last three years. In total, 40 strength-trained male athletes using AAS for at least 3 years (group A) and 40 strength-trained athletes, nonusers (group B) participated in this cross-sectional study. Moreover, 40 age-matched healthy nonsmokers and nonathletes, who did not use any medication were served as controls (group C).

All volunteers were examined in the Laboratory of Sports Medicine of the Aristotle University of Thessaloniki, in Greece, an authorized sports cardiology center in Greece. All tests were conducted in the morning and interpreted by the same cardiologist blinded to the identity of the participants. They were asked to refrain from exercise and all dietary sources of caffeine and alcohol 24 h before their examination. The evaluation included clinical history, clinical examination, resting electrocardiogram, an echocardiographic study, a head-up tilt test using a 30 min protocol to evaluate baroreflex sensitivity and short-term heart rate variability (HRV), and a maximum cardiopulmonary exercise testing on a treadmill.

All participants gave written informed consent. The study was conducted under the Declaration of Helsinki. The Ethics Committee of the Aristotle University of Thessaloniki approved the study protocol (Approval Number EC-65321/2012, Thessaloniki, 16 July 2012).

## 2.1. Measurements

### 2.1.1. Echocardiographic Study

Transthoracic echocardiography was performed using Vivid S70 (GE Medical, Horten, Norway) with an M5S phased-array transducer. All echocardiographic images were obtained and stored by an experienced cardiologist–ultrasonographer blinded to the identity of the participants. The studies were analyzed offline by two cardiologists using the Echopac version 201 (GE, Horten, Norway).

Measurements of the left ventricle (LV) and its walls were performed in the parasternal long-axis view by M-mode approach according to the American Society of Echocardiography guidelines [14]. LV mass was estimated from parasternal views using the Devereux formula:  $0.8\{1.04[(LVEDD + IVSd + PWd)^3 - LVEDD^3]\} + 0.6$ , where LVEDD, IVSd, and PWd represent LV end-diastolic diameter, interventricular septal, and posterior wall thickness in diastole, and RWT was calculated with the formula:  $(2 \times \text{posterior wall thickness}) / (\text{LV internal diameter at end-diastole})$ . LVMI was corrected for body surface area (BSA). LV ejection fraction (LVEF) and LA volume were estimated using the biplane method of disks. LA maximal volume (LAVi) was measured at the end-systole and corrected for BSA. LV diastolic function was assessed according to the American Society of Echocardiography and the European Association of Cardiovascular Imaging guidelines [15]. Pulsed-wave (PW) Doppler was performed in the apical four-chamber view to obtain mitral inflow velocities. E-wave and A-wave peak velocities and their ratio E/A was measured.

PW tissue Doppler imaging (TDI) was performed in the apical four-chamber view to acquire mitral annular velocities at the septal and lateral wall and measure early diastolic peak E' velocity and late diastolic A' velocity in both walls. E/E' average ratio was obtained averaging the e' velocity from the septum and lateral sides of the mitral annulus and was used to estimate LV filling pressures. TR systolic jet velocity was obtained with CW Doppler from parasternal and apical four-chamber view with color flow imaging to obtain the highest Doppler velocity.

### 2.1.2. Arterial Baroreflex Sensitivity and Heart Rate Variability Assessments

Baroreflex sensitivity was assessed by the Task Force Monitor 3040i device (CNSystem, Graz, Austria). After lying in a supine position for 5 min, all participants were placed in a 60° head-up position for 30 min. During each test, the RR intervals (RRI) were assessed from a continuous electrocardiogram, while continuous arterial Blood pressure (BP) was obtained using photoplethysmography on the middle finger. Baroreflex sensitivity (BRS) was assessed by spectral analysis of systolic BP (SBP) and RRI changes and was estimated using the average regression of the baroreflex slope of the SBP/RRI relationship. Moreover, the baroreflex effectiveness index (BEI), which indicates the ramps in RRI and SBP, was estimated. The ramp count and event count were also estimated. The ramp count indicates at least 3 consecutive beats, where the SBP rose or fell, while the event count indicates the number of baroreceptor sequences, where for at least 3 consecutive beats, there is a rise or fall of SBP with a subsequent shortening or lengthening of RRI.

By the same device, power spectral analysis of the short-term heart rate variability (HRV) was obtained for assessing cardiac autonomic activity. The low frequency (0.04–0.15 Hz) spectral component of the R–R interval using normalized units (LFnu–RRI) was estimated as a marker of sympathetic activity. On the other hand, the high frequency (0.15–0.4 Hz) spectral component of the R–R interval (HFnu–RRI) was estimated as a marker of cardiac vagal activity. Finally, their ratio (LF/HF ratio) was estimated as a marker of sympathovagal balance.



### 2.1.3. Cardiopulmonary Exercise Testing

Finally, each participant underwent a maximal cardiopulmonary exercise testing on a Trackmaster treadmill (Full Vision Inc, Newton, KS, USA) using a Bruce protocol. There was a continuous electrocardiogram, while BP was measured at the end of each 3 min stage. Expiration gases were analyzed using Med Graphics Breeze Suite CPX Ultima spirometric device (Medical Graphics Corp, Saint Paul, MN, USA). Maximum oxygen consumption ( $\text{VO}_2\text{max}$ ) was defined as the highest oxygen consumption obtained in the final 30 s of the test, characterized by a plateau of oxygen uptake despite further increases in work rate (steady time). The respiratory exchange ratio was higher than 1.10 in all tests. Measurements at maximum exercise included SBP and diastolic blood pressure (DBP), heart rate (HR), pulmonary ventilation (VE), and total exercise time (ExTime).

### 2.2. Statistical Analysis

Continuous variables were expressed as mean  $\pm$  standard deviation. The Kolmogorov–Smirnov test was used to test the normality, a condition fulfilled by the data analyzed. Changes of variables within the groups were evaluated by one-way analysis of variance, with a group being the independent variable. Correlation coefficients were calculated according to Pearson analysis. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS, Chicago, IL, USA), version 20.0 software for Windows. The significance level was  $p < 0.05$ .

## 3. Results

The physical characteristics of the participants are presented in Table 1. There were no statistically significant differences in all variables among the three groups.

**Table 1.** Physical characteristics of the study population (mean  $\pm$  S.D.).

Groups	A	B	C
Age (years)	27.4 $\pm$ 8.6	26.9 $\pm$ 7.8	27.1 $\pm$ 5.6
Height (cm)	1.75 $\pm$ 0.08	1.74 $\pm$ 0.08	1.76 $\pm$ 0.07
Weight (kg)	81.5 $\pm$ 15.2	81.3 $\pm$ 10.9	80.1 $\pm$ 8.1
BMI	26.6 $\pm$ 2.6	26.2 $\pm$ 2.3	25.9 $\pm$ 2.2
Training age (years)	10.3 $\pm$ 3.3	10.1 $\pm$ 3.6	-
Duration of AAS use (years)	4.3 $\pm$ 0.5		-

A: AAS users; B: nonusers; C: controls; BMI: body mass index; AAS: androgen–anabolic steroids.

Results obtained from the cardiopulmonary exercise testing are presented in Table 2. All tests were terminated due to volitional exhaustion. The SBP at rest was found in group A to be increased by 8.8% ( $p < 0.05$ ) and 7.4% ( $p < 0.05$ ), in comparison with groups B and C, respectively. In group A, SBPmax was higher by 6.8% ( $p < 0.05$ ) and by 8.8% ( $p < 0.05$ ), compared to groups B and C, respectively. Additionally, group A had increased ExTime by 11.0% and  $\text{VO}_2\text{max}$  by 15.1% ( $p < 0.05$ ), compared to group B, and by 17.5% ( $p < 0.05$ ) and by 18.3% ( $p < 0.05$ ), compared to C, respectively.

**Table 2.** Results from the cardiopulmonary exercise testing (mean  $\pm$  S.D.).

Groups	A	B	C
HRrest (beats/min)	73.1 $\pm$ 12.1	72.5 $\pm$ 11.3	72.5 $\pm$ 9.3
HRmax (beats/min)	184.0 $\pm$ 11.3	184.7 $\pm$ 13.5	180.0 $\pm$ 11.7
SBPrest (mmHg)	127.0 $\pm$ 6.7 <sup>a,b</sup>	116.7 $\pm$ 8.0	118.2 $\pm$ 9.2
SBPmax (mmHg)	174.0 $\pm$ 13.3 <sup>a,b</sup>	162.9 $\pm$ 14.6	160.0 $\pm$ 12.3
DBPrest (mmHg)	75.7 $\pm$ 9.9	76.7 $\pm$ 9.1	77.2 $\pm$ 5.7
DBPmax (mmHg)	75.0 $\pm$ 7.8	75.0 $\pm$ 8.8	76.3 $\pm$ 6.6
ExTime	12.1 $\pm$ 1.1 <sup>a,b</sup>	10.9 $\pm$ 1.2	10.3 $\pm$ 0.9
VO <sub>2</sub> max (mL/kg/min)	46.6 $\pm$ 6.4 <sup>a,b</sup>	40.5 $\pm$ 7.1	39.4 $\pm$ 6.1
VE max (L/min)	107.7 $\pm$ 20.0	108.6 $\pm$ 19.4	109.1 $\pm$ 18.2

A: AAS users; B: nonusers; C: controls; <sup>a</sup>  $p < 0.05$  A versus B; <sup>b</sup>  $p < 0.05$  A versus C; HR: heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure; ExTime: exercise time; VO<sub>2</sub> max: maximum oxygen consumption; VEmax: maximum ventilation.

Table 3 presents the results of the baroreflex sensitivity and HRV assessments. There were no statistically significant differences in ramp count, event count and HFnu-RRI among the three groups. The BRS and BEI indices in group A were found to be decreased by 13.8% ( $p < 0.05$ ) and 10.7% ( $p < 0.05$ ), compared to B, and by 16.1% ( $p < 0.05$ ) and 6.4% ( $p < 0.05$ ), compared to C, respectively. Moreover, LFnu-RRI and LF/HF ratio were increased in AAS users by 24.2% ( $p < 0.05$ ) and 25.5% ( $p < 0.05$ ), compared to group B, and by 27.6% ( $p < 0.05$ ) and 46.8% ( $p < 0.05$ ), compared to group C, respectively.

**Table 3.** Results of the Baroreflex sensitivity and HRV assessments (mean  $\pm$  S.D.).

Groups	A	B	C
BRS (ms/mmHg)	9.4 $\pm$ 2.3 <sup>a,b</sup>	10.9 $\pm$ 1.8	11.2 $\pm$ 1.9
BEI (%)	65.7 $\pm$ 10.4 <sup>a,b</sup>	73.6 $\pm$ 9.2	70.2 $\pm$ 10.3
Ramp Count	333.6 $\pm$ 74.3	369.3 $\pm$ 74.9	355.5 $\pm$ 61.7
Event Count	172.1 $\pm$ 55.6	182.7 $\pm$ 45.5	176.5 $\pm$ 54.3
HFnu-RRI (%)	19.4 $\pm$ 4.7	20.5 $\pm$ 4.1	21.3 $\pm$ 4.3
LFnu-RRI (%)	97.5 $\pm$ 8.3 <sup>a,b</sup>	78.5 $\pm$ 8.7	76.4 $\pm$ 8.8
LF/HF ratio	6.9 $\pm$ 3.9 <sup>a,b</sup>	5.5 $\pm$ 3.6 <sup>c</sup>	4.7 $\pm$ 3.7

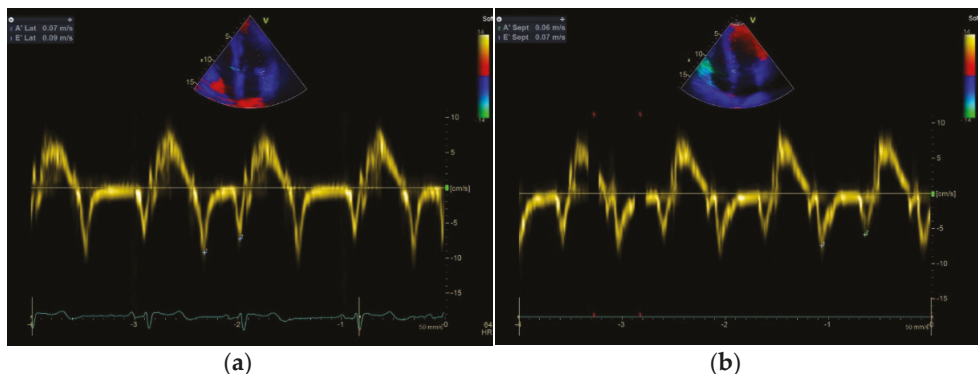
A: AAS users; B: nonusers; C: controls; <sup>a</sup>  $p < 0.05$  A versus B; <sup>b</sup>  $p < 0.05$  A versus C; <sup>c</sup>  $p < 0.05$  B versus C; BRS: baroreflex sensitivity; BEI: baroreflex effectiveness index; HFnu-RRI: high-frequency spectral component of the R-R interval using normalized units; LFnu-RRI: low-frequency spectral component of the R-R interval using normalized units; LF/HF ratio: low-frequency/high-frequency ratio.

Results obtained from the echocardiographic study are listed in Table 4. Group A demonstrated a significant increase in LVM and LVMI by 9.3% ( $p < 0.05$ ) and 8.9% ( $p < 0.05$ ), compared to group B, and by 38.4% ( $p < 0.05$ ) and by 39.1% ( $p < 0.05$ ), compared to group C, respectively. Moreover, group A had increased RWT by 16.2% ( $p < 0.05$ ), LAVi by 22.3% ( $p < 0.05$ ) and TR peak velocity by 44.4% ( $p < 0.05$ ), compared to group C. Finally, in group A septal E' and lateral E' were by 38.0% ( $p < 0.05$ ) and 32.1% ( $p < 0.05$ ) decreased, compared to group B, and by 44.1% ( $p < 0.05$ ) and 35.0% ( $p < 0.05$ ), compared to group C, respectively. On the other hand, group A demonstrated a significant increase in E/E' aver by 32.0% ( $p < 0.05$ ), compared to group B, and by 60.4% ( $p < 0.05$ ), compared to group C (Figure 1).

