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Full Body Photobiomodulation Therapy to Induce Faster Muscle Recovery in Water Polo Athletes: **Preliminary Results**

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Abstract

Objective: To investigate the recovery of male water polo athletes applying full body photobiomodulation therapy (PBMT) regarding inflammation and muscle damage markers, testosterone and cortisol hormonal responses, heart rate variability (HRV), maximal voluntary contraction, and squat jump (SJ) after official water polo matches.

Background: PBMT has been applied locally on specific muscle groups to induce faster recovery and improve the performance of athletes and nonathletes. However, many sports modalities require movement of the whole body, and a full body PBMT could be more adequate to irradiate large muscle areas homogeneously and faster.

Methods: In a randomized, parallel, and double-blinded design, 13 athletes (whole team) aged 18 years attended the study and were allocated into two groups: PBMT (dose of 6.9 J/cm², irradiance of 46.17 mW/cm², 5 min irradiation) and placebo treatment. The study was conducted during the 2019 Brazilian under 20 water polo championship. All athletes were assessed by blood samples and neuromuscular evaluation. Immediately after each match, all athletes received PBMT (effective or placebo).

Results: No significant interactions (raw values and percentage related to baseline) were observed for testosterone and cortisol, tumor necrosis factor-alpha, interleukin-6, creatine kinase concentration, maximal isometric voluntary contraction, SJ test, and HRV. Only an isolated interaction (decrease) was found for lactate dehydrogenase (LDH) response after the first match (p=0.004, post-hoc p=0.038).

Conclusions: The parameters of the full body PBMT of this study did not induce faster recovery of inflammatory, muscle damage (excepting LDH), testosterone, cortisol, HRV, and neuromuscular responses during repeated days of water polo matches.

Keywords: low-level laser therapy, light-emitting diode, inflammation, cortisol, testosterone

Introduction

N THE PAST two decades, photobiomodulation therapy ▲ (PBMT) at the near-infrared light spectrum emitted by low-level lasers and/or light-emitting diodes (LEDs) has emerged as a potential ergogenic therapy to accelerate muscle performance and recovery. 1-3 The justification for these effects comes from decades of publications based on animal models and randomized clinical trials (RCTs), which show many positive effects of PBMT on cell proliferation, metabolism, tissue repair, and attenuation of muscle damage and inflammation.³ As the central pillar of light-tissue interaction, cytochrome c oxidase in the mitochondrial respiratory chain can absorb photons of light that in turn stimulates increments in mitochondrial activity as respiration and synthesis of energy [adenosine triphosphate (ATP)], beyond stimulate muscle glycogen synthesis and increase antioxidant enzyme activity.³

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Although there seems to be a consensus about the treated area, that is, irradiate as much muscle area as possible, 1-3 some RCTs findings of exercise performance and muscle recovery are controversial even using the same device and/or similar parameters of PBMT. For instance, Baroni et al.⁵ and Vanin et al.⁶ disagree about the effectiveness of light energy of 30 J applied per site of irradiation on quadriceps muscle group on peak torque at maximal voluntary contraction (MVC) protocol and muscle damage marker [creatine kinase (CK)]. While Baroni et al.⁵ reported better results with 30 J per site of irradiation (or 180 J total energy), Vanin et al.⁶ reported the worst results (similar to placebo).

Moreover, the effects seen in RCTs¹⁻³ are modest compared with those seen in animal models.^{4,7} One possible reason is the unknown ideal dose of light, or ideal parameters of PBMT to be used in RCTs, despite some studies being optimistic in recommending a guideline for this purpose, even the literature points out controversial results as mentioned above. In this way, when using animal models, it is easier to irradiate a whole muscle group or body because the size and cross-sectional areas of animal muscles are significantly smaller than human muscles. Therefore, the use of a full body irradiation in humans can be an alternative to mimic or simulate the same conditions (size area) of muscle irradiation applied in animal models. Moreover, full body PBMT could help to better understand and translate the effects of this therapy in animal models^{4,7} to athletes submitted to the stress of exercise regarding the muscle recovery.

