

# The impact of foam rolling recovery tool on oxidative stress biomarkers and performance in-water polo players: a randomized controlled trial

Josu Barrenetxea-Garcia<sup>1</sup>, Natalia Busto<sup>2</sup>, Julio Calleja-González<sup>3,4</sup>, Eduardo Sáez de Villarreal<sup>5</sup>, Andrea Perazzetti<sup>6</sup>, Sergi Nuell<sup>7</sup>, Susana Garai<sup>8</sup>, Juan Mielgo-Ayuso<sup>2,\*</sup>

<sup>1</sup>Water polo Club Leioa Waterpolo, Leioa, Spain

<sup>2</sup>Department of Health Sciences, Faculty of Health Sciences, University of Burgos, Burgos, Spain

<sup>3</sup>Department of Physical Education and Sport, Faculty of Education and Sport, University of the Basque Country (UPV/EHU), Vitoria, Spain

<sup>4</sup>Faculty of Kinesiology, University of Zagreb, Zagreb, Croatia

<sup>5</sup>Physical Performance Sports Research Center (PPSRC), Universidad Pablo de Olavide, Sevilla, Spain

<sup>6</sup>Department of Movement, Human and Health Sciences, University of Rome 'Foro Italico', Rome, Italy

<sup>7</sup>EUSES, Escola Universitària de la Salut i l'Esport, Universitat de Girona, Girona, Spain

<sup>8</sup>Uribe Integrated Health Organization, Sopolana Primary Care Unit, Sopolana, Spain

This randomized controlled trial aimed to evaluate the effects of foam rolling (FR) recovery tool on oxidative stress biomarkers and sport-specific performance in male and female water polo (WP) players during a 7-week competitive period. The study also explored sex-based differences to guide tailored recovery strategies. Thirty-four WP players were recruited, with 27 completing the protocol (13 males and 14 females), and randomly assigned to a foam roller group or a control group (CG). Testing was conducted pre- and postintervention, measuring in-water boost, throwing speed, 20-m sprint swim, and oxidative stress biomarkers: Ferric reducing ability of plasma (FRAP), glutathione, oxidative damage in proteins (ODPs), total thiols. Dietary intake was evaluated via a validated food frequency questionnaire in week 6. The FR positively influenced throwing speed ( $P=0.021$ ) and antioxidant capacity in male players (FRAP,  $P=0.006$ ). However, no significant improvements in

sprint or boost performance were observed in females, with ODP increasing in both sexes, particularly in CG females ( $P<0.001$ ). Regression analyses showed that FRAP improvements significantly predicted 20-m sprint performance ( $P=0.027$  for females,  $P=0.043$  for males). Dietary analysis revealed adequate protein and antioxidant intake but suboptimal carbohydrate consumption. These findings suggest that FR may enhance specific performance outcomes and antioxidant capacity, particularly in male WP players, though its effect on oxidative damage appears limited. Managing oxidative stress through recovery tools like FR and nutritional strategies remains essential for optimizing performance in high-intensity team sports.


**Keywords:** Team sports, Myofascial release therapy, Food intake, Antioxidants, Athletes

## INTRODUCTION

Oxidative stress occurs when the production of reactive oxygen species (ROS) surpasses the body's antioxidant defenses, leading to damage in cellular components such as lipids, proteins, and DNA (Canals-Garzón et al., 2022). This imbalance presents significant implications for both health and athletic performance (Clemente-Suárez et al., 2023). High-intensity intermittent sports, such as

water polo (WP), exacerbate oxidative stress due to increased oxygen consumption, mechanical strain, and metabolic demands. Thus, without effective recovery strategies, the accumulation of oxidative stress can impair muscle function, delay recovery process, and hinder athletic success (Margaritelis et al., 2015; Powers et al., 2020).