**Table 4.** Echocardiographic results (mean ± S.D.).

Groups	A	B	C
IVSd (mm)	12.2 ± 1.6 <sup>b</sup>	11.8 ± 1.1	10.6 ± 1.0
PWd(mm)	11.9 ± 1.4	11.5 ± 1.3	9.8 ± 1.3
LVEDD (mm)	51.8 ± 3.8	50.2 ± 4.3	49.9 ± 3.7
LVM (g)	227.0 ± 27.6 <sup>a,b</sup>	208.7 ± 28.3 <sup>c</sup>	164.0 ± 17.3
LVMi(g/m <sup>2</sup> )	115.2 ± 6.3 <sup>a,b</sup>	105.8 ± 5.8 <sup>c</sup>	82.8 ± 5.4
RWT (cm)	0.43 ± 0.05 <sup>b</sup>	0.44 ± 0.05 <sup>c</sup>	0.37 ± 0.04
EF (%)	62.7 ± 5.2	62.3 ± 5.0	62.9 ± 5.7
LAVi (ml/m <sup>2</sup> )	30.7 ± 1.8 <sup>b</sup>	28.2 ± 2.1	25.1 ± 1.9
TR peak velocity (m/s)	1.3 ± 0.2 <sup>b</sup>	1.1 ± 0.2	0.9 ± 0.2
MVE (cm/s)	73.8 ± 3.9	75.0 ± 3.2	72.1 ± 3.5
MVA (cm/s)	44.2 ± 2.1	45.1 ± 2.0	46.3 ± 1.9
E/A	1.65 ± 0.29	1.67 ± 0.28	1.52 ± 0.30
E/E' aver	9.24 ± 1.42 <sup>a,b</sup>	7.00 ± 0.9	5.76 ± 0.8
Septal E' velocity (cm/s)	6.2 ± 0.70 <sup>a,b</sup>	10.0 ± 0.9	11.1 ± 0.82
Septal A' velocity (cm/s)	5.6 ± 0.6	6.7 ± 0.6	7 ± 0.7
Lateral E' velocity (cm/s)	9.1 ± 0.6 <sup>a,b</sup>	13.4 ± 0.5	14 ± 0.5
Lateral A' velocity (cm/s)	7.1 ± 0.5	7.9 ± 0.6 <sup>c</sup>	8 ± 0.7

A: AAS users; B: nonusers; C: controls; <sup>a</sup>  $p < 0.05$  A versus B; <sup>b</sup>  $p < 0.05$  A versus C; <sup>c</sup>  $p < 0.05$  B versus C; IVSd: septal wall thickness in diastole; PWd: posterior wall thickness in diastole; LVEDD: LV end diastolic diameter; LV mass: left ventricular mass; LVMi: LV mass index; RWT: relative wall thickness; EF: ejection fraction; LAVi: LA maximal volume index; TR: tricuspid regurgitation systolic jet velocity; MVE: mitral peak E-wave velocity; MVA: mitral peak A-wave velocity; Septal E' velocity: mitral annular early diastolic peak E -wave velocity in septum; Septal A': mitral annular late diastolic peak A -wave velocity in septum; Lateral E': mitral annular early diastolic peak E -wave velocity in the lateral wall; Lateral A': mitral annular late diastolic peak A -wave velocity in the lateral wall; E/E' aver = ratio of the early diastolic transmitral flow velocity to the average of septal and lateral early diastolic mitral annular velocity.



**Figure 1.** An example of tissue Doppler recordings of lateral (a) and septal (b) annular velocities from an athlete in group A.

In group A, significant correlations were obtained between (a) the years of AAS use and E/E' aver ( $r = 0.609, p = 0.001$ ); (b) BRS and E/E' aver ( $r = -0.426, p = 0.006$ ) as well as Lateral E' ( $r = 0.325, p = 0.041$ ); (c) VO<sub>2</sub>max and LVMi ( $r = 0.372, p = 0.018$ ).

#### 4. Discussion

The use of AAS by athletes has been associated with cardiovascular disorders, either acute (such as arrhythmogenic SCD, thromboembolic episodes, or myocardial infarction) or chronic (such as hypertension, atherosclerosis, or LV hypertrophy and dysfunction) [1–7]. Our results indicate that systemic use of AAS in strength-trained athletes leads to altered cardiac autonomic and hemodynamic function when assessing spontaneous BRS and short-term HRV indices. Moreover, AAS use seems to enhance LV hypertrophy and may accelerate LV diastolic dysfunction, depending on the intake years. An association between indices of early diastolic dysfunction and BRS depression was found in our anabolic users. Additionally, an increased maximal cardiopulmonary efficiency was established in AAS users; this finding was associated with an increased LVMI.

The BRS indicates the function of the baroreflex arch and is strictly linked with heart rate and blood pressure fluctuations. Measurement of BRS respects the interbeat interval in milliseconds per unit change, known as HRV, and blood pressure in mm Hg. The increase or decrease in HRV in response to a reduction or elevation of BP by baroreflex may occur by activating either sympathetic or parasympathetic limb, or both [16]. BRS follows the synergy between the vascular and autonomic functions to guide the BP fluctuations within normal levels. Thus, BRS modulates blood pressure fluctuations by changing the HR, myocardial contractility, and peripheral resistance [17].

Exercise training can improve BRS in healthy individuals and patients with cardiac autonomic disorders [18,19]. Subramanian et al. [16] supported that athletic training positively influences baroreflex and autonomic function. Short-term exercise training lowered standing HR in postural orthostatic tachycardia syndrome, attributable to a training-induced increase in BRS [20]. Reduced BRS was shown to be associated with high blood pressure, whereas resetting the baroreceptor working range to a higher level was observed in hypertension [21,22]. Additionally, impaired baroreflex sensitivity was found in depressive disorders; such abnormality may be a predisposing factor for sudden death in patients with underlying cardiac disease [23]. Reduced BRS was also associated with obesity, diabetes, and metabolic syndrome in adolescents and adults and was an acknowledged cardiovascular risk factor [24–26]. The long-time appearance of increased BP creates a “resetting” of the baroreflex so that the BP responses to exercise are regulated around a higher defined point [27]. In the early stages of BRS dysfunction, subjects may have normal resting BP, and the abnormal pressure responses may be disclosed during effort [28]. In hypertensive patients with LV hypertrophy, the LV diastolic function has independent associations with BRS parameters obtained at rest [29]. Several studies in heart failure patients showed that sympathetic hyperactivity is triggered by lower BRS [30,31]. In heart failure patients, the increased plasma levels of angiotensin II, due to the activation of the renin–angiotensin system, cause alteration on baroreflex control of sympathetic activity and HR directly in the vasomotor and cardiac centers in the brain and the peripheral nerve terminals, facilitating norepinephrine free and inhibiting acetylcholine release [30–35]. These patients may appear with abnormalities of the sinus node.

In our study, the tilt test results showed that AAS users presented significantly lower BRS and BEI compared to nonusers by 13.8% and 10.7%, respectively. Recently, there was similar evidence for lower BRS and sympathovagal imbalance in AAS users [13]. Moreover, a correlation between BP and spontaneous BRS and arterial stiffness was observed. Beutel et al. [10] reported the appearance of hypertension with differential hemodynamic changes and alterations in the reflex control in HR after chronic stanzolol administration in rats. Testosterone-treated animals presented rest bradycardia, cardiac hypertrophy, alterations in baroreflex activity, and enhanced response to sodium nitroprusside [12]. It was suggested that chronic administration of either testosterone or cocaine elicits functional changes in the activity of brain neurons regulating baroreflex responses [10,12]. Additionally, direct effects of both drugs on the heart may mediate the baroreflex activity; chronic cocaine administration increases chronotropic actions of catecholamines, while testosterone

inhibits noradrenaline reuptake from the heart, increases the levels of the pore-forming subunits, and in addition, the activity of T-type calcium channel causing reduced reflex bradycardia [12]. Although animal studies do not necessarily apply to humans, similar mechanisms could, in future studies, justify the effects of AAS on BRS in athletes. Moreover, long-term treatment with AAS in rats reduced the sensitivity of the Bezold–Jarisch reflex control of bradycardia and BP, possibly due to cardiac hypertrophy [11].

Sedentary individuals exhibit raised sympathetic tone even at rest and higher reactivity to any stress [32,33]. Subramanian et al. [16] observed that HRV (total power and SDNN) was higher in athletes. The parasympathetic tone was higher in terms of higher RMSSD and higher HF power. We reported similar cardiac autonomic adaptations in our previous studies in athletes and patients with chronic diseases following exercise training [34,35]. In the present study, there were decreased short-term HRV indices during the head-up tilt test in the strength training athletes using AAS, compared to nonuser athletes, representing a shift towards sympathetic modulation predominance. Similarly, chronic abuse of high doses of AAS in bodybuilders led to cardiac autonomic dysfunction [8]. Dos Santos et al. [13] supported a relationship between AAS use in athletes and imbalance in autonomic control of both the periphery and the heart. In experimental studies, chronic high-dose AAS administration in rats caused impairment of parasympathetic cardiac modulation, decreased HF power, and HRV [9,10,36]. It was supported that cardiac autonomic dysfunction caused by AAS use may induce arrhythmias and sudden cardiac death [10].

Regular endurance, resistance, and combined training improve BP levels in hypertensive patients [37]. The reduction of high BP with exercise training is mainly due to attenuation in peripheral vascular resistance, caused by a reduction in sympathetic nerve activity and an increase in arterial lumen diameters [38]. The relationship between cardiac sympathetic overactivity and its association with cardiovascular diseases, such as hypertension and heart failure, is well established [39]. Controversies exist concerning the effects of AAS on BP. Some investigators have observed increased BP in strength trainees using AAS, whereas others have not [1,2,5,6]. Neto et al. [8] supported that the high BP at rest was associated with increased sympathetic modulation and enhanced cardiac hypertrophy in bodybuilders using AAS [8]. In our study, a significant increase was found in SBP at rest and at maximum effort in AAS users, compared to nonusers and controls. A similar increase in SBP at rest and during maximal workload in athletes using AAS, compared to nonusers, was observed by D'Andrea et al. [40]. They suggested that AAS can also cause sodium and water retention, with a consequent increase in blood volume and pressure.

It is well known that exercise training causes cardiac structural and functional adaptations. The type (concentric or eccentric or mixed hypertrophy) depends on the type of exercise training (aerobic, strength, or mixed). Strength-trained athletes often demonstrate concentric type LV hypertrophy. That hypertrophy is benign, as associated with normal systolic and diastolic properties. The present study showed increased relative wall thickness and LV mass index in AAS users, compared to nonuser athletes. This reinforces that AAS enhances the LV hypertrophy observed in the strength-trained athletes as a cardiac adaptation to training. In similar studies, significant increases in posterior and septal wall thickness, LVM, and chamber diameter in AAS users, compared to nonuser strength-trained athletes, were reported [41–43]. Moreover, our results indicate that long-term AAS use may accelerate LV diastolic dysfunction, depending on the years of intake, which may be early identified with TDI use. Tissue Doppler imaging in the estimation of diastolic function in AAS users showed a significantly increased  $E/E'$  average ratio and reduced early diastolic tissue velocities (septal  $E'$  and lateral  $E'$ ). Early subclinical impairment of both systolic and diastolic myocardial function, mightily associated with mean dosage and duration of AAS use, was noticed by D'Andrea et al. [40]. Notin et al. suggested that the decrease in LV relaxation properties might have been due to an alteration in the active properties of the myocardium since no wall thickening was obtained in AAS-using bodybuilders [44]. The impairment of LV function in long-term AAS users is an early indication of LV dysfunction and may be sufficient to increase the risk of heart failure [45]. An autopsy study of

cardiac dimensions in 173 AAS users also demonstrated a significantly elevated cardiac mass [46]. This cardiac remodeling has similar characteristics to hypertrophic cardiomyopathy, showing a prevalence of cardiac fibrosis and impairment of systolic and diastolic LV function [46]. In an experimental study in rats, interstitial collagen increases, leading to loss of diastolic function [47]. It was suggested that high concentrations of AAS by activating cytoplasmic androgen receptors, cell membrane receptors, and secondary transmitters stimulate the renin–angiotensin–aldosterone system. Leading to an increased synthesis of myocardial fibers, LV hypertrophy, and hypertension [1]. In our study, no LV systolic function alteration was demonstrated in AAS users. On the contrary, Alizade et al. [48] reported that peak systolic right ventricle free wall strain and strain rate were reduced in bodybuilders using AAS. In former AAS users, impaired LV systolic function was also reported [49]. Other studies have reported no significant difference in cardiac dimensions and systolic and diastolic function between AAS users and nonuser weightlifters [50–52]. However, the assessment techniques play a certain role in the credibility of the findings. D’Andrea et al. supported that the strain rate imaging was a more sensitive technique that allowed more accurate evaluation of ventricular regional wall motion in AAS users than Doppler myocardial imaging and conventional ultrasound [40].

We demonstrated a correlation between early LV diastolic dysfunction indices and spontaneous BRS depression in athletes using AAS. The increased sympathetic activity in AAS users may be the link between diastolic dysfunction and BRS reduction. A similar correlation has not yet been described in the literature. An association between LV diastolic function and depressed BRS was demonstrated in different cardiovascular conditions, but the mechanism and causality were not established [53,54]. It was also reported that diastolic dysfunction evoked significant heart baroreflex impairment in hypertensive patients [29,55].

AAS abuse causes an increase in muscle mass, can speed up muscle recovery after intense training or injury, and allow users to train longer and harder. In an experimental study in rats, the administration of AAS increased the respiratory muscle mass and the diaphragm [56]. These effects on both skeletal and respiratory muscles may contribute to the improved performance observed in our AAS users. Thus, the cardiopulmonary test results showed that AAS users had significantly longer time to exhaustion by 11% and maximal oxygen uptake by 15.1%, compared to the nonuser athletes. Interestingly, aerobic capacity was positively correlated with LVMI. A few studies demonstrated that AAS increases endurance performance in athletes [51]. No significant correlations were found between cardiopulmonary testing indices and baroreflex sensitivity or HRV parameters in our athletes.

The results of our study should be interpreted in light of some limitations. The use of AAS was based on athletes’ reports, and the dose of AAS in each athlete was not quantified. We did not perform body composition measurements in the participants. Moreover, we did not measure the arterial stiffness involved in the whole mechanism of the BRS. Additionally, we did not use strain and strain rate imaging for the assessment of myocardial function. The parameters that could have influenced the autonomic activity, such as diet, stress, and environmental influence, were not measured. Finally, although we demonstrated a correlation between LV diastolic function indices and BRS in AAS users, we cannot support a cause–effect relationship.

## 5. Conclusions

Long-term AAS use in strength-trained athletes decreases BRS and short-term HRV. An improvement in aerobic capacity was found in AAS users, which was positively correlated with LVMI. An essential finding of the study is the correlation between early left ventricular diastolic dysfunction indices and reduced BRS in AAS users. The exact mechanism of this relationship should be explained in future, more extensive studies.