Besides PBMT purposed role in the recovery process, some immunoregulatory proteins are strongly associated with muscle damage and tissue repair. It is well documented that exercise training may impose an inflammatory milieu increasing of: tumor necrosis factor-alpha (TNF- α), interleukin-1 β (IL-1 β), and interleukin-6 (IL-6) in early inflammation stages, followed by elevation of anti-inflammatory biomarkers, such as transforming growth factor-beta (TGF- β), interleukin-10 (IL-10), and insulinlike growth factor 1 (IGF-1). All these proteins contribute to the process of new muscle fibers formation and regenerating damaged fibers restoring skeletal muscle tissue. 9,10 In this line, the inflammatory process is necessary to regulate the muscle adaptation front exercise; however, the relationship between PBMT and inflammatory responses as well as the influence of the PBMT in this scenario using full body PBMT is unclear.

Thus, the purpose of the present study was to investigate whether the use of full body PBMT applied on water polo athletes after official matches can induce faster muscle recovery [improve MVC, squat jump (SJ), and heart rate variability (HRV)]; increase testosterone; and decrease cortisol, inflammation (TNF-α and IL-6), and muscle damage markers [CK and lactate dehydrogenase (LDH)]. We hypothesized that full body PBMT would promote better muscle recovery and decrease inflammation and muscle damage markers compared with placebo therapy.

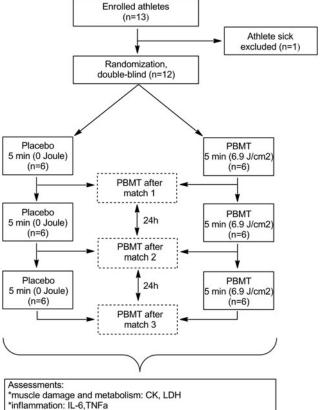
Methods

This randomized, double-blind, and placebo-controlled trial enrolled a whole team composed of 13 young male water polo athletes (18 \pm 1 years, 85.2 \pm 14.4 kg of body mass, and 182.6 ± 5.3 cm of height), with at least 4 years of practice. This study design was chosen to avoid any possible interference of previous use of PBMT on placebo condition.

Athletes were allocated randomly and secretly into two groups, PBMT and placebo, and according to their technical level (reported by coach) and position (parallel design) (Fig. 1). The study was conducted during the 2019 Brazilian under 20 water polo Championship performed for 5 days (Wednesday to Sunday), and each team played three matches. All procedures were conducted according to the Declaration of Helsinki and approved by the local ethics committee (approved number 1.139.070).

The sample size was calculated based on the necessary number of athletes to obtain significant differences between both groups (PBMT and placebo) regarding CK. A possible large effect (Cohen's d of 1.1) of full body irradiation was considered on CK assessment among four repeated measures (baseline, match 1, 2, and 3). In addition, the statistical power was considered as 80% and alpha (α) of 5%. Thus, a total sample size calculated was 12 athletes. As the whole team had 13 athletes, all were enrolled in the present study.

Before each match (2–3 h) were measured the HRV in rest, followed by blood samples to analyze testosterone and cortisol; CK and LDH; TNF-α and IL-6; and MVC and SJ;



*performance: Jump, maximal voluntary isometric contraction *hormone: testosterone, cortisol, testosterone/cortisol ratio

Full-body photobiomodulation therapy (PBMT): Joovv Elite system *Irradiance: 46.17 mW/cm2

Time of irradiation: 150 s (front) plus 150 s (back) muscles

*Fluency or energy density: 6.9 J/cm2

Continuos mode

*Irradiation at 30 cm distance

FIG. 1. Study design. CK, creatine kinase; LDH, lactate dehydrogenase; IL-6, interleukin 6; TNF- α , tumor necrosis factor-alpha.

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as hormonal, muscle damage, inflammation, and neuro-muscular responses, respectively. These procedures were performed on the first day of the championship week (Monday) to set a baseline, and before each other three matches. These assessments were performed by investigators blinded to PBMT and allocation. Saturday, the team did not play any match and these procedures were not performed. Immediately after each match, the athletes were treated with PBMT or placebo using a full body irradiation device. The order of application of PBMT and placebo was counterbalanced every day, that is, the irradiation was alternated between each athlete of the group PBMT and each athlete of the placebo group until all athletes of both groups received their respective therapy.

Heart rate variability

The HRV was measured with RR intervals recorded during 6 min (data of the first minute were discarded from analysis), with athletes seated and monitored by V800 Polar monitor (Polar, Kempele, Finland), and the root mean square of the successive normal sinus RR interval difference (RMSSD) was determined using the Kubios software.