In particular, WP being a full-contact aquatic sport, combines aerobic and anaerobic demands with high-intensity intermittent actions such as sprint swimming, in-water boosts, and throwing

\*Corresponding author: Juan Mielgo-Ayuso  <https://orcid.org/0000-0002-6554-4602>  
Department of Health Sciences, Faculty of Health Sciences, University of Burgos,  
Burgos 09001, Spain  
Email: jfmielgo@ubu.es

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(Botonis et al., 2019; de Villarreal et al., 2014). In addition to technical and tactical skills, it has been argued that muscular strength and power, anthropometric characteristics, and WP throwing ability are the most important factors that give a clear advantage in elite competitions (de Villarreal et al., 2014; Sáez de Villarreal et al., 2015). These actions create substantial oxidative stress, especially during periods of high training loads and competition, when recovery is critical for maintaining performance (Varamenti et al., 2013). Despite the high physiological demands of WP, research on interventions to mitigate oxidative stress and support recovery in players is limited (Barrenetxea-Garcia et al., 2022; Barrenetxea-Garcia et al., 2024b). This gap highlights the need for recovery strategies that directly address the oxidative stress burden inherent to sport.

Among them, one such strategy is foam rolling (FR), which has gained attention as a practical recovery tool for athletes (Hendricks et al., 2020). This self-myofascial release technique involves applying pressure to muscle groups using a foam roller, with reported benefits such as reduced muscle soreness, enhanced range of motion, and improved blood flow (Beardsley and Škarabot, 2015; Cheatham et al., 2015). Improved blood flow and nutrient delivery may also alleviate oxidative stress, potentially enhancing recovery and performance. However, while the general benefits of FR are well-documented, its specific effects on oxidative stress biomarkers and sport-specific performance metrics, such as throwing speed, 20-m sprints, and in-water boosts, remain underexplored (Barrenetxea-García et al., 2024a; Healey et al., 2014). Understanding these potential effects is particularly important in WP, where oxidative stress management is crucial for performance sustainability.

Adding to this complexity, sex-based differences play a key role in oxidative stress responses and recovery efficacy. Therefore, hormonal differences, especially the role of estrogen in females, influence antioxidant capacity (Arias-Loza et al., 2013). Concretely, estrogen not only acts as a direct scavenger of ROS, also upregulates key antioxidant enzymes, such as superoxide dismutase and glutathione peroxidase (Viña et al., 2005). This hormonal advantage may provide females with greater baseline protection against oxidative damage, potentially reducing the perceived benefits of external recovery interventions like FR (Wiewelhoeve et al., 2019). Conversely, males, who experience higher oxidative stress due to lower estrogen levels, may derive greater benefits from FR in mitigating ROS (Powers and Jackson, 2008). These physiological differences underscore the importance of tailoring recovery strategies to personal needs (Calleja-González et al., 2018; Calleja-González et al., 2019), including sex-specific protocols, given that so far this

phenomenon has not yet been demonstrated in any significant way to our conscience.

Therefore, the present study aims to assess the impact of a FR recovery tool on oxidative stress biomarkers and sport-specific performance metrics in WP players. Specifically, this randomized controlled trial investigates whether FR influences throwing speed, 20-m sprint times, and in-water boost heights, and evaluates how sex affects these outcomes. By addressing these questions, this study seeks to fill the existing research gap, providing evidence-based insights into the role of FR in managing oxidative stress and enhancing recovery in WP players. These findings will support the development of tailored recovery strategies for players, offering practical guidance for coaches and health professionals.

## MATERIALS AND METHODS

### Design

Participants were randomly assigned to either the foam roller group (FRG) or the control group (CG) using sealed opaque envelopes. The participants were categorized based on their sex (male and female) and assigned to either the FRG or the CG (Table 1). The CG continued their usual post-training routines without any additional intervention, such as stretching or self-myofascial release. This approach aligns with a Passive CG model, where participants maintain their regular activities without specific recovery treatments. This design was chosen to allow comparison with the experimental group while reflecting real-world practices during competitive periods. Male participants ( $n = 13$ ) were further divided into FRG ( $n = 6$ ) and CG ( $n = 7$ ). Similarly, female participants ( $n = 14$ ) were allocated into FRG ( $n = 7$ ) and CG ( $n = 7$ ). This allocation strategy ensured balanced groups for the intervention and control conditions while allowing for sex-specific analyses of performance and oxidative stress biomarker changes. Testing was conducted before (pretest) and after (posttest) the 7-week training pe-