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Review

# Effects of Variable-Resistance Training versus Constant-Resistance Training on Maximum Strength: A Systematic Review and Meta-Analysis

Yiguan Lin <sup>1</sup>, Yangyang Xu <sup>2</sup>, Feng Hong <sup>3</sup>, Junbo Li <sup>4</sup>, Weibing Ye <sup>5,\*</sup> and Mallikarjuna Korivi <sup>5,\*</sup>

<sup>1</sup> Department of Public Instruction, Tourism College of Zhejiang, Hangzhou 311231, China; linyiguan@tourzj.edu.cn

<sup>2</sup> Student Affairs Office, Medical College, Shandong Yingcai University, Jinan 250104, China; xuyangyang@sdycu.edu.cn

<sup>3</sup> Department of Sports Operation and Management, Jinhua Polytechnic, Jinhua 321000, China; 20201015@jhc.edu.cn

<sup>4</sup> Physical Education Department, Zhejiang University of Science and Technology, Hangzhou 310023, China; 100111@zust.edu.cn

<sup>5</sup> Institute of Human Movement and Sports Engineering, College of Physical Education and Health Sciences, Zhejiang Normal University, Jinhua 321004, China

\* Correspondence: ywbls@zjnu.cn (W.Y.); mallik.k5@gmail.com or mallik@zjnu.edu.cn (M.K.); Tel.: +86-137-5799-5718 (W.Y.)

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**Abstract:** Greater muscular strength is generally associated with superior sports performance, for example, in jumping, sprinting, and throwing. This meta-analysis aims to compare the effects of variable-resistance training (VRT) and constant-resistance training (CRT) on the maximum strength of trained and untrained subjects. PubMed, Web of Science, and Google Scholar were comprehensively searched to identify relevant studies published up to January 2022. Fourteen studies that met the inclusion criteria were used for the systematic review and meta-analysis. Data regarding training status, training modality, and type of outcome measure were extracted for the analyses. The Cochrane Collaboration tool was used to assess the risk of bias. The pooled outcome showed improved maximum strength with VRT, which was significantly higher than that with CRT (ES = 0.80; 95% CI: 0.42–1.19) for all the subjects. In addition, trained subjects experienced greater maximum-strength improvements with VRT than with CRT (ES = 0.57; 95% CI: 0.22–0.93). Based on subgroup analyses, maximum-strength improvement with a VRT load of  $\geq 80\%$  of 1 repetition maximum (1RM) was significantly higher than that with CRT (ES = 0.76; 95% CI: 0.37–1.16) in trained subjects, while no significant differences were found between VRT and CRT for maximum-strength improvement when the load was  $< 80\%$  (ES = 0.00; 95% CI:  $-0.55$ – $0.55$ ). The untrained subjects also achieved greater maximum strength with VRT than with CRT (ES = 1.34; 95% CI: 0.28–2.40). Interestingly, the improved maximum strength of untrained subjects with a VRT load of  $< 80\%$  of 1RM was significantly higher than that with CRT (ES = 2.38; 95% CI: 1.39–3.36); however, no significant differences were noted between VRT and CRT when the load was  $\geq 80\%$  of 1RM (ES =  $-0.04$ ; 95% CI:  $-0.89$ – $0.81$ ). Our findings show that subjects with resistance training experience could use a load of  $\geq 80\%$  of 1RM and subjects without resistance training experience could use a load of  $< 80\%$  of 1RM to obtain greater VRT benefits.

**Keywords:** dose–response; training intensity; elastic bands; chain; training load

## 1. Introduction

Maximum strength is the maximum force a muscle can generate in a single isometric voluntary contraction [1]. The performance of athletes, especially in powerlifting and weightlifting, is directly associated with their maximum strength. Athletes in sports

such as track-and-field, wrestling, and basketball also require maximum strength for better performance [2,3]. Constant-resistance training (CRT) is a type of training that uses constant weight loads to improve the maximum strength of an individual [4]. However, CRT does not produce effective muscle stimulation over the entire range of motion because of the “sticking point” [5–7]. Variable-resistance training (VRT), also called accommodating-resistance training [8], uses an elastic band or chain and is an alternative training method to CRT. VRT facilitates different weight loads and helps to overcome the sticking point during resistance training. VRT can reduce skeletal muscle resistance in the weakest area of motion, provide greater resistance in areas with more strength, and get closer to human strength curves to make the muscles function over a broader range [9]. As a result, VRT has the potential to increase motor unit recruitment and firing rates and improve training benefits [10–12]. Many studies have shown that VRT is effective in improving maximum strength [13–15]. However, there is inconsistent evidence to support this hypothesis [16]. In addition, VRT has been shown to produce greater stimulation of muscles during the eccentric phase, thereby increasing the rate of force development and obtaining a greater muscle stretch–shortening cycle [17–19]. The training benefits of VRT are associated with neuromuscular adaptations. VRT can activate muscle fibers to participate in contractile movement to a greater extent [20,21]. VRT produces appropriate instability in the exercise and keeps muscles in a state of tension during the eccentric phase, which can help athletes recover from injuries. Therefore, VRT is beneficial in post-operative rehabilitation [4].

With the increase in research on VRT, contradictory research data have emerged. The results of several of studies have not found that VRT is better than CRT for the development of maximum strength [16,22,23]. In a study by Cronin et al. [24], participants performed supine jump squat training with a load of 8–15 repetition maximum (RM) with or without elastic bungees. The results revealed that maximum strength of participants with elastic bungees was not better than that of participants without bungees (non-bungee squat) [24]. In a similar study, participants used a combination of chains and without chains for jump squat training with a 30% 1RM load. The results also showed that VRT did not effectively improve maximum strength [24]. Ebben et al. [25] also showed that there were no significant differences in neuromuscular activation between VRT and CRT through electromyography (EMG) of the hamstrings and quadriceps.

Two recent meta-analyses attempted to address the influential role of VRT over CRT on gaining of muscular strength in different populations. These two studies reported no significant differences in the development of maximum strength between VRT and CRT [26,27]. Furthermore, these studies [26,27] were limited with a smaller number of included articles, inadequate details of subjects/training loads, lack of subgroup analysis, and results seem to be inconsistent with the widely held view. Thus, whether VRT contributes to maximum-strength improvement and quantifying the dosage of appropriate exercise for optimal strength are problems that need clarification [28]. Based on current reviews, the VRT development of maximum strength is still controversial, so further analysis is needed to unequivocally determine the effects of VRT. The purpose of this study was to verify the impact of VRT on maximum strength and to analyze the factors that limit the beneficial effects of VRT on improving maximum strength. The hypothesis was that the effects of VRT and CRT on maximum strength are the same.

## 2. Materials and Methods

### 2.1. Search Strategy

We followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA, 2020) guidelines [29] (Supplementary Materials). We searched the databases for relevant articles up to 31 January 2022 without restricting the starting date. The literature retrieval was carried out independently by researchers Y.L. and W.Y. The articles included in our study were obtained by searching for randomized and non-randomized controlled trials published in English. Articles related to variable-resistance training were searched

in the PubMed, Web of Science, and Google Scholar databases using combinations of the following keywords: variable-resistance training; accommodating resistance; chain training; elastic training; rubber band; maximum strength; compensatory acceleration training; squat, bench press; barbell deadlift. The database search was limited to peer-reviewed English journal articles. After retrieving the publications, the reference lists were searched twice for a more comprehensive inclusion of other articles of potential interest.

## 2.2. Inclusion and Exclusion Criteria

Before inclusion, the searched articles and abstracts were screened; then, the full text of each article was obtained. A strict review was then conducted in accordance with the inclusion criteria. The analysis did not limit the subjects' age, sex, training basis, sports specialty, or body composition. Inclusion criteria were as follows: (1) at least one group in the experiment was trained in variable-resistance mode; (2) the outcome measure was maximum strength; (3) the study was published in a peer-reviewed English journal.

Studies were excluded if (1) the maximum-strength index was not reported in the experiment; (2) the studies included only an abstract without full text; (3) the studies did not provide sufficient outcome data; (4) studies were duplicate publications; (5) no comparisons of the effects before and after training modes were conducted; and (6) there was a lack of a CRT group.

The articles were independently screened by two investigators (Y.L. and W.Y.) according to the inclusion and exclusion criteria. By reading the abstracts and text, articles that did not conform to our requirements were excluded. We then continued reading the full text of articles that met the inclusion criteria. If investigators' opinions were not unanimous, another review author (M.K.) was invited to negotiate and reach a consensus.

## 2.3. Quality Evaluation

The Cochrane Collaboration tool was used to determine the risk of bias for the included trial as described in the handbook [30]. The included full-text articles were assessed by two of the three review authors (Y.L., Y.X., and W.Y.), and the risk-of-bias tool was independently applied to each study. The differences were resolved by discussing with other review authors (J.L. and M.K.). Sources of biases, such as selection bias (random sequence generation and allocation concealment), performance bias (blinding of participants and personnel), detection bias (blinding of outcome assessment), attrition bias (incomplete outcome data), and reporting bias (selective reporting) were detected for all the included studies. The outcome of the risk of bias is fully described in Section 3, Results.

## 2.4. Data Extraction

The basic information from the articles that met our criteria, including authors; sex, age, and number of participants; training basis; training methods; training arrangements (training cycle, number of weekly training sessions, number of groups, number of repeats); and load, is presented in Table 1. This task was undertaken by one author (Y.L.). The second and third authors (Y.X. and F.H.) checked the extracted data for accuracy and completeness. A quality assessment was conducted by another review author (W.Y.). Disagreements were resolved by consensus or by another author (M.K.).

## 2.5. Outcome Measures

All studies used maximum strength as the evaluation indicator. The maximum-strength index was measured using a barbell for the 1RM in kilograms (kg). The maximum strength of the subjects was tested before and after training, and the change in maximum strength before and after training was measured.

## 2.6. Data Analysis

We employed the Cochrane Collaboration Review Manager (RevMan, Copenhagen, Denmark) version 5.3 for the statistical analyses. The  $I^2$  test was used to test the heterogeneity of each trial, and 25%, 50%, and 75% of the values represented low, medium, and high statistical heterogeneity. If there were no significant differences in the heterogeneity test, a fixed effects model was employed for the meta-analysis; a random effects model was used when there was high heterogeneity in the heterogeneity test. For continuous outcome variables with the different test units and methods, the standardized mean difference (SMD) and 95% confidence intervals (CIs) were selected as the effect sizes for the combined analysis. Meta-regression analysis was performed for VRT duration and load to identify their influential role on gaining maximum strength. Then, subgroup analysis was performed to determine the optimum load of VRT that could effectively improve maximum strength.

Meta-analysis data were extracted from the change values of the VRT and control groups before and after the intervention, namely, the mean  $\pm$  SD of the change values before and after training. When relevant data were unavailable, the filling method was adopted based on the research study by Bellar et al. [31], and a correlation coefficient of 0.986 was obtained. Based on the correlation coefficient, the SD changes before and after training in the remaining included articles were obtained. The calculation formula was as follows [30]:

$$SD_{\text{change}} = \sqrt{[SD_{\text{pre}}]^2 + [SD_{\text{post}}]^2 - 2 \times \text{corr} \times SD_{\text{pre}} \times SD_{\text{post}}}$$

where  $SD_{\text{change}}$  is the standard deviation of change values before and after training;  $SD_{\text{pre}}$  is the standard deviation before training;  $SD_{\text{post}}$  is the standard deviation after training; corr is the correlation coefficient.

**Table 1.** Details of the studies included in the meta-analysis.

Study	#		Sex	Age (Years)	Experience	Training Methods		Training Arrangement		Intensity (%)	
	VRT	CRT				VRT	CRT	PMR	PVR	PCR	
Sawyer et al. 2021 [32]	20	20	Male	18–25	Trained	Squat + elastic	Squat	3 × 3w [5 × (1–7)]	50–93	20	80
Arazi et al. 2020 [33]	12	12	Female	24 ± 4	Untrained	Squat + chain	Squat	3 × 8w [(3–5) × (6–12)]	65–85	15	85
	12	12	Female	24 ± 4	Untrained	Bench press + chain	Bench press	3 × 8w [(3–5) × (6–12)]	65–85	15	85
Kashiani et al. 2020 [34]	17	16	Male	22 ± 2	Untrained	Overhead press + chain	Overhead press	3 × 12w [3 × (8–12)]	70–80	35	65
	17	16	Male	22 ± 2	Untrained	Overhead press + elastic	Overhead press	3 × 12w [3 × (8–12)]	70–80	35	65
Katushabe et al. 2020 [35]	9	8	Male	21 ± 2	Trained	Squat + elastic	Squat	— × 6w [3 × (5–10)]	-	20	80
	9	8	Male	21 ± 2	Trained	Deadlift + elastic	Deadlift	— × 6w [3 × (5–10)]	-	20	80
Archer et al. 2016 [24]	11	10	Male	24 ± 2	Trained	Squat jump + chain	Squat jump	3 × 1w [5 × 3]	30	20	80
	16	16	Female	24 ± 6	Trained	Squat + elastic	Squat	2 × 10w [(3–4) × (6–10)]	75–85	27–58	42–73
Ataee et al. 2014 [8]	8	8	Male	21 ± 2	Trained	Squat + chain	Squat	3 × 4w [1 × 5]	85	20	80
	8	8	Male	21 ± 2	Trained	Bench press + chain	Bench press	3 × 4w [1 × 5]	85	20	80
Bellar et al. 2011 [31]	11	11	Male	24 ± 3	Untrained	Bench press + elastic	Bench press	2 × 13w [5 × 5]	85	15	85
	10	11	Mixed	20 ± 1	Untrained	Bench press + elastic	Bench press	3 × 24w [(3–6) × (6–10)]	67–95	20–35	65–80
Shoepet et al. 2011 [16]	10	11	Mixed	20 ± 1	Untrained	Squat + elastic	Squat	3 × 24w [(3–6) × (6–10)]	67–95	20–35	65–80
	10	9	Female	20 ± 2	Trained	Bench press + chain	Bench press	2 × 8w [3 × (4–6)]	80–90	5	95
Burnham et al. 2010 [36]	12	12	Male	20 ± 1	Trained	Bench press + elastic	Bench press	4–5 × 7w [(5–6) × (4–6)]	85	-	-
	12	12	Male	20 ± 1	Trained	Bench press + chain	Bench press	4–5 × 7w [(5–6) × (4–6)]	85	-	-
Chigiarelli et al. 2009 [37]	13	12	Male	21 ± 1	Trained	Bench press + chain	Bench press	2 × 9w [(5–7) × (5–10)]	60–95	10–20	80–90
	16	16	Male	21 ± 2	Trained	Squat + elastic	Fast squat	2–3 × 13w [4 × 10]	75–85	-	-
McCurdy et al. 2009 [23]	16	16	Male	21 ± 2	Trained	Squat + elastic	Slow squat	2–3 × 13w [4 × 10]	75–85	-	-
	16	16	Male	21 ± 2	Trained	Squat + elastic	Bench press	3 × 7w [(3–6) × (2–10)]	72–98	20	80
Rheaet et al. 2009 [38]	23	21	Mixed	20 ± 1	Trained	Bench press + elastic	Squat	3 × 7w [(3–6) × (2–10)]	72–98	20	80
	23	21	Mixed	20 ± 1	Trained	Squat + elastic	Squat	3 × 7w [(3–6) × (2–10)]	72–98	20	80

Note: The content of the study design comprises training times per week × training weeks [(sets) × (repetitions)], excluding warm-up and relaxation. VRT = variable-resistance training; CRT = constant-resistance training; w = week; PMR = percentage of maximum repetitions; PVR = percentage of variable resistance; PCR = percentage of constant resistance; n = number of participants.