Blood collection and whole blood stimulation with lipopolysaccharide in vitro

Blood samples were collected from the antecubital vein using ethylenediaminetetraacetic acid vacutainer tubes (BD Vacutainer®) and divided into two tubes. Four milliliters of blood was incubated with lipopolysaccharide (Escherichia coli, type: 0111: B4; Sigma, St. Louis, MO), with a final concentration of 10 ng/mL (during 60 min at 37°C, with constant and slow rotation) to measure TNF- α and IL-6 [coefficient of variation (CV) = 4.9 and 3.1, respectively; R&D System, Minneapolis], while another 4 mL of blood was centrifuged to measure CK (CV=4.4%, Lote 11790; Biosystems, Spain) and LDH (CV=4.8%, Lote 11581; Biosystems, Spain) in random-access analyzer (A-15; Biosystems, Barcelona, Spain) using a specific filter for each analysis. Testosterone (CV=5.6%, Lote 37K4J8; Monobind) and cortisol (CV = 6.1%, Lote 36K1I8; Monobind) were measured by enzyme-linked immunosorbent assay using a microplate reader (SpectraMax Plus 384; Molecular Devices).

Maximal voluntary contractions

Each athlete performed three isometric MVC of the knee extensors muscles of the self-reported dominant leg, with rest interval of 1 min between each MVC. Each athlete remained seated in a specific chair with knee and hip flexed at 90°, and trunk and thighs fixed by a belt. 11 The force produced was recorded by a load cell (strain gauge sensibility: 2.0 ± 0.02 mV/V; maximum capacity: 100 kgf; MKControle, São Paulo, Brazil). The load cell data were acquired by an analog module (National Instruments, Austin) with a sampling rate of 1000 Hz and subsequently filtered with a second-order low-pass Butterworth filter with a cutoff frequency of 5Hz in MatLab 7.9 (MathWorks®). Peak force (Fpeak) was assumed as the mean around 50 ms (totaling 100 ms) during the highest force value. While the mean force (Fmean) was assumed as a mean over 100 ms during the force plateau.

Squat jump

Athletes performed three SJ interspaced by 1 min on a force plate with a sample acquisition of 600 Hz (CEFISE, Brazil). During the SJs, participants were instructed to keep hands on the waist (akimbo), flex the knees to approximately 90°, stop in this position and jump as quickly and high as possible falling with parallel legs. The peak force (Fpeak) was assumed as the highest value during the concentric phase. Height was measured by the sum of displacement in the contact phase and air phase. Work was assumed as a product of body mass, acceleration of gravity (9.81 m/sec²), and the total displacement of the subject during the jump. While the rate of force development (RFD50ms) was assumed as the difference between the highest value on 50 ms of the concentric phase and baseline (bodyweight) strain values divided by 50.

Full body PBMT and placebo

Full body PBMT was performed using a Joovv Elite system comprising six panels of 76 red $(660\pm10\,\mathrm{nm},\,80\,\mathrm{mW})$ each at 30 cm of distance) and 74 infrared $(850\pm10\,\mathrm{nm},\,65\,\mathrm{mW})$ each at 30 cm of distance) LEDs, totaling 900 LEDs distributed over an area of $12,193\,\mathrm{cm}^2$ ($1.2193\,\mathrm{m}^2$). The athletes were positioned 30 cm far from the device, and the irradiation time in continuous mode (without a pulse) corresponded to 5 min (2'30" to front plus 2'30" back). During all PBMT irradiation or placebo conditions, all athletes were wearing only water polo trunks (Fig. 2). All PBMT or placebo interventions were applied in a darkened room.

In the front position, all athletes performed an external rotation of the hip joint to expose adductor muscles to irradiation since this muscle group is heavily required to swim.¹³ All athletes were blindfolded to irradiation and wore headphones (hearing a standard song) during both therapies (effective or placebo).

PBMT effective dose applied was 6.9 J/cm² based on previous studies,^{4,7} with an irradiance of 46.17 mW/cm².

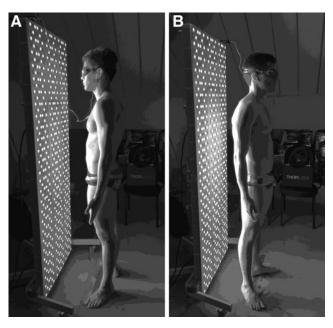


FIG. 2. Full body photobiomodulation therapy applied in front **(A)** and back **(B)** muscles of water polo athletes.