**Table 1.** Participant characteristics separated by gender and intervention groups

Characteristic	Male		Female	
	CG (n=7)	FRG (n=6)	CG (n=7)	FRG (n=6)
Height (cm)	180.33±6.98	176.71±6.32	165.13±4.73	167.25±5.83
Body mass (kg)	74.33±11.74	75.50±7.22	62.38±6.21	57.83±5.04
Age (yr)	20.17±3.54	20.43±4.08	19.63±3.02	19.33±3.67
Experience (yr)	10.67±3.20	11.14±3.67	9.88±2.42	9.33±2.34

Values are presented as mean ± standard deviation.  
CG, control group; FRG, foam roller group.

riod. The players trained 4 times per week, with sessions lasting 2 to 3 hr depending on the day and participated in one competitive match each weekend. Training sessions comprised 25% physical, 30% technical, and 45% tactical work. All practices were conducted at the Sakoneta Sport Centre (Leioa), where the official pool (25 × 12.50 m) maintained a water temperature of 26.5°C, external temperature 30° and a humidity level of 70%. Measurements were performed in the same venue and under consistent environmental conditions in order to minimize external variability. During the study, WP male players competed in the First Regional League, while WP females participated in the Second National League. The 7-week intervention period was chosen based on the duration of the in-season competitive phase and previous research investigating the chronic effects of recovery interventions (Beardsley and Škarbot, 2015; Hendricks et al., 2020). This timeframe was considered sufficient to observe adaptations in performance and oxidative stress biomarkers while fitting within the athletes' real competitive schedule.

All participants were fully informed of the study procedures and voluntarily signed informed consent forms. In cases involving minors, consent was obtained from parents or guardians. Before the study began, each participant underwent a physical examination conducted by the team physician to confirm the absence of contraindications such as injury, disease, or medication use. The study was approved by the Human Research Ethics Committee of the University of Basque Country (Approval code: M10\_2022\_056) and complied with the ethical principles outlined in the Declaration of Helsinki. The study was registered under Trial ID: AC-TRN12622000550707 by Australian New Zealand Clinical Trials Registry. Data handling followed Spain's Organic Law 15/1999 on Personal Data Protection and adhered to the ethical standards required under Law 14/2007 for human experimentation, as outlined in the Spanish Official State Gazette (n° 159).

Participants in the FRG performed self-myofascial release using a foam roller (38 cm × 13 cm) after every training session, as detailed in Barrenetxea-García et al. (2024a). Participants were instructed on proper execution, familiarized with the exercises, and supervised during all sessions. The protocol consisted of 6 exercises targeting major muscle groups involved in WP performed under supervision to ensure proper technique. Each exercise involved rolling for 60 seconds with a 30-sec rest period (Wiewelhove et al., 2019). The CG followed their regular post-training routine without FR. The players were familiar with the protocols during the pre-season, and it was answered every day before training and competitive sessions.

## Participants

A total 34 male and female WP players were recruited from the Leioa Waterpolo Club in Spain. Only 27 participants completed the study complete protocol. The sample size was calculated using G\*Power 3.1.9.2 program based on a repeated measures analysis of variance (ANOVA) (within-between interaction), considering an effect size (ES)  $f=0.25$  (medium), alpha level = 0.05, power = 0.80, 2 groups, and 2 time points (pre- and postintervention). The analysis indicated that a minimum of 24 participants would be required (Faul et al., 2009). Inclusion criteria were as follows: (a) No diagnosed muscle injuries within 2 months prior to or during the intervention. (b) Completion of at least 80% of the training sessions. (c) A minimum of 5 years of competitive WP experience. On the other hand, exclusion criteria proposed participants were allowed to withdraw from the study at any time. They were also advised to avoid any additional strenuous physical activities outside of the programmed intervention and to maintain consistent hydration, sleep, and dietary habits. The use of drugs or other substances that could influence study outcomes was strictly prohibited. In addition to the programmed training intervention, for the duration of the experiment before testing day, participants were given specific instructions to follow; (a) ensuring a minimum of 8 hr of restful sleep, (b) consuming the appropriate amount of carbohydrates as prescribed for each individual, while also maintaining proper hydration, and (c) wearing identical footwear during both the pre and posttests.