### 3. Results

#### 3.1. Search and Exclusion Results

Following a systematic search, we retrieved a total of 2436 articles. After removing the duplicate records (1132), 331 records were marked as ineligible by automation tools, and 467 records were removed for other reasons. From the remaining (506) records, 471 were excluded according to our study criteria, leaving 35 articles. Finally, there were 35 articles relevant to our study. The remaining 35 articles were further evaluated, and 21 were screened out for the following reasons: 3 studies did not report average or standard deviations [17,40,41]; 1 study did not report maximum-strength indicators [42]; 15 studies did not compare the effects before and after the training intervention [15,20,21,25,43–53]; 2 studies had no CRT group [54,55]. Finally, a total of 14 studies were included in the meta-analysis. The specific screening steps are shown in Figure 1.

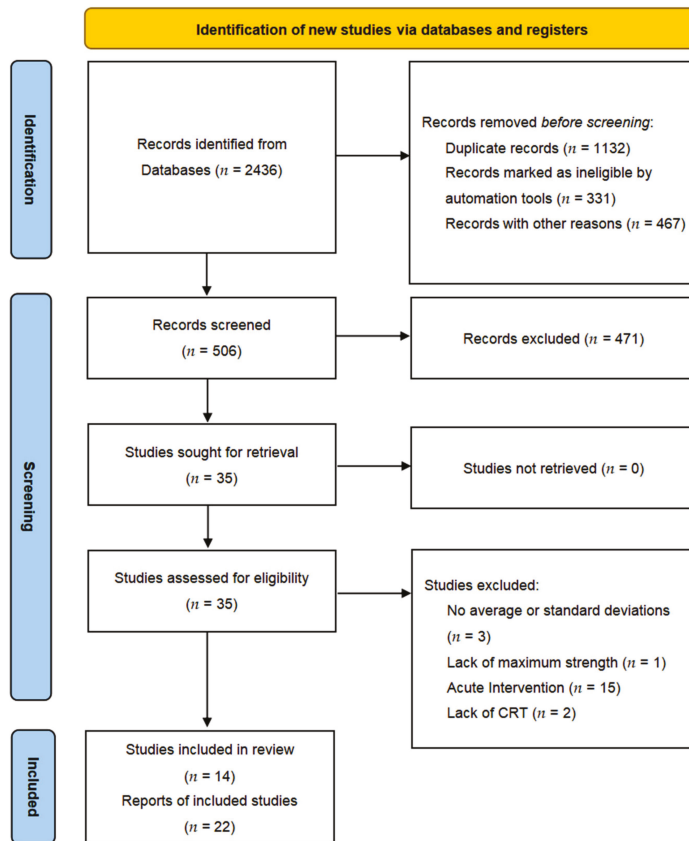


Figure 1. Preferred Reporting Items for the Systematic Review and Meta-Analysis (PRISMA) flow diagram of article selection.

#### 3.2. Description of Included Studies

In our systematic review and meta-analysis, we included 14 studies comprising 22 reports. These studies were intercontinental, and published between 2008 and 2021. The reports involved 414 participants (trained and untrained) with a mean age between 18 and 30 years. The specific details are shown in Table 1. Of the included studies, three studies only recruited female participants, nine only involved male participants, and two

involved male and female participants. In terms of training, four studies were conducted on untrained subjects, and 10 studies were conducted on trained subjects. The main training methods were squatting and bench pressing. The VRT forms included chain and elastic resistance combined with barbells. In terms of the training period, 10 studies were  $\leq 10$  weeks, and four studies were  $>10$  weeks. The percentage of maximum repetitions was from 30 to 95%; the proportion of the variable load component accounted for 10–35% of the total load.

### 3.3. Meta-Analysis Results of VRT and CRT Modes on Maximum Strength

A total of 22 reports that comprised both trained and untrained participants were included for the meta-analysis [8,16,22–24,31–39]. As shown in Figure 2, VRT and CRT significantly differed in the improvement of the maximum strength of the subjects (ES = 0.80; 95% CI: 0.42–1.19). However, high statistical heterogeneity ( $I^2 = 78\%$ ) was detected in our analysis.

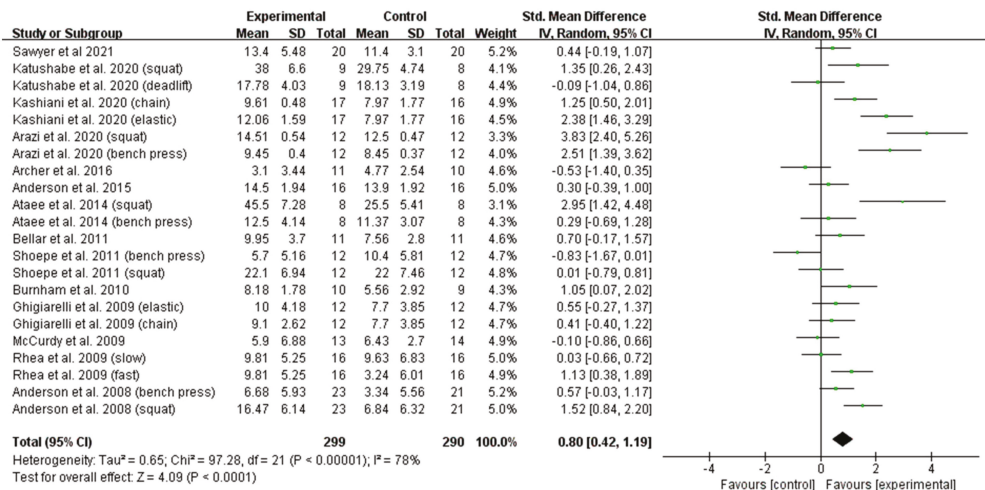


Figure 2. Forest plot of maximum-strength development comparison between VRT and CRT.

### 3.4. Influence of VRT and CRT on Maximum Strength of Trained Subjects

The meta-analysis conducted on the studies of only trained subjects showed that VRT favored a significantly higher improvement of maximum strength than CRT (ES = 0.57; 95% CI: 0.22–0.93; Figure 3) [8,22–24,32,36–39]. Based on the VRT workload, we then subgrouped the studies into  $<80\%$  and  $\geq 80\%$  1RM. As reported in Figure 3, the effect of VRT with a load of  $\geq 80\%$  1RM on the maximum-strength development was significantly higher than that of CRT (ES = 0.76; 95% CI: 0.37–1.16). However, no significant differences were observed between VRT and CRT in the improvement of maximum-strength when the load of VRT was  $<80\%$  1RM (ES = 0.00; 95% CI:  $-0.55$ – $0.55$ ; Figure 3).



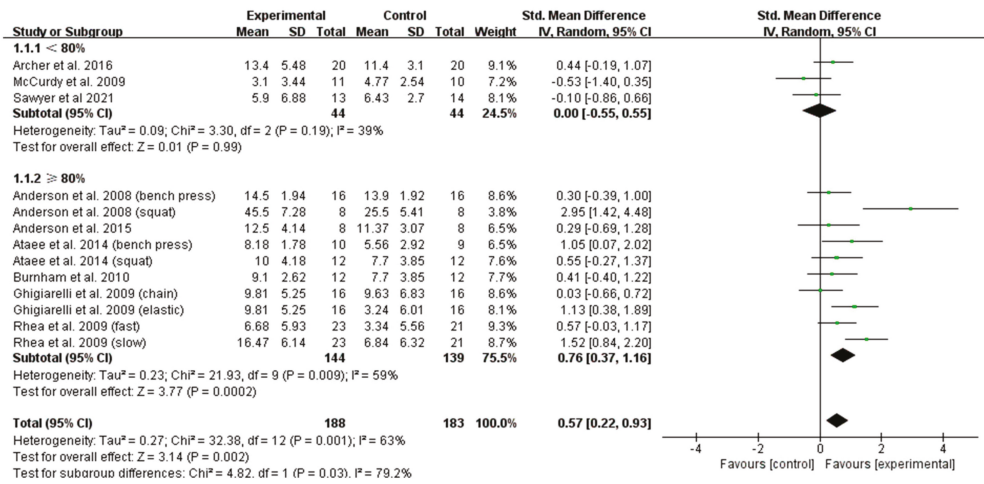


Figure 3. Forest plot of maximum-strength development: comparison between VRT and CRT after sensitivity analysis in trained subjects.

### 3.5. Influence of VRT and CRT on Maximum Strength of Untrained Subjects

As shown in Figure 4 [16,31,33,34], maximum-strength gains were significantly higher with VRT than CRT in the untrained subjects (ES = 1.34; 95% CI: 0.28–2.40). Interestingly, the subgroup analysis showed that the effect of VRT with a load of <80% 1RM on maximum-strength gain was significantly greater than that of CRT (ES = 2.38; 95% CI: 1.39–3.36). Nevertheless, we found no significant differences between VRT and CRT in the development of maximum strength when the load of VRT was ≥80% 1RM (ES = −0.04; 95% CI: −0.89–0.81) in the untrained subjects (Figure 4).

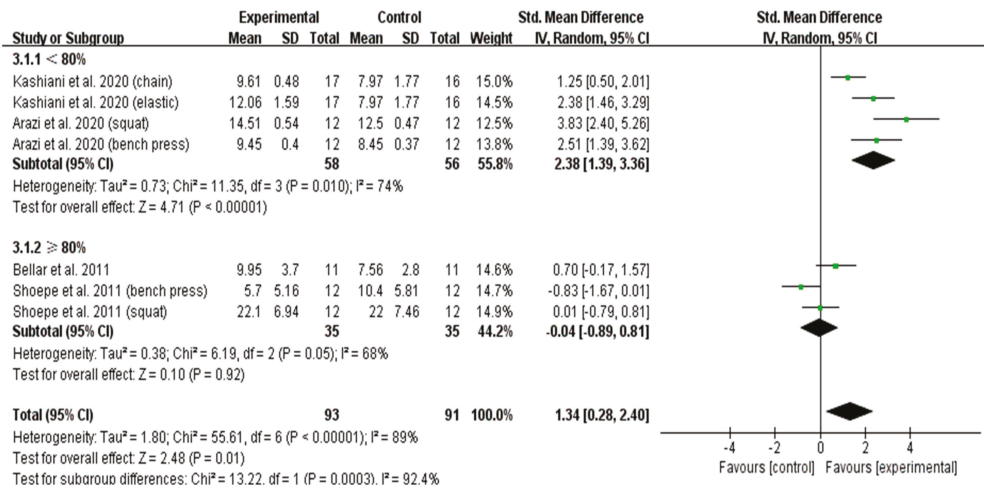


Figure 4. Forest plot of maximum-strength development: comparison between VRT and CRT after sensitivity analysis in untrained subjects.

### 3.6. Risk of Bias in the Results

We used the Cochrane collaborative method to assess the risk of bias (Figure 5) [8,16,22–24,31–39]. For the selection bias, 12 trials reported random sequence generation and two non-randomized groupings; no reports of concealment and blinding were documented, and all the literature was rated as having a high risk. The outcome variable evaluation was not mentioned in 14 studies, and all articles were assessed as being unclear. In our evaluation, one study identified a reporting bias. No experiments indicated follow-up bias or other biases.

	Random sequence generation (selection bias)	Allocation concealment (selection bias)	Blinding of participants and personnel (performance bias)	Blinding of outcome assessment (detection bias)	Incomplete outcome data (attrition bias)	Selective reporting (reporting bias)	Other bias
Anderson et al. 2008	+	?	-	?	+	+	+
Anderson et al. 2015	+	?	-	?	+	+	+
Arazi et al. 2020	+	?	-	+	+	+	+
Archer et al. 2016	+	?	-	?	+	+	+
Ataee et al. 2014	+	?	-	?	+	+	+
Bellar et al. 2011	+	?	-	?	+	+	+
Burnham et al. 2010	+	?	-	?	+	+	+
Ghigiarelli et al. 2009	+	?	-	?	+	+	+
Kashiani et al. 2020	+	?	-	?	+	+	+
Katushabe et al. 2020	+	?	-	?	+	+	+
McCurdy et al. 2009	-	?	-	?	+	-	+
Rhea et al. 2009	+	?	-	?	+	+	+
Sawyer et al. 2021	+	?	-	?	+	+	+
Shoepe et al. 2011	-	?	-	?	+	+	+

Figure 5. Summary of the risk of bias of studies included in this meta-analysis. Green indicates a low risk of bias, yellow indicates unclear bias, and red indicates a high bias risk.

#### 4. Discussion

To the best of our knowledge, this is the first systematic review and meta-analysis to investigate the effect of VRT and VRT load on maximum-strength gain in comparison with CRT. Our results show that VRT was better than CRT in improving the maximum strength of trained and untrained subjects. Furthermore, the VRT-improved maximum strength depended on the workload. The subgroup analysis showed that the VRT beneficial effect was better when the untrained subjects used a <80% 1RM load and when the trained subjects used a load of  $\geq 80\%$  of 1RM.

Many studies have indicated that a  $\geq 80\%$  1RM load is the most conducive to developing muscle strength and have used this as the boundary value of the load [2,56]. During strength training, as the resistance increases, the speed of the movement gradually decreases, resulting in a “sticking region” at the weakest position of the joint. When athletes use CRT in heavy-load training, they often fail to lift weights in the concentric phase because of the sticking region of their movements, thus reducing the degree of stimulation produced by training on the target muscles. When VRT is used, the resistance of weak muscle points is reduced, which, in turn, reduces the probability of weight lifting failure. At the same time, the resistance gradually increases in the latest stage of the action and exceeds the maximum weight that could be lifted when CRT is used, thus producing greater stimulation of the target muscles. Therefore, it is likely that the concentric stage of VRT is the most favorable component to facilitate the development of maximum strength, especially in the latest stage of the concentric action [57]. Israel et al. [15] showed, using EMG, that in the squat movement, the activation of vastus lateralis was the highest in the early stage of the concentric phase and late stage of the eccentric phase under VR conditions. During squat and bench presses, VRT is able to provide progressive resistance to match the human strength curves [4]. The early stage of the concentric phase and the late stage of the eccentric phase are the stages in which the greatest resistance occurs, and stimulation with a heavy load is necessary to increase strength.