TABLE 1. IRRADIATION PARAMETERS (UNITS)

Manufacturer	Joovv		
Wavelength	Red	Infrared	
Center wavelength (nm)	660±10	850±10	
Number of LEDs (6 panels)	456	444	
Beam area (cm ²) each LED	2.54	2.54	
Operating mode	Continuous	Continuous	
Distance from the body (cm)	30	30	
Radiant power (mW)	80	65	
Power density (mW/cm ²)	25.47	20.70	
Time of irradiation front (sec)	150	150	
Time of irradiation back (sec)	150	150	
Energy density (J/cm ²)	3.82	3.10	
Total average radiant power (mW)	(80+6)	5)=145	
Total average power density (mW/cm ²)	(25.47 + 20)	.70) = 46.17	
Total average energy density (J/cm ²)	(3.8+3.1)=6.9		
Total area of six panels (cm ²)	12,193		
Distance from power meter (cm)	30	30	
Area sensor power meter (cm ²)	3.14	3.14	

LED, light-emitting diode.

Placebo (PLA) condition consisted of fake irradiation with the device turned-off. It is important to highlight that an investigator blinded to all data collection and analysis performed all irradiations. The order of treatments was chosen through simple balanced randomization. All PBMT parameters were measured previously by a power and energy meter (PM100D; Thorlabs, Inc.) equipped with light sensor S310C (area of 3.14 cm²) and are described in Table 1.

Statistical analyses

After checking the normality of data by the Shapiro–Wilk test, the raw and percentage of different data from PBMT and placebo were analyzed using a two-way repeated-measures analysis of variance with SIDAK post-hoc if necessary, assuming p < 0.05.

Results

Initially, the team investigated won all three matches played during the competition. The HRV RMSSD measured at baseline and before each match showed no difference between therapies evidenced by nonsignificant interaction (p=0.61) (Fig. 3). In addition, no significant interaction (p=0.34) was observed for RMSSD expressed in percentage values from the baseline moment.

Table 2 shows the data of (raw and delta percentage) CK, LDH, testosterone, cortisol, testosterone/cortisol ratio, IL-6, and TNF- α results. For absolute values, no significant interaction was observed for any variable (p > 0.11). A significant interaction was only found for LDH delta percentage with lower values in the PBMT compared with the placebo group 24h after the match 1 (p = 0.004, post hoc p = 0.038).

Table 3 shows the results of the jump test and MVC (raw and delta percentage data). No significant interaction was observed for any variable obtained by the jump test and MVC (p > 0.25). Finally, intention-to-treat analysis was applied in this study.

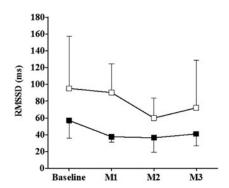
Discussion

The current study is the first in the literature to investigate the use of full body PBMT and applied this therapy to athletes during an official championship. However, the findings of the current study demonstrated that full body PBMT was not effective to produce faster recovery regarding neuromuscular, HRV RMSSD, inflammatory, CK, testosterone, and cortisol responses during repeated days after official water polo matches using the parameters of light reported in this study. Only an isolated positive effect between the groups was found for LDH after the first irradiation (i.e., day of match 2).

In the last few years, several studies have reported beneficial effects of PBMT on recovery and performance; however, the major of these findings were reported in animals models or in isolate muscle efforts, such as irradiations on quadriceps femoris muscles submitted to knee flexion/extension in isokinetic exercise protocols, or irradiation on biceps brachii submitted to elbow flexion/extension in isotonic or isometric exercise protocols. ^{1–3}

The mechanisms of action of PBMT to improve exercise performance and muscle recovery have extensively been discussed in the literature, which reports improvement in mitochondrial metabolism and ATP synthesis; oxidative stress defense through upregulation of antioxidant enzyme activity as superoxide dismutase, glutathione peroxidase, and catalase; prevention of muscle damage measures by CK levels in bloodstream; modulation of inflammatory process in muscle damage; and gene expression modulation through upregulation of protein synthesis (hypertrophy) and downregulation of

FIG. 3. Root mean square of the successive differences of heart rate variability at rest. Raw values and percentage difference from baseline. M1, before first match; M2, before second match; M3, before third match; PBMT, photobiomodulation therapy; PLA, placebo.