## Methods

Performance tests and order: the 20-m sprint swim, throwing speed and in-water boost evaluated using validated methods:

### 20-m sprint swim

Conducted in a 25-m pool using an electronic timing system (Casio HS-3V-1). Participants performed 3 sprints at maximum speed with a 5-min rest among attempts and the best time was recorded (Barrenetxea-Garcia et al., 2023; Barrenetxea-García et al., 2024a; de Villarreal et al., 2014; Martin et al., 2021; Sáez de Villarreal et al., 2015). The 20-m sprint swim test was deemed to be reliable based on an ICC = 0.85 (0.81–0.89).

### Throwing speed

Measured with a radar gun (Stalker SOLO) during 3 sets of 3 throws with 15 sec among throws and one minute between sets. The highest velocity excluding outliers was recorded (Barrenetxea-Garcia et al., 2023; Barrenetxea-García et al., 2024a; de Villarreal

et al., 2014; Martin et al., 2021; Sáez de Villarreal et al., 2015). The WP throwing speed was deemed to be reliable based on an ICC = 0.87 (0.84–0.90).

#### ***In-water boost***

Evaluated using a vertical jump test in water, captured with a video camera at 50 Hz. The maximum height of hand contact on a board with a centimeter scale was analyzed from 3 trials (Barrenetxea-García et al., 2024a; de Villarreal et al., 2014; Martin et al., 2021; Sáez de Villarreal et al., 2015). The in-water boost test was deemed to be reliable based on an ICC = 0.90 (0.87–0.93).

To evaluate the impact of FR on oxidative stress and recovery, specific biomarkers were analyzed from blood samples collected pre- and postintervention. Blood samples were obtained via venipuncture from the antecubital vein by a qualified nurse following standardized procedures. Samples were processed immediately with plasma and serum separated by centrifugation at 1,500×g for 15 min at 4°C. Aliquots were stored at -80°C until analysis. The following biomarkers were measured:

#### ***Total proteins (g/dL)***

Total protein levels in plasma were determined using a colorimetric method based on the Biuret reaction where proteins form a complex with copper ions in an alkaline solution. The intensity of the color change was measured spectrophotometrically at 540 nm which correlates with the protein concentration.

#### ***Total thiols (μmol/mg protein)***

Total thiols were assessed using Ellman's reagent, 5,5'-dithio-bis-(2-nitrobenzoic acid), which reacts with sulfhydryl groups to produce a yellow chromogen measurable at 412 nm. The concentration of thiols was normalized to protein levels to account for individual differences in plasma protein content.

#### ***Antioxidant capacity by ferric reducing ability of plasma (μmol Trolox/mL)***

The ferric reducing ability of plasma (FRAP) assay quantified the antioxidant potential of plasma by measuring the reduction of a ferric-tripyridyltriazine complex to its ferrous form. This reduction generates a blue color with an absorbance peak at 593 nm. Results were expressed in Trolox equivalents a vitamin E analog used as a standard.

#### ***Glutathione (GSH, μg/mg protein)***

GSH levels were measured using a glutathione reductase recy-

cling assay. This method involves the reduction of 5,5'-dithio-bis-(2-nitrobenzoic acid) by GSH, forming a yellow product measurable at 412 nm. The assay quantifies both reduced and oxidized glutathione, providing insight into the redox status of the athlete.

#### ***Oxidative damage in proteins (nmol/mg protein)***

Protein oxidation was assessed by measuring protein carbonyl content, a widely recognized marker of oxidative damage. Carbonyl groups in oxidized proteins were derivatized with 2,4-dinitrophenylhydrazine, forming hydrazones