This meta-analysis shows that trained subjects obtained a better effect from VRT with a training load  $\geq 80\%$  of 1RM. When training with a smaller load, the load does not reach the limit of muscle strength, and there is no sticking region [7]. At the same time, the muscles do not bear the overload at the latest stage of the movement. VR is not enough to stimulate the growth of strength to a great extent, and the benefits brought by VR are reduced. For trained subjects, less than 80% 1RM loads did not properly stimulate the muscles, so the training effect of VRT was not significant compared with CRT. However, movement speed may be a factor in maximum strength. Rhea et al. [38] found that the increase in maximum strength was more significant with slow training. This may be related to slower training during the concentric phase contributing to the increased cross-sectional area of type I and type II-a skeletal fibers [46]. In general, when the load is small, VRT may trigger higher movement speeds, which affects the increase in maximum strength to some extent [43,44,46]. Cronin et al. [41] and Archer et al. [24] both used lower loads for power training, and the results showed that the VRT group had a lower maximum-strength increase than the CRT group. However, Stevenson et al. [47] argued that VRT can increase the speed of the eccentric phase but can harm the speed of the concentric phase. Recent analyses of the mechanism by which VRT increases maximum force revealed that the speed increase in VRT mainly occurred during the eccentric phase and that eccentric acceleration may contribute to the maximum increase in strength [57,58].

Another meta-analysis concluded that a <80% 1RM VRT load had a more significant effect on untrained subjects. Strength improvement mainly depends on muscle and nerve adaptation. Muscle adaptation includes improved energy reserves, increased muscle fiber size, and capillary density. Nerve adaptation includes the activation of motor units, intermuscular coordination, and changes in the discharge frequency of motor neurons [59]. Hakkinen et al. [60] found that the first 8 weeks of strength training mainly improved nerve adaptability, while the second 8 weeks increased the muscle fiber size. Several

studies have suggested that the maximum strength increased by VRT is mainly related to improvements in neuromuscular adaptation [20,39]. For trainers who have had long-term strength training, their power may increase to a higher level. However, it becomes complicated to increase other muscles' sizes and strength. Adding a chain or elastic band to the free weights or changing the state of the body movement can provide a new stimulus for the muscles and improve the coordination between the muscles in the fight against unfixed resistances, thus improving the development of strength. Mina et al. [21] compared the effects of VRT and CRT on post-activation potentiation (PAP). Their results showed that warming up with VRT was more beneficial in improving subsequent 1RM performance. A recent study by Smith et al. [9] reported that VRT showed shorter electrochemical (reflex-EMDE-M) and mechanical (reflex-EMDM-F) activities after four weeks of training. These studies also support the opinion that VRT training can improve neuromuscular adaptation. For individuals with no training experience, strength enhancement is mainly based on neural adaptation; the mobilization ability of muscles is weak, and the excessive load may not produce optimal stimulation of muscles. Therefore, it is more appropriate to use a load of <80% of 1RM for VRT.

In VRT, the ratio of VR to CR is also an aspect worth exploring. Some research groups have suggested that if the goal is to develop maximum strength, training with VR accounts for 15–35% of the total resistance and produces a better training effect [17,39,61]. If the purpose of training is to develop explosive force, a VR load accounting for 10–20% of the total resistance should be used for training [44,61]. In two studies, 80% of 1RM (5% VR, 75% CR) and 85% of 1RM (5% VR, 80% CR) were used to carry out a comparative study of Olympic clean and snatch exercises. The results revealed that there were no significant differences between the force output of Olympic clean and snatch in the VRT form and that of CRT. The subjects also reported that it was more challenging to carry out Olympic clean and snatch exercises in the VRT form [13,50]. Therefore, the impact of different training actions should also be considered in the implementation of training regimes, which is again a topic that requires further research.

There are a few limitations to the present study. First, due to the lack of necessary data, several relevant studies were excluded, which resulted in a relatively lower number of included studies. Second, there were no allocation concealment and blinding methods in the studies' quality evaluations, so the risk of bias was high. Third, some of the loads used in the included studies were varied. In our study, an intermediate load value was considered for the total load, but there may have been some deviation. Despite these limitations, overall, our meta-analysis provides new ideas and conclusions that emphasize the beneficial effects of VRT and its load on improving the maximum strength of trained and untrained athletes.

## 5. Conclusions

Our findings suggest that VRT is better than CRT in improving maximum strength. Trainers of different levels should choose the corresponding load of VRT for training according to the actual situation. Our findings recommend that trained subjects could use a load of  $\geq 80\%$  of 1RM and that untrained individuals could use a load of <80% of 1RM to attain greater VRT benefits. Future research should refine the training load, such as distinguishing between different-level trainers using the training benefits of VRT with different loads, and the proportion of variable resistance in the total load. In addition, the specific differences in the training effects of the two forms of VRT, the iron chain, the elastic band, and the impact of the training cycle are still worthy of further research.

**Supplementary Materials:** The following materials are available online at <https://www.mdpi.com/article/10.3390/ijerph19148559/s1>, PRISMA 2020 checklist and PRISMA 2020 abstract checklist.

**Author Contributions:** Conceptualization, M.K., W.Y. and Y.L.; Methodology, Y.L. and F.H.; Validation, W.Y., Y.L. and M.K.; Formal analysis, Y.X. and Y.L.; Data curation, Y.L., Y.X. and J.L.;

Writing—original draft preparation, Y.L. and F.H.; Writing—review and editing, J.L. and W.Y.; Visualization, Y.X. and Y.L.; Supervision, M.K. and W.Y.; Project administration, W.Y. and M.K.; Funding acquisition, Y.L. All authors have read and agreed to the published version of the manuscript.

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Review

# An Overview on How Exercise with Green Tea Consumption Can Prevent the Production of Reactive Oxygen Species and Improve Sports Performance

Hadi Nobari <sup>1,2,\*</sup>, Saber Saedmocheshi <sup>3</sup>, Linda H. Chung <sup>4</sup>, Katsuhiko Suzuki <sup>5</sup>, Marcos Maynar-Mariño <sup>6</sup> and Jorge Pérez-Gómez <sup>1</sup>

- <sup>1</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Caceres, Spain; jorgepg100@gmail.com
  - <sup>2</sup> Department of Physical Education and Sports, University of Granada, 18010 Granada, Spain
  - <sup>3</sup> Department of Physical Education and Sport Sciences, Faculty of Humanities and Social Sciences, University of Kurdistan, 66177-15175 Sanandaj, Kurdistan, Iran; saedsaber384@gmail.com
  - <sup>4</sup> Research Center for High Performance Sport, Campus de los Jerónimos, Catholic University of Murcia, Guadalupe, 30107 Murcia, Spain; lhchung@ucam.edu
  - <sup>5</sup> Faculty of Sport Sciences, Waseda University, Tokorozawa 359-1192, Japan; katsu.suzu@waseda.jp
  - <sup>6</sup> Department of Physiology, School of Sport Sciences, University of Extremadura, 10003 Caceres, Spain; mmaynar@unex.es
- \* Correspondence: hadi.nobari1@gmail.com

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**Abstract:** Free radicals are reactive products that have multiple effects on the human body. Endogenous and exogenous antioxidants manage the overproduction of free radicals. However, an imbalance between free radicals and antioxidant factors causes oxidative stress. Exercise and physical activity are factors that increase oxidative stress and disrupts the body's homeostasis. Intensity and duration of training, training characteristics, and fitness level can have positive or negative effects on oxidative stress. Green tea consumption is recommended for the prevention of a variety of diseases, health maintenance, and weight loss. The effectiveness of green tea is primarily due to the presence of catechins and polyphenols, specifically (–)-epigallocatechin-3-gallate, which has antioxidant and anti-inflammatory properties based on clinical and animal studies. This review investigates the effect of green tea exercise and their interactive effects on free radicals and sports improvement.

**Keywords:** antioxidant status; nutrition; performance; physical activity; reactive oxygen species (ROS)

## 1. Introduction

Regular exercise is fundamental for maintaining a healthy lifestyle. Specifically, >150 min of moderate-intensity aerobic activity or >75 min of intense physical activity (PA) per week is recommended for cardiovascular health and optimal function of body organs, while 300 min of moderate-intensity exercise per week is recommended to lower the risk of cancer [1]. These recommendations are based on many systematic reviews and meta-analyses of epidemiological studies [2]. However, exercise has both positive and negative effects on the inflammatory status, oxidative stress, and function of body organs. While moderate-intensity exercise improves immune function as opposed to sedentary behavior, immune system activity can be harmed by prolonged and intense exercise [3,4]. Biomarkers, such as lipid peroxidation, oxidized glutathione, and total radical-trapping antioxidant parameter, have important roles in health and body function as well as during exercise [4]. However, there is limited research regarding the effect of exercise on immune system activity and oxidative stress [5].

Exercise is commonly divided into aerobic and resistance training. Aerobic exercise (e.g., running, walking, swimming, and cycling) improves the cardiovascular system and



significantly reduces reactive oxygen species (ROS) and various ROS-related reactions. Aerobic exercise increases the presence of antioxidants and improves the expression of antioxidant enzymes, thereby counteracting exercise-induced oxidative stress [6,7]. On the other hand, resistance training provokes an increase in oxidative and inflammatory stress despite promoting neural and structural (i.e., muscle fibers) adaptations. The rest period lowers oxidative stress, where there is repair of minor injuries induced by resistance training due to increased levels of oxidative and inflammatory stressors. Strenuous physical exercise can stimulate high production of ROS and reactive nitrogen species (RNS) in skeletal muscle. Depending on the exercise level, duration, frequency, sex, age, and fitness, high concentration of reactive species can be harmful to the body if it is not neutralized by the available antioxidant enzymes [8]. Nevertheless, the adaptations that occur from regular and intense exercise reduce overall oxidative stress over time [8].

An imbalance between oxidants and the production of free radicals in the body with the body's antioxidant status leads to an increase in ROS, which ultimately leads to oxidative stress [9]. Oxidative stress induced by physical activity can reduce the effectiveness of endogenous antioxidants and may lower athletic performance [10]. However, the consumption of rich sources of antioxidants (such as polyphenols and flavonoids) can decrease oxidative stress levels [10]. One source of antioxidants is tea, which is the second most consumed beverage in the world after water [11]. Evidence shows that tea was first used in China around 2737 BC not only as a beverage but also for medicinal purposes [12]. There are three types of tea found in nature: green tea, oolong tea, and black tea. The elements of tea are mainly polyphenols, caffeine, and minerals, along with small amounts of vitamins, amino acids, and carbohydrates. The polyphenol that predominates in tea varies depending on the type of tea. For example, catechin is the major polyphenol in green tea, while tannin is the major polyphenol in black tea [12]. These natural substances have antioxidant properties with very limited side effects. Green tea has been shown to significantly reduce free radicals and oxidative stress [12], which will be discussed in more detail in the following sections. Therefore, green tea consumption together with physical activity can be more effective in eliminating or reducing oxidative stress than drinking green tea or doing exercise alone. Given the wide range of signaling pathways involved with exercise and tea consumption, this overview will only address oxidative stress, the antioxidant effectiveness of green tea and exercise, and their interaction.

## 2. Production of Free Radical and oxidants

ROS are metabolic by-products that include superoxide ( $-O_2$ ), hydrogen peroxide ( $H_2O_2$ ), hydroxyl ( $-OH$ ), and single oxygen ( $-O_2$ ) radicals. Protein phosphorylation, activation of multiple transcription factors, apoptosis, immunity, and differentiation require a certain amount of ROS production, which is produced by the cell at low levels. However, overproduction of ROS can damage important structures within the cell, such as proteins, lipids, and nucleic acids [13,14]. The mitochondria are the primary organelles that produce ROS, but endothelial and inflammatory cells can also produce ROS because of cellular respiration under physiological and pathological conditions [13,14]. Although these cells have a special capacity to produce intrinsic ROS [15,16], they cannot reach the same quantity as mitochondrial ROS [15]. ROS production is dependent on enzymatic and nonenzymatic reactions. Enzymatic reactions, such as respiratory chain reactions, are capable of producing ROS [15]. Nonenzymatic reactions involve oxygen reacting with organic matter, thereby yielding free radicals [15]. The production of free radicals can be endogenous or exogenous. The endogenous pathway can result from immune cell activation, inflammation, ischemia, infection, cancer, excessive exercise, mental stress, and aging, while the exogenous pathway can arise from external factors, such as pollution, smoking, drugs, etc.

## 3. Physiological Levels of Reactive Oxygen Species (ROS)

At physiological levels, ROS performs a variety of beneficial body functions. For example, ROS is needed for the synthesis of certain cellular structures and is required by

the host defense immune system to counteract external factors. Specifically, phagocytes synthesize and store free radicals to be released when exposed to pathogens. Free radicals play an important regulatory role in cellular signaling pathways and have a function in certain cell types, such as fibroblasts, endothelial cells, vascular smooth muscle cells, cardiac myocytes, and thyroid tissue [17]. In addition, the production of free radicals is physiologically beneficial to human health, such as for patients with granulomatous disease who cannot produce O<sub>2</sub>. At the physiological level, mitochondrial ROS, especially superoxide anion and hydrogen peroxide, play a positive role in responding to factors such as vascular shear stress and reducing vascular resistance to blood flow [18]. Assessing the availability of oxygen is very important for cellular health because it allows cells to initiate adaptive reactions to survive and access oxygen. Schumacker observed that the electron transfers chain acts as an O<sub>2</sub> deficiency marker by releasing ROS in response to hypoxia, which in turn initiates a signaling mechanism to react appropriately to this process, such as by increasing production and stabilizing HIF-1 [19]. In the physiological state of ROS production, skeletal muscle can also be a target organ for regulating oxidation and oxidative stress. Because muscles need a lot of energy during exercise, this process can increase mitochondrial ROS production. Several pathways can increase ROS production in skeletal muscle, including muscle contraction, insulin, and hypoxia. For example, ROS can be a signal mediator in regulating skeletal muscle glucose uptake during muscle activity [18].

#### 4. Overproduction of Free Radicals in the Body: Role of Mitochondria in Oxidative Stress Production and Immune System Response

The overproduction of free radicals and oxidants (i.e., nonphysiological level) causes oxidative stress, which can be responsible for several pathological diseases that affect different tissues and organs and is one of the underlying factors that can be harmful to overall health. This lack of balance between free radical production and antioxidant neutralization can consequently be harmful to body tissues (e.g., cell membranes, lipids, proteins, lipoproteins, and deoxyribonucleic acid structures) and can activate destructive mechanisms [20]. For example, the overproduction of  $\cdot\text{OH}$  radicals and peroxynitrite leads to lipid peroxidation, which in turn damages the cell membrane. Families of ROS, such as single oxygen, superoxide anion (O<sub>2</sub><sup>-</sup>), H<sub>2</sub>O<sub>2</sub>, peroxy radical (i.e., ROO $\cdot$ ), and  $\cdot\text{OH}$  radical, are highly reactive. Nitrogen-containing active species, such as nitric oxide (NO) and peroxynitrite anion (ONOO<sup>-</sup>), are also present. The stimulants of active species are radiation, drugs, xenobiotics, and toxins. In addition, exercise can produce free radicals if the intensity, duration, or volume of training is high, which can damage the inner membrane of the mitochondria (where aerobic respiration takes place) and disrupt cell homeostasis [5].