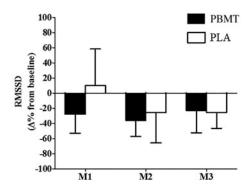


TABLE 2. BLOOD MARKERS FOR PBMT AND PLACEBO CONDITIONS MEASURED AT BASELINE AND BEFORE EACH MATCH

				PBMT							Placebo			
	Baseline	Before M1	7/2	Baseline Before M1 $\Delta\%$ Before M2	%∇	Before M3	7%	Baseline	Before M1	%∇	Before M2	%∇	Before M3	$\Delta\%$
CK (U/L) LDH (U/L) Testosterone	289.0±294.9 403.0±124.9 4.3±1.5	474.6±560.1 320.0±43.7 4.2±1.7	55.3 ± 40.8 -16.6 ± 19.4 19.2 ± 90.9	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$51.8 \pm 56.5 \\ -37.3 \pm 16^{a} \\ 35.6 \pm 80.3$	470.9±577.5 309.4±63.8 5.0±0.1	49.6 ± 44.2 -20.0 ± 17.3 37.5 ± 81.5	287.0±98.6 373.3±88.9 4.9±0.2	382.5 ± 127.2 283.3 ± 44.1 5.0 ± 0.2	42.8 ± 75.4 -20.4 ± 16.9 2.5 ± 4.6	464.3 ± 202.9 302.7 ± 32.0 4.9 ± 0.2	74.5±76.2 -14.7±1.0 1.5±5.8	364.8 ± 157.4 286.6 ± 42.5 4.9 ± 0.2	32.2 ± 41.6 -19.9 ± 9.4 0.9 ± 5.0
$(\eta g/mL)$ Cortisol $(\eta g/mL)$ Testosterone/	10.1 ± 7.4 0.7 ± 0.5	$16.7 \pm 7.2 \\ 0.3 \pm 0.0$	$131.1 \pm 182.1 \qquad 19.1 \pm 1.1 \\ -20.9 \pm 85.0 \qquad 0.3 \pm 0.0$		$188.6 \pm 171.4 \\ -17.7 \pm 89.2$	19.6 ± 0.4 0.3 ± 0.0	$191.4 \pm 169.1 \\ -19.0 \pm 88.0$	$11.1 \pm 6.8 \\ 0.7 \pm 0.5$	19.9 ± 0.8 0.3 ± 0.0	169.0 ± 182.8 -43.5 ± 34.5	$14.6 \pm 7.2 \\ 0.5 \pm 0.4$	$101.9 \pm 191.3 \\ 7.7 \pm 107.0$	20.5 ± 0.3 0.2 ± 0.0	177.6 ± 189.5 -46.6 ± 31.5
Corusoi ratio IL-6 (pg/mL) TNF-α (pg/mL)	9.9±5.3 192.2±78.2	6.6 ± 3.9 127.7 ± 83.8	-18.4 ± 67.4 -37.9 ± 31.7	6.5 ± 5.0 205.7 ± 116.9	-9.7 ± 104.7 -1.8 ± 40.4	5.5 ± 4.0 278.4±148.7	-33.2 ± 62.8 41.4 ± 49.2	20.4 ± 18.3 364.5 ± 131.8	13.2 ± 15.5 82.0 ± 42.1	55.6 ± 244.7 -74.0 ± 18.0	24.7 ± 43.2 177.0 ± 102.0	8.2 ± 104.8 -48.1 ± 25.3	10.3 ± 10.4 370.1 ± 154.2	-42.4 ± 36.0 0.8 ± 31.7

Values are expressed as mean±SD.

^aCompared with the same moment at the placebo group.

CK, creatine kinase; LDH, lactate dehydrogenase; IL-6, interleukin-6; M1, first match; M2, second match; M3, third match; Δ%, percentage difference from baseline; PBMT, photobiomodulation therapy; TNF-α, tumor necrosis factoralpha.

Table 3. Maximal Isometric Voluntary Knee Extensor and Jump Test Outcomes for PBMT and Placebo Conditions Measured at Baseline and Before Each Match

Ì		2.1		0.5 0.7 1.8
	∇	-7.0 ± 12.1 -13.0 ± 17.0	$\Delta\%$	-24.7±10.5 -3.9±9.7 -4.0±9.4 -12.9±21.8
	Before M3	60.4±11.6 44.5±8.3	Before M3	2272.3±486.4 16.0±2.0 133.4±18.7 7.9±2.6
	%∇	-2.4 ± 10.4 -5.0 ± 15.2	%∇	-21.6±9.3 -5.6±5.8 -6.3±7.3 -3.5±31.7
Placebo	Before M2	63.4 ± 10.7 48.6 ± 8.0	Before M2	2264.3±282.4 15.8±2.2 132.1±14.9 9.1±2.4
	%∇	-3.2 ± 9.6 -5.6 ± 9.7	%∇	-9.7±7.4 -1.7±6.6 -2.0±5.7 7.8±53.8
	Before MI	62.8±9.9 48.7±8.4	Before M1	2621.1±409.8 16.4±2.2 141.0±14.8 7.9±2.4
	Baseline	64.9±8.4 51.6±7.4	Baseline	2969.3±577.6 16.8±2.2 143.9±21.5 9.7±3.7
	%∇	-8.8 ± 3.9 -10.6 ± 6.4	$\Delta\%$	$-22.4 \pm 16.4 \\ 0.0 \pm 4.7 \\ 2.1 \pm 5.9 \\ -35.4 \pm 12.1$
	Before M3	57.3±9.2 44.6±6.4	Before M3	2202.3±890.2 15.8±1.0 134.6±32.2 6.9±2.4
	%∇	-4.2 ± 2.7 -7.9 ± 10.5	%∇	-20.8±9.5 1.3±9.4 1.8±9.3 -18.2±23.6
PBMT	Before M2	60.3 ± 9.8 46.4 ± 10.1	Before M2	∞
	%∇	-7.7 ± 2.8 -12.5 ± 17	%∇	-2.0 ± 4.0 1.5 ± 6.5 1.1 ± 7.6 -32.4 ± 27.2
	Before M1	57.8±7.1 43.8±9.9	Before M1	2720.7±743.8 16.1±1.1 133.4±33.2 7.5±3.6
	Baseline	62.8±8.8 50.3±9.4	Baseline	2783.6±804.9 15.8±1.2 131.7±29.3 10.5±2.9
	MVC	Fpeak (kg) Fmean (kg)	CMJ	Fpeak (N) Height (cm) Work (J) RFD50 msec (Nkg/sec)

Values are expressed as mean±SD.

2%, percentage difference from baseline; Fpeak, peak of force; Fmean, mean force; RFD50ms, rate of force; M1, first match; M2, second match; M3, third match; MVC, maximal voluntary contraction; SJ, squat jump.

protein degradation (atrophy) signaling.^{1–3,14} All these effects seem to be dependent on the light dose applied over the muscle tissue (dose response) reported in a therapeutic window around 60–300 J for large muscles^{1,2} when PBMT is applied before or after the exercise. In addition, when applied before the exercise, how many minutes and hours before the exercise seem to also be very important.^{7,15}

Regarding PBMT and muscle performance and recovery in sports involving swimming, Zagatto et al.¹³ were the pioneer in investigating the effect of repeated PBMT by a laser probe applied over adductor muscles of lower limbs (48 J of total energy) on cytokines, muscle damage marker, and performance during 5 days of water polo training. Nevertheless, Zagatto et al.¹³ reported small positive effects of PBMT on inflammation and muscle damage markers, and moderate effect on performance, likely anticipating the inflammation and recovery processes. Moreover, these authors highlighted the need of recovery strategies for high training loads quantified by session-rating of perceived exertion during an intensified training week ranging from 600 to 1300 a.u. per day in water polo athletes, such as in championships involving swim modality. However, a huge limitation reported in that study was the small area covered by PBMT using a laser probe, which was solved in the present study (full body irradiation). Although using full body irradiation, the PBMT was not effective to enhance recovery and/or performance. These results might be affected by not achieving the large effect expected on reduction of CK in bloodstream, and then, the sample size was small to the study, although a whole team was enrolled during a championship (application in real world).

Another possible reason for the no significant effects of full body PBMT is the light dose and parameters used, as reported in previous review about the known biphasic doseresponse of PBMT¹⁶ and also addressed recently. The in this context, the present study applied 6.9 J/cm² similarly to previous animal studies (7.2 J/cm²) that showed increased muscle performance and recovery, the but this dose did not produce significant results in the present study, demonstrating a lack in the effectiveness of translation of the PBMT dose from animal model to clinical trial.