Mitochondria are complex organelles with a bilayer membrane, and any dysfunction in this organelle can trigger small oxidative stress signals. As mitochondria primarily function to generate energy and release various ions, they are in turn vulnerable to these charged ions. For example, high production of free radicals during mitochondrial deoxyribonucleic acid (DNA) damage causes an increase in oxidative stress on the mitochondria and can consequently damage it. Recently, thioredoxin 2 (Trx2), a low-redox protein in the mitochondria with two redox regions (C90 and C93), has attracted the attention of researchers [21]. Trx2 is found almost exclusively in tissues with high metabolic levels, such as liver, brain, and heart. Trx2 helps maintain intracellular stability through reversible oxidation of disulfide. It also regulates the function of many apoptosis-related factors, such as apoptosis signal regulator kinase 1 (ASK1) and nuclear factor kappa B (NF- $\kappa$ B) [22]. Low levels of Trx2 expression causes the release of cytochrome c from the mitochondria, followed by activation of caspase-3 and -9. Furthermore, Trx2 reduces mitochondrial reactive oxygen species (mctROS) and tumor necrosis factor (TNF)-dependent apoptosis. Thus, low-redox mitochondrial protein Trx2 is a regulator of oxidative stress.

Research has shown that mitochondria can regulate the response of immune cells via oxidative stress reactions [23]. For example, the mitochondrial process regulates the function of memory T cells [24,25]. ROS are an important factor in stimulating the immune

system [8]. When responding to oxidative stress, the immune system produces factors that consequently cause oxidative stress. This means active species do not recognize their own factors, so the immune-response-induced oxidative stress may also attack their own factors, creating a vicious cycle. However, the immune system also fights against the oxidative stress agent by activating its complex antioxidant system (which include enzymes, minerals, and vitamins) to neutralize the reactive species [26]. Reactive active species release proinflammatory cytokines, TNF-alpha (TNF- $\alpha$ ) and interleukins, which eventually initiate an inflammatory response. Accumulation of these inflammatory factors releases the body's defense factors, such as interferon-gamma (IFN- $\gamma$ ), cluster of differentiation 14 (CD14), or TNF- $\alpha$ . In addition, the immune response produces an increase in energy and physiological costs, an example of the latter being a fever, which is produced by cytokine activity. A fever of above two degrees Fahrenheit has a metabolic cost of 175 KJ in a person. Thus, this can delay recovery after exercise [27,28]. Regardless, the endogenous antioxidant defense system eliminates these active species. Furthermore, consuming exogenous antioxidant supplements can help maintain proper immune system function [29].

## 5. Neutralizing Free Radicals: Green Tea

Natural interventions to reduce the negative effects of exercise and improve body function during exercise has found its place in the athletes' training program. The use of supplements is often promoted to athletes with claims of improving performance, but it may not clearly specify their ergogenic effects. Thus, extensive research is being done on the role of their effectiveness on sports performance [30].

Among dietary and natural supplements, tea is popular and widely used due to its antioxidant, anti-inflammatory, and anticarcinogenic properties [29]. Among them, black tea is highly consumed (78% popularity), followed by green tea (20% popularity) [31]. Green tea is a very potent beverage because of its antioxidant, anti-inflammatory, anticarcinogenic, and antiallergic properties [32]. Additionally, green tea decreases the symptoms of metabolic syndrome and diabetes by controlling blood glucose, lowering cholesterol, etc. [31].

### 5.1. Green Tea Chemical Composition and Its Biological Characteristics

The chemical formula of green tea consists of proteins and amino acids (20–25% raw material), such as glutamic acid, tryptophan, glycine, serine, aspartic acid, tyrosine, valine, leucine, threonine, arginine, sinus, and carbohydrates (5–7% raw material; cellulose, glucose, fructose, and sucrose). It also contains lipids (nickel linoleic acid and linoleic acid), vitamins (B, C, and E), caffeine, chlorophylls, and carotenoids. Unstable compounds (aldehydes, alcohols, esters, lactones, and hydrocarbons), minerals, and essential elements (5% dry weight; Ca, Mg, Cr, Mn, Fe, Cu, Zn, Mo, Se, Na, P, Co, Sr, Ni, K, F, and Al) are also included. Green tea has a rich source of polyphenols, such as flavonoids. Flavonoids are phenolic derivatives that differ in their concentration in green tea [33]. Catechins are the most important flavonoids in green tea.

The components in green tea have medicinal properties because of the presence of polyphenols, specifically flavonoids. These flavonoids contain a high proportion of catechins (80–90%) compared to other teas. Green tea has four main catechins: epigallocatechin gallate (EGCG; 60%), epigallocatechin (EGC; 20%), epicatechin-3-gallate (ECG; 14%) and epicatechin (EC; 6%). Among them, EGCG has the most health benefits, as it is effective in maintaining cardiovascular health and metabolism. In green tea, the amount of catechins varies, although a standard extract has been acquired for its use in supplementation [31]. Catechins cannot be completely extracted from fresh green tea leaves; therefore, there are large differences in the extract concentrations obtained. In addition, catechins are relatively unstable and can vary in amount under different conditions. Therefore, it is not possible to estimate the doses of catechins used in animal studies due to their lack of quantification [10,31,34].

The metabolic reactions for all catechins follow the same pathways of phase II detoxification reactions. Animal studies show that catechin uptake occurs in the intestine and

liver [35]. Catechins are partially bound to intestinal mucosa, liver, and kidneys, and about 5% of catechins are transported freely (i.e., unbound) through the bloodstream. For optimal biochemical functioning, catechins must attach to certain biochemical components (e.g., glutathione via liver enzymes) to improve their water solubility and facilitate their excretion in bile and urine. Thus, large amounts of catechins (including bile-attached catechins) are not absorbed in the small intestine and are transported to the large intestine, where they are broken down to smaller metabolites by resident microbes and reabsorbed for excretion via the urine as valerolactones [36]. Catechins were initially thought to have low viability of about 8–9% and very high detoxification [37]. However, studies have observed that the bioavailability of catechins increases by about 40% due to antimicrobial activity [38,39], where catechins were present up to 48 h after ingestion in human urinary excretion. This increase in bioavailability of catechins is explained by the conversion of larger compounds (EGCG and ECG) to smaller compounds (such as UDP-glucuronosyltransferases and sulphotransferases) via the phase II detoxification reaction.

### 5.2. Effectiveness and Life Span of Green Tea Catechins

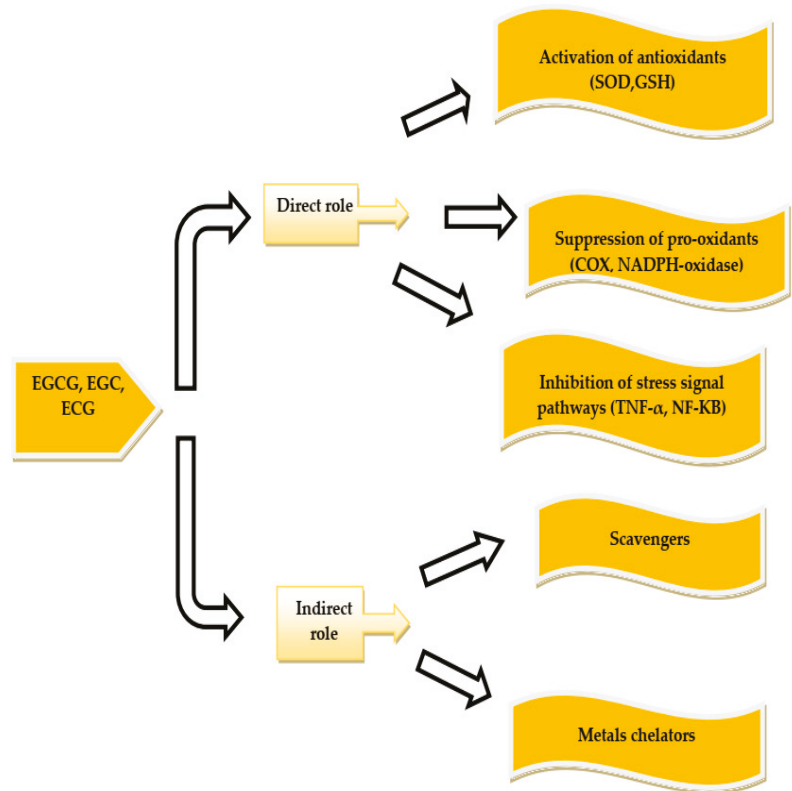
Pharmacological research demonstrates that green tea catechins have an effectiveness in the range of 2–13% in mice [40]. Some properties that can contribute to the oral bioavailability of green tea include its low solubility in gastrointestinal fluid, low permeability of the membrane, its degradation and metabolism in the gastrointestinal tract, and its transmission through intestinal epithelial membrane. Green tea catechins are stable at pH <6.5, but EGC and EGCG are rapidly degraded at pH >7.4. The concentration of polyphenols is higher in the fasted state than at postprandial [40]. Isbrucker et al. found that 20 mg·kg<sup>-1</sup> body mass per day of EGCG had no side effects in animals [41]. Most animal studies have used 0.1% green tea extract, equivalent to 10 mg per 100 mL of dissolved water, because it showed effectiveness with the lower amount [41].

Gallate-type catechins (e.g., EGCG and ECG) have a longer life span than nongalactic catechins (EGC and EC) because they are bound to proteins, which prevent premature excretion. The short half-life of nongalactic catechins occurs when it crosses the intestinal wall. The lasting effect of the maximum serum level of green tea catechins is about one hour in the fasted state and about two hours in the satiated state [42]. Green tea catechins are shown to be more stable at low pH and in the fasted state [43]. Alimentation raises the pH of the gut, a condition that destroys catechins.

### 5.3. Antioxidant Function Green Tea

Green tea is a rich source of antioxidants. Structural properties of green tea catechins play a role in their antioxidant function. The presence or absence of the galvanic portion and the number and position of the –OH groups on the ring define their structural property. The presence of the carboxyl group enables catechins to interact with the biological substance and gives its antioxidant function by binding to hydrogen or transferring electrons and hydrogen, or both [44]. The antioxidant activity of tea appears to inhibit lipid-inhibiting alkoxyl and peroxy radicals [45]. For example, the antioxidant activity of green tea extract is due to the increase in superoxide dismutase activity and catalase expression. These enzymes protect cells against oxidative stress [46]. The proposed mechanism for this process is the reduction of nitric oxide concentration in the plasma, which directly lowers the activity of oxygen species [13,47]. Malondialdehyde, a marker of oxidative stress, also decreases after consuming green tea [13,47]. Elevated levels of Phase II antioxidant enzymes have been observed after drinking green tea by increasing levels of polyphenols in the small intestine, lungs, and skin of mice [48] and rat prostate [49] and the oral cavity of hamsters [50]. The underlying mechanism of this process can be through the activation of MAPKs by green tea polyphenols [51]. Activation of the Nrf2 signaling pathway is also involved in this process [52]. Stimulation of phase II enzymes not only neutralizes free radicals and oxidative stress but can also have a detoxifying effect on carcinogens, such as aflatoxin B1 [52]. Studies have shown that catechins have a direct (antioxidant) or indirect

(increased activity or expression) effect on oxidative stress. The following Figure 1 shows the effect of green tea catechins on oxidative stress [45].



**Figure 1.** Mechanism regarding the effect of green tea catechins on oxidative stress.

#### 5.4. Green Tea Used as an Ergogenic Aid

Green tea is an ergogenic supplement that optimizes mitochondrial function and increases lipolysis and fatty acid metabolism, thereby reducing fatigue and improving endurance performance. After a Japanese study examined positive effects of catechins in rats [45], there was an increase in investigations regarding the effects of green tea catechins in animal models [45,46]. One study observed that mice who exercised with consumption of green tea extract (GTE; 0.2% and 0.5% supplementation groups) improved exercise performance compared to the exercise only and control groups, suggesting that the presence of catechin EGCG stimulated lipid metabolism [46].

#### 6. Sport Performance and Drinking Green Tea

In recent years, many studies have studied the effect of polyphenols, especially catechins, on athletic performance. Most of these studies have examined the effect of catechins on exercise-induced muscle damage and their biological and physiological role in improving physical function. Until a few years ago, as catechins are antioxidants, their role in preventing muscle damage was studied [47]. However, in recent years, more studies have significantly examined the effect of polyphenols on athletic performance. Studies on polyphenols and exercise include antioxidant supplements, such as green tea extract (GTE). Although the majority of studies on green tea have been done on animals, lately, a

large number of studies have been done on human athletic performance. Green tea extract contains a large amount of catechins that increase daily energy consumption in humans. For example, short-term consumption of green tea extract on healthy untrained men increased the amount of energy available during 30 min of cycling at 60% of the maximum oxygen consumption. However, the chemical process of green tea and green tea extract in the face of oxidative stress remains unknown. It has been reported that the inhibitory capacity of catechins is due to the presence of a hydroxyl group at the 5 prim position, which increases their ability to inhibit free radicals [47].

### 7. Production ROS during Exercise Training and Its Consequences

There are physiological factors that cause oxidative stress, but the effect of exercise on oxidative balance is difficult because it depends on many factors such as sex, age, and exercise status as well as intensity, duration, and frequency of exercise. Aging and exercise are commonly known as oxidative stressors [48,49]. Research has shown that metabolism, oxygen consumption, and production of active species due to intense exercise are 20 times higher than at rest [50]. Muscle contractions promotes the production of oxidants, which has importance in physiological function of the body. However, prolonged or intense exercise can lead to oxidative damage to cell compounds, and exercise-induced oxidative stress has been shown to affect muscle function [50]. Figure 2 illustrates the effect of exercise on the production of reactive active species.

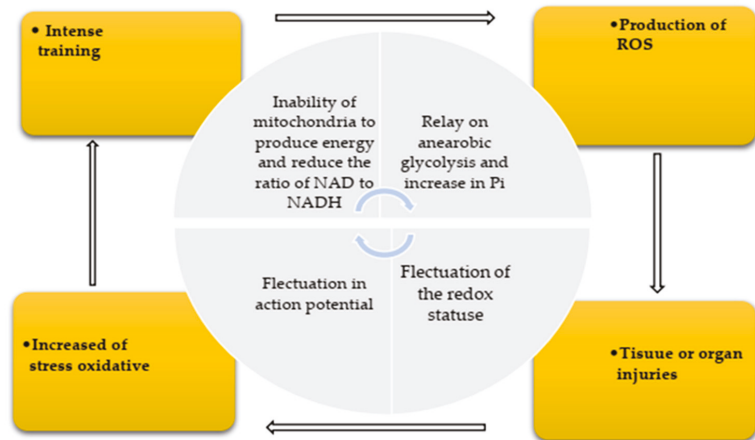


Figure 2. Production of reactive oxygen species (ROS) during exercise training.

Numerous studies have shown an increase in oxidative stress factors after chronic aerobic and strenuous exercise [50,51]. Exhausting strenuous exercise provides a good model for assessing the effects of oxidative stress on the body [50]. It is well known that intense and prolonged exercise elicits both lipid peroxidation and muscle damage rather than increasing the capacity of antioxidants [50]. However, some studies have observed that endurance training has no significant effect on inflammatory markers [52]. Nevertheless, research studies have shown that intense, moderate, and long-term aerobic exercise produce free radicals and oxidative stress [52]. Free radicals cause damage to lipids, proteins, carbohydrates, and DNA, among others [53]. Under normal conditions, about 2–5% of oxygen is converted to free radicals by organs.

### 8. Green Tea and Prevention of Oxidative Stress Production during Exercise

In general, the body can neutralize exercise-induced oxidative stress through antioxidant defense [20], but this defense can become overwhelmed by exercise-induced ROS

production. The oxidative stress produced in the body is neutralized through internal and external antioxidant systems. Because intense, irregular, and prolonged PA increases energy requirements in body tissues, especially in active muscles, oxygen consumption is greater and enzymatic antioxidant defense systems cannot cope with the higher production of free radicals alone [54]. Thus, nonenzymatic antioxidant system, in the form of natural and oral supplements, can help combat oxidative stress [54]. Consumption of antioxidants, such as green tea polyphenols and catechins, improves the body's antioxidant status and reduces the damaging effects of radicals during exercise, although these supplements do not appear to have a direct beneficial effect on performance [55,56].

Alessio was the first to examine the effects of green tea catechins on exercise-induced oxidative stress [57]. Five to six weeks of green tea consumption increased the athlete's antioxidant capacity, which in turn increased the level of total antioxidants in plasma during and after an intense running exercise session as well as prevented or inhibited excessive lipid peroxidation caused by the exercise. Several studies have shown that green tea has a protective effect (i.e., increased plasma antioxidant and decreased plasma lipid hydroperoxide levels) against oxidative stress produced during and after exercise [58,59]. In addition, green tea consumption inhibits or reduces plasma creatine kinase and xanthine oxidase activities after exercise. Numerous studies have shown that long-term use of antioxidant supplements lowers the production of free radicals and can diminish or even inhibit signaling from reactive species that are harmful to the body [60].

On the other hand, research has shown that exercise activates the redox-sensitive transcription factor NF- $\kappa$ B, which retranscribes the expression of antioxidant factor genes [50]. NF- $\kappa$ B is the main stimulus in inflammation and the main element of the innate immune response [45]. In addition, it plays an essential role in adaptive immune responses and regulates embryonic development, lymphopoiesis, and osteogenesis [61]. NF- $\kappa$ B is a redox-sensitive transcription factor that is overproduced and proposed to be involved in the regulation of cellular activity, including inflammation, immune response, growth, and cell death [61]. NF- $\kappa$ B remains in the cytoplasm as long as the kappa B inhibitor (I $\kappa$ B) 2 is inactive, but it is transported to the nucleus when I $\kappa$ B is activated [61]. EGCG reduces NF- $\kappa$ B binding to the nucleus and reduces the expression of transcription factor P65, the NF- $\kappa$ B subunit, by TNF- $\alpha$  [31]. Exercise and muscle contractions stimulate the sarcoplasmic release of calcium, increase ROS, and activate numerous signaling cascades, such as mitogen-activated protein kinase (MAPK) [31]. Various cell samples have been shown to increase intracellular calcium, ROS accumulation, and MAPK activation to enable NF- $\kappa$ B, suggesting that exercise can also activate NF- $\kappa$ B [52]. Acute treadmill exercise increases I $\kappa$ B kinase  $\alpha$  and  $\beta$  (IKK $\alpha/\beta$ ) phosphorylation, nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor, alpha (I $\kappa$ B $\alpha$ ) phosphorylation, and NF- $\kappa$ B activity in rat skeletal muscle [39]. While I $\kappa$ B $\alpha$  phosphorylation level increases during exercise, nuclear-bound NF- $\kappa$ B activity peaks at postexercise intervals [7]. NF- $\kappa$ B activation is a local event in contracting muscle as this process can occur in the absence of exercise-induced systemic factors [60]. Drug inhibitors p38 and extracellular signal-regulated kinase 1/2 (ERK1/2) are able to slow IKK $\alpha/\beta$ -regulated muscle contraction by 39% and 35%, respectively and by 76% in combination [62]. Alieso et al. showed that green tea EGCG protects the kidneys against lipid peroxidation [57]. However, Pingitore et al. demonstrated that the use of antioxidant supplements can have negative effects on the normal function of athletes, where low levels of oxidative stress reduce the positive reactions associated with hormones because some free radicals play an important role in signaling processes related to cell function [20].

Tea polyphenols act as antioxidants by reducing or inhibiting ROS and nitrogen through accumulating active metal ions, and they may indirectly act as antioxidants through other mechanisms that inhibit enzymes. "Pro-oxidant" and induced antioxidant enzymes protect tissues, cells, and plasma compounds against oxidative damage (see Figure 3).

**Imbalance between oxidants and antioxidants (Disease or exercise training)**

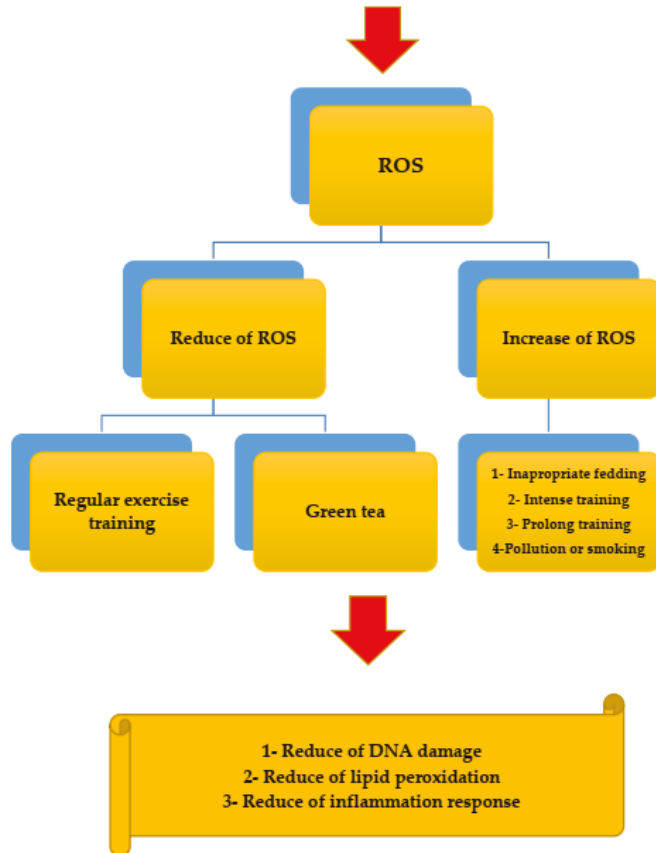


Figure 3. The relationship between green tea and reactive oxygen species (ROS).

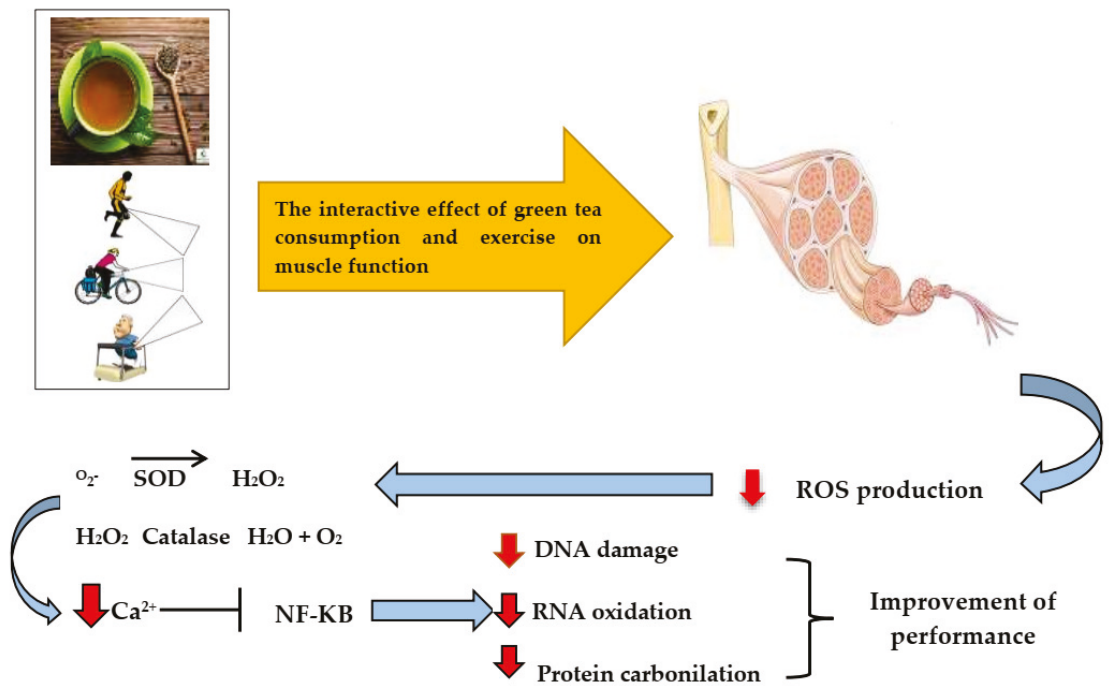
**9. How Can Green Tea Improve Sports Performance?**

The use of natural strategies and interventions to improve athletic performance, in addition to overall health, has attracted the attention of athletes at all levels of sport, and green tea consumption has particularly been demonstrated to be effective [9,36]. Meta-analysis studies have shown that green tea consumption improves performance without any side effects [25,34,36]. Further meta-analysis investigations on antioxidant effects and the presence of polyphenols in green tea have shown its beneficial effects on exercise performance [36,42]. There are many mechanisms for the effectiveness of polyphenols. Although it is not possible to address them all here, we will mention some of them [63]. Natural interventions, such as plant extracts and phytochemicals, enhance physical function, improve recovery after exercise, maintain overall health [35], and have minimal side effects [64,65]. In addition, regular green tea consumption (434 mL·day<sup>-1</sup>) was found to reduce body fat and decrease waist-to-pelvis ratio compared to the control group (i.e., no green tea ingestion). Similar to these findings, rodent models with high fat diet and body fat mass lost weight with green tea catechin consumption [66,67].

Green tea polyphenols and catechins also affect SIRT1, which has a role in increasing the activity of PGC-1α after deacetylation, consequently improving mitochondrial function [68,69]. Another action of green tea polyphenols and catechins is increased endothelial



NO synthesis and vasodilation [70,71]. The production of NO improves the perfusion of oxygen to the active muscles and improves athletic performance [72,73]. Thus, GTE supplementation can reduce oxidative stress through its polyphenols [74] and produce NO to improve maximum oxygen uptake, thus delaying fatigue [74]. Additionally, green tea consumption reduces muscle pain caused by improper exercise, bruising, and subsequent injuries. The reduction of exercise-induced fatigue because of GTE supplementation has great practical applications in sports performance in both amateur [75] and professional [52] athletes because it may be the limiting factor in achieving a personal record or successful performance. Oxidative stress not only accumulates during exercise [75] but also does so after exercise; thus, the consumption of green tea during and following exercise can lower oxidative stress (i.e., lipid peroxidation) [76]. Plasma triglyceride levels can indicate the effectiveness of catechins, which has been shown to be reduced with exercise in normal mice [77]. In addition, the consumption of green tea extract lowered triglyceride levels in Zucker mice and mice fed with a sucrose-rich diet. Studies have demonstrated that green tea flavonoids have an insulin-like activity and increase insulin activity [77]. The Figure 4 shows the effect of exercise combined with green tea consumption on exercise performance.



**Figure 4.** Possible mechanisms related to the effects of green tea catechins and exercise training on performance. Exercise and concomitant consumption of green tea reduce oxidative factors and suppress the activation of inflammatory factors by regulating calcium release, reduce DNA and RNA damage, release caspases due to TRX activation, and ultimately lead to improved performance.

Ichinose et al. examined the effect of drinking green tea with moderate-intensity exercise and found that concomitant use of the two interventions increased metabolism of fat, which was the predominant fuel during exercise [78]. The predominance of the fat energy source reduced  $VO_2$  during exercise. This process resulted in a 10% reduction in post-workout oxygen consumption in both groups, which was consistent with other studies [79,80]. They found that exercise combined with drinking green tea resulted in

metabolic adaptation and reduced energy expenditure at the same intensity of exercise. This reduction in energy leads to a decrease in the amount of basal metabolism. They observed this mechanism by measuring the concentration of free fatty acids after exercise and reported that the level of free fatty acids was significantly higher immediately after exercise in trained mice. Beta-oxidation cycle enzymes are important during exercise and fatty acid oxidation [81]. Murase et al. [82] found that drinking tea with exercise improved the activity of beta-oxidation enzymes and improved mitochondrial enzymes. They also found that drinking tea with exercise delayed the onset of fatigue [82]. According to studies and observation of the interaction between exercise and nutritional interventions, new insights can be provided for the use of natural interventions during physical activity to improve athletic performance or maintain health.

One of the limitations of our review is that there is a wide range of oxidative stress factors that could not be addressed. Additionally, this review only mentioned few of the many effects of green tea on performance and health. The amount of green tea required to obtain the maximum benefits is also unclear. Moreover, it is not clear how much physiological oxidative stress production is beneficial and how much harms the body. Most of the research has been done on healthy and young people, and there have been few investigations examining oxidative stress factors associated with metabolic syndrome and other diseases. Therefore, more studies are needed to answer these questions.

## 10. Conclusions

Due to its many properties, green tea improves physical and physiological function of the body during exercise by diminishing oxidative stress; however, more research is still needed. Assuming that green tea improves antioxidant function in active muscles while performing strength and endurance training, further studies should examine different training models to better understand its benefits on athletic performance. Due to the fat burning, weight loss, and mitochondrial function of green tea, most studies of exercise protocols associated with green tea have used aerobic exercise protocols. Due to the properties of green tea in changing body composition, weight loss, maintaining lean body mass, reducing body fat, reducing waist circumference, and reducing body fat percentage, green tea consumption combined with resistance training exercises causes more changes in anthropometric characteristics compared to strength training alone. Therefore, much research is needed on the effectiveness of different exercise models with green tea supplementation in physiological contexts and signaling and molecular pathways.

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Brief Report

# Potential Improvement in Rehabilitation Quality of 2019 Novel Coronavirus by Isometric Training System; Is There “Muscle-Lung Cross-Talk”?