Taking a deep discussion about PBMT dose and considering an average of 2 m² (20,000 cm²) of the body area for each athlete, a total energy of 138,000 J were applied over the body surface, that is, 69,000 J/m² or 6.9 J/cm² in front and back. This amount of energy may be too much compared with previous studies^{1,2} that identified a therapeutic window for large muscles such as quadriceps femoris muscles around 60–300 J. Regarding the light dose, 6.9 J/cm² may also be considered too much if compared with previous studies^{1,2} that applied 60-300 J over quadriceps femoris muscles that have $\sim 1000 \,\mathrm{cm}^2$ of surface area [a thigh of $25 \times 40 \,\mathrm{cm}$ (width × length)] resulting in 0.2 J/cm² (if considered 200 J of total energy/1000 cm²). However, these studies^{1,2} did not apply the PBMT over the entire surface area of muscles uniformly. For this reason, we cannot assume indeed a dose of 0.2 J/cm² and suppose that 6.9 J/cm² was too much.

All studies until now have used laser probes with $\sim 0.03 \, \mathrm{cm}^2$, or cluster devices with $\sim 20{\text -}30 \, \mathrm{cm}^2$. Consequently, the energy density applied with laser probes generally varied from ~ 50 to $250 \, \mathrm{J/cm}^2$ (from 2 to 7 J per point of irradiation), and with cluster devices generally varied from 1 to $2 \, \mathrm{J/cm}^2$ (from 30 to $50 \, \mathrm{J}$ per site of irra-

diation).^{1,2} Unfortunately, we can see different dosimetry depending on the device used for PBMT, making it difficult to standardize an applicable light dose and energy to achieve positive results in muscle performance and recovery. Thus, it is worth raising an important discussion about energy and energy density (dose): Which one (energy or energy density) is the most important in muscle performance and recovery? What is the muscle area necessary to be irradiated by PBMT to promote better muscle performance/recovery in physical exercises?

Another important difference between the present study and previous clinical trials is the mode of irradiation at a distance, which evokes a light dispersion/reflection (not measured) that can decrease the light absorption by the body. ^{18–20} Thus, it is important to highlight the pioneering of the present study and the difficulty to compare our results with previous RCTs in this research field, mainly regarding the light dose and parameters applied in contact mode over small areas of muscle tissue irradiation compared with the present study. We suggest more investigation to establish ideal parameters, including power of the light, distance from the device, irradiance, time of irradiation, and dose for full body irradiation protocols.

As the practical application of the present study, the beneficial effects of PBMT still remain under investigation. Full body PBMT using the light parameters of the present study (mode of irradiation, dose, irradiance, and time of light exposure) was not beneficial to decrease inflammation markers and muscle damage and improve performance as maximal isometric voluntary contraction and SJ test. Only an isolated effect was found reducing LDH concentration. In addition, the use of full body PBMT did not result in harmful effects, but for their wide use, recommendation for further robust findings is needed. In this sense, a strength of the present study is to introduce for the first time the use of full body PBMT, which should be better investigated regarding its light parameters (distance from the panel, dose, irradiance, time of light exposure) effects during training programs¹⁴ or when applied as preconditioning therapy (irradiation of muscles before the exercise or matches) assessing also the time response to light. 7,15

Conclusions

The full body PBMT applied after water polo matches, using the light parameters of the present study (mode of irradiation, dose, irradiance, time of light exposure), did not induce a faster recovery of neuromuscular, inflammatory, CK, and testosterone and cortisol hormonal responses during repeated days of water polo championship, while an isolated positive effect between groups was found only for LDH (reduced muscle damage).

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Author Disclosure Statement

No competing financial interest.

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References

- Ferraresi C, Huang YY, Hamblin MR. Photobiomodulation in human muscle tissue: an advantage in sports performance? J Biophotonics 2016;9:1273–1299.
- Vanin AA, Verhagen E, Barboza SD, Costa LOP, Leal-Junior ECP. Photobiomodulation therapy for the improvement of muscular performance and reduction of muscular fatigue associated with exercise in healthy people: a systematic review and meta-analysis. Lasers Med Sci 2018;33: 181–214.
- 3. Ferraresi C, Hamblinc MR, Parizotto NA. Low-level laser (light) therapy (LLLT) on muscle tissue: performance, fatigue and repair benefited by the power of light. Photonics Lasers Med 2012;1:267–286.
- Ferraresi C, Parizotto NA, Pires de Sousa MV, et al. Light-emitting diode therapy in exercise-trained mice increases muscle performance, cytochrome c oxidase activity, ATP and cell proliferation. J Biophotonics 2015; 8:740–754.
- Baroni BM, Leal Junior EC, De Marchi T, Lopes AL, Salvador M, Vaz MA. Low level laser therapy before eccentric exercise reduces muscle damage markers in humans. Eur J Appl Physiol 2010;110:789–796.
- Vanin AA, De Marchi T, Tomazoni SS, et al. Pre-exercise infrared low-level laser therapy (810 nm) in skeletal muscle performance and postexercise recovery in humans, what is the optimal dose? A randomized, double-blind, placebocontrolled clinical trial. Photomed Laser Surg 2016;34: 473–482.
- Ferraresi C, de Sousa MV, Huang YY, Bagnato VS, Parizotto NA, Hamblin MR. Time response of increases in ATP and muscle resistance to fatigue after low-level laser (light) therapy (LLLT) in mice. Lasers Med Sci 2015;30: 1259–1267.
- 8. Leal-Junior ECP, Lopes-Martins RAB, Bjordal JM. Clinical and scientific recommendations for the use of photobiomodulation therapy in exercise performance enhancement and post-exercise recovery: current evidence and future directions. Braz J Phys Ther 2019;23: 71–75.
- Peake JM, Neubauer O, Della Gatta PA, Nosaka K. Muscle damage and inflammation during recovery from exercise. J Appl Physiol (1985) 2017;122:559–570.
- 10. Yang W, Hu P. Skeletal muscle regeneration is modulated by inflammation. J Orthop Translat 2018;13:25–32.
- Milioni F, Vieira LH, Barbieri RA, et al. Futsal matchrelated fatigue affects running performance and neuromuscular parameters but not finishing kick speed or accuracy. Front Physiol 2016;7:518.

 Bosco C, Luhtanen P, Komi PV. A simple method for measurement of mechanical power in jumping. Eur J Appl Physiol Occup Physiol 1983;50:273–282.

- 13. Zagatto AM, de Paula Ramos S, Nakamura FY, de Lira FS, Lopes-Martins RA, de Paiva Carvalho RL. Effects of lowlevel laser therapy on performance, inflammatory markers, and muscle damage in young water polo athletes: a doubleblind, randomized, placebo-controlled study. Lasers Med Sci 2016;31:511–521.
- 14. Ferraresi C, Bertucci D, Schiavinato J, et al. Effects of light-emitting diode therapy on muscle hypertrophy, gene expression, performance, damage, and delayed-onset muscle soreness: case-control study with a pair of identical twins. Am J Phys Med Rehabil 2016;95:746–757.
- Rossato M, Dellagrana RA, Sakugawa RL, Lazzari CD, Baroni BM, Diefenthaeler F. Time response of photobiomodulation therapy on muscular fatigue in humans. J Strength Cond Res 2018;32:3285–3293.
- 16. Huang YY, Chen AC, Carroll JD, Hamblin MR. Biphasic dose response in low level light therapy. Dose Response 2009;7:358–383.
- 17. de Souza Malta E, de Lira FS, Machado FA, Zago AS, do Amaral SL, Zagatto AM. Photobiomodulation by led does not alter muscle recovery indicators and presents similar outcomes to cold-water immersion and active recovery. Front Physiol 2019;9:1948.
- Paolillo FR, Milan JC, Aniceto IV, et al. Effects of infrared-LED illumination applied during high-intensity treadmill training in postmenopausal women. Photomed Laser Surg 2011;29:639–645.
- Paolillo FR, Corazza AV, Borghi-Silva A, Parizotto NA, Kurachi C, Bagnato VS. Infrared LED irradiation applied during high-intensity treadmill training improves maximal exercise tolerance in postmenopausal women: a 6-month longitudinal study. Lasers Med Sci 2013;28:415–422.
- Paolillo FR, Corazza AV, Paolillo AR, et al. Phototherapy during treadmill training improves quadriceps performance in postmenopausal women. Climacteric: the journal of the International Menopause Society 2014;17:285–293.

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