Hadi Nobari <sup>1,2,3,\*,†</sup>, Mohamad Fashi <sup>4,\*,†</sup>, Arezoo Eskandari <sup>5</sup>, Jorge Pérez-Gómez <sup>2</sup> and Katsuhiko Suzuki <sup>6,\*,†</sup>

<sup>1</sup> Department of Physical Education and Sports, University of Granada, 18010 Granada, Spain

<sup>2</sup> HEME Research Group, Faculty of Sport Sciences, University of Extremadura, 10003 Cáceres, Spain; jorgepg100@gmail.com

<sup>3</sup> Department of Exercise Physiology, Faculty of Sport Sciences, University of Isfahan, Isfahan 81746-7344, Iran

<sup>4</sup> Department of Biological Sciences in Sports, Faculty of Sports Science and Health, Shahid Beheshti University, Tehran 198396-3113, Iran

<sup>5</sup> Department of Exercise Physiology, Faculty of Physical Education and Sports Science, Tehran University, Tehran 141793-5840, Iran; A.eskandari\_1988@yahoo.com

<sup>6</sup> Faculty of Sport Sciences, Waseda University, Saitama 359-1192, Japan

\* Correspondence: hadi.nobari1@gmail.com (H.N.); m\_fashi@sbu.ac.ir (M.F.); katsu.suzu@waseda.jp (K.S.)

† These authors contributed equally to this study.

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**Abstract:** The novel Coronavirus Disease 2019 (COVID-19) crisis is now present in more than 200 countries. It started in December 2019 and has, so far, led to more than 149,470,968 cases, 3,152,121 deaths, and 127,133,013 survivors recovered by 28 April 2021. COVID-19 has a high morbidity, and mortality of 2%, on average, whereas most people are treated after a period of time. Some people who recover from COVID-19 are left with 20 to 30% decreased lung function. In this context, exercise focused on skeletal muscle with minimal lung involvement could potentially play an important role. Regular exercise protects against diseases associated with chronic low-grade systemic inflammation. This long-term effect of exercise may be ascribed to the anti-inflammatory response elicited by an acute bout of exercise, which is partly mediated by muscle-derived myokines. The isometric training system seems to have this feature, because this system is involved with the skeletal muscle as the target tissue. However, no studies have examined the effect of exercise on the treatment and recovery of COVID-19, and, more importantly, “muscle–lung cross-talk” as a mechanism for COVID-19 treatment. It is suggested that this theoretical construct be examined by researchers.

**Keywords:** COVID-19; immune response; chronic diseases; exercise; oxidative stress; anti-inflammatory treatment; fibroblast growth factor 21; cytokines; myokines

## Dear Editor

Coronavirus Disease 2019 (COVID-19) is a new infectious disease caused by the coronavirus 2 that causes extreme acute respiratory syndrome (SARS-CoV-2). The majority of COVID-19 patients have mild to moderate symptoms, with about 15% progressing to extreme pneumonia and about 5% developing acute respiratory distress syndrome. The COVID-19 crisis is now present in more than 200 countries. It started in December 2019 and has, so far, led to more than 149,470,968 cases, 3,152,121 deaths, and 127,133,013 survivors recovered by 28 April 2021. COVID-19 is too new to have enough information about it; therefore, it may become an unprecedented pandemic. It has also spread to many Asian, American, and European countries. Causing respiratory tract infection and fibrosis, COVID-19 could result in morbidity and mortality, especially in those with impaired immune systems or a lack of existing immunity to the new virus [1]. In severe cases of COVID-19 infection, the virus can enter the blood stream and infect endothelial and

other target cells in the kidneys, esophagus, bladder, ileum, heart tissues, and central nervous system; multiple organ failure associated with hyper activation of the immune system is observed. Patients with COVID-19 infection in critical condition often have high systemic inflammatory parameters, including levels of creatine protein and cytokines (e.g., interleukin 6 (IL-6), IL-8, tumor necrosis factor alpha (TNF- $\alpha$ ), and etc.) [2]. In general, cytokine formation, inflammation, cell death, and other pathophysiological processes are linked to respiratory viral infections, which may be linked to a redox imbalance or oxidative stress. NADPH oxidase-2 (NOX-2) is overexpressed in hospitalized COVID-19 patients, resulting in increased oxidative stress, according to Violi et al. Other authors have found that blocking NOX-2 enhances disease phenotypes in macrophages by reducing oxidative stress, which is consistent with these findings [3].

Endothelial cells can mobilize NOX proteins in response to pro-inflammatory cytokines (e.g., IL-1, IL-6, and TNF- $\alpha$ ) and other agonists [4]. This contributes to local oxidative stress, which leads to endothelial dysfunction. NOX proteins may be mobilized by endothelial cells, leading to local oxidative stress and, as a result, endothelial dysfunction [5]. This causes muscle damage, either directly or indirectly, through oxidative damage to biomolecules and the activation of pro- or anti-inflammatory cytokines [6]. There is, however, insufficient information on COVID-19 and exercise. Fever, heavy pneumonia, ribonucleic acid (RNA) aemia, as well as the occurrence of ground-glass opacities and acute cardiac injury are some of the markers associated with COVID-19. In patients with COVID-19, substantial high blood levels of cytokines and chemokines (chemokine ligand (CCL) 2 and CCL5 and their receptors (CCR) 2 and CCR5) have been found. Chemokines have biological effects, such as inducing host immune cells to travel to the site of infection, controlling lymphocyte and other leukocyte transmission via peripheral lymph tissues, and promoting the growth of non-lymphatic organs [7]. The “cytokine storm” causes a pro-inflammatory climate, which is linked to significant tissue damage and contributes to COVID-19 patients’ fatal outcomes. The relation between inflammation and oxidative stress has been well established [8]. Nuclear factor-kappa B (NF- $\kappa$ B) and Toll-like receptor 4 (TLR4) signaling pathways, which are primarily triggered by viral pathogens such as SARS-CoV-2, can intensify the host inflammatory response, eventually leading to acute lung injury. TNF- $\alpha$  and IL-6, which are formed by skeletal muscle, T-cells, macrophages, and natural killer cells, are elevated as part of this response [9]. This disease, which is followed by a cytokine storm, affects a variety of tissues, most notably the lungs, resulting in acute lung injury of varying degrees. The cause of some patients’ rapid progression from acute respiratory distress syndrome to septic shock, accompanied by multiple organ failure and death, is cytokine storm. Acute lung injury and sepsis-induced multiple organ failure, two of the most common causes of morbidity and mortality in serious illness, have also been linked to oxidative stress. As a result, oxidative stress has long been a promising therapeutic target in serious illness, and antioxidants have long been studied in critically ill patients. Reactive oxygen species (ROS) are important mediators of the inflammatory responses, and different levels of exercise have mediate effects on ROS development. The inside membrane of the mitochondria is a source of ROS. Different levels of oxidative stress are caused by different levels of severity, volume of training, and exposure to a stressful setting [6]. Hence, antioxidants have anti-inflammatory properties and can be useful in the treatment of cytokine storm [10]. In addition, the transcription factor nuclear factor erythroid 2-related factor 2 (Nrf2) is responsible for the adaptation of cells under electrophilic or oxidative stresses [11]. Under normal conditions, Nrf2 is located in the cytoplasm bound to its inhibitor Keap1, which targets Nrf2 for ubiquitination and subsequent degradation. In the presence of electrophiles or ROS, the Keap1-Nrf2 complex dissociates and Nrf2 migrates to the nucleus, where it stimulates transcription of the target genes with antioxidant response element sequences in their promoters [11,12].

Exercise’s anti-inflammatory effects have been studied through three different mechanisms: (1) a decrease in visceral fat mass; (2) increased synthesis and release of anti-inflammatory cytokines (e.g., interleukin 1 receptor antagonist (IL-1ra), interleukin 4 (IL-4),

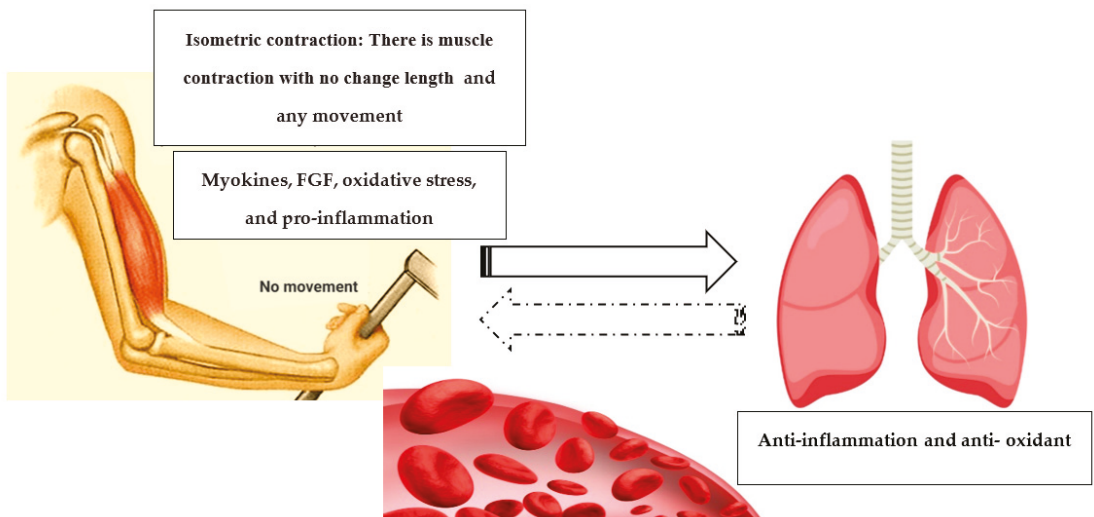
interleukin 10 (IL-10), interleukin 11 (IL-11), and interleukin 13 (IL-13)), by contracting skeletal muscle, such as myokines; and (3) TLR expression on monocytes and macrophages is reduced as a result [13]. In addition, exercise is a way in which it is possible to induce Nrf2 migrates to the nucleus, and this stimulates antioxidant response element and pro/anti-inflammatory balance [14].

Several studies have demonstrated that physical fitness and moderate-intensity training also have reverse correlations with risks of disease and premature fatality [15,16], which can be that immune response function and improved types of immune markers can be increased by exercise in some diseases. These diseases can include low-grade inflammation, hypertension, stroke, osteoporosis, cancer (e.g., colon, lung, stroke, and breast), chronic infectious disease, metabolic syndrome, cardiovascular disease, type 2 diabetes mellitus, cognitive impairment and obesity [6,17,18].

It is safe to exercise during the COVID-19 epidemic in healthy individuals, taking the necessary precautions, such as exercising at home. After COVID-19 disease, due to the spread of infection, the patients will not be able to exercise [16,19]. COVID-19 mortality is on average 2%, whereas most people are treated after a specified period. Some people who recover from COVID-19 are left with 20 to 30% less lung function, and gasping for breath when they walk quickly. In this context, exercise could potentially play an important role during recovery from COVID-19. Due to respiratory tract infection, performing aerobic exercises for recovered patients will be extremely difficult. Therefore, exercise focused on skeletal muscle with minimal lung involvement should be a priority [20]. The isometric training system seems to have this feature. Isometric training is used in the rehabilitation and physical preparation of athletes, patients, and the general public. Isometric training refers to muscle contraction where the muscle-tendon unit remains at a constant length. An example of this is pushing against an immovable object such as a wall. Suitable training using this system consists of 5–10 repetitions of 5 s per contraction, 5 days a week. Isometric contractions have a number of benefits, according to some sources; (i) in rehabilitative environments, isometric training allows for a precise application of force within pain-free joint angles; (ii) since the maximal isometric force is greater than that of concentric contractions, isometric training can help to trigger force overload which is associated with neuromuscular adaptations; and (iii) a professional who is familiar with a sport's physical demands will be able to use isometric training to target particular weak points in a range of motion, which can help with success and injury prevention [21–23]. By altering excitatory and inhibitory functions in the corticomotor pathways, isometric contractions may also provide an acute analgesic effect and allow for painless dynamic loading. Isometric contractions are also a highly accurate way to measure and monitor changes in force production. The isometric training method appears to aid immune system resilience by reawakening antioxidant defenses such as the glutathione system. Without the involvement of other antioxidant systems, the glutathione system could provide a primary protection against ROS produced during the performed isometric system, preventing oxidative damage to cellular lipids [21,24,25]. According to self-reported findings by four of the patients recovered from COVID-19, doing home isometric exercises has a great impact on the speed of return to normal life during post-illness recovery (Kashan, Esfahan, Iran: unpublished observation). The isometric training system exercises the skeletal muscle as the target tissue, where it can induce useful adaptations. These adaptations include, in part, the anti-inflammatory and antioxidant effects of exercise and could play an important role during the pandemic. Myokines are cytokines or peptides synthesized and released by myocytes and immune cells in muscle tissue in response to muscular contractions [26]. Myokines are implicated in the autocrine regulation of metabolism in muscles as well as in the para/endocrine regulation of other tissues and organs including the adipose tissue, liver, and brain and lung through their receptors “muscle–lung cross-talk”. For example, decreased levels of irisin, a skeletal muscle cell-derived myokine, are related to reduced lung function [27]. Irisin secretion was found to be linked to physical activity in a previous study. Irisin could play a role in oxidant stress inhibition through an oxidative-restraint



pathway involving Nrf2, which leads to cell apoptosis [28]. Furthermore, by upregulating PPAR expression and suppressing inflammatory cytokine levels, fibroblast growth factor 21 (FGF-21) reduces pulmonary hypertension and suppressing inflammatory cytokine levels. FGF is a group of 18 polypeptides with biochemical properties that are structurally and functionally significant. To regulate glycolipid metabolism, enhance insulin resistance, prevent liver disease, and promote the browning of white adipose tissue, FGF-21 can enter the blood in autocrine, endocrine, and paracrine patterns [29]. Exercise can increase FGF-21 secretion and expression, effectively controlling brown adipose tissue activation and white adipose tissue browning to aid fat loss. These improvements can be linked to an increase in cardiorespiratory health [30,31]. This mechanism can be seen in Figure 1.



**Figure 1.** The relationship between isometric training system exercise and the release of myokines and FGF as anti-inflammatory effects that suppress inflammatory reactions (muscle–lung cross-talk). FGF: fibroblast growth factor.

The benefits of pulmonary rehabilitation are well known and the existing programs could be used as one of the rehabilitation referral pathways for COVID-19 survivors who present with symptoms and/or impairments in physical function. The main component of pulmonary rehabilitation programs is exercise training, which includes aerobic and/or resistance training, and these exercises have been demonstrated to decrease the negative effects that prolonged sedentary behavior and inactivity during a hospitalization period have on physical function [32]. The basic mechanism of these adaptations about COVID-19 is not fully understood. In addition, to date, no studies have examined the effect of exercise on the recovery of COVID-19, and more importantly “muscle–lung cross-talk” as a mechanism for COVID-19 rehabilitation. We encourage exercise physiology and immunology researchers to examine the theoretical constructs presented here.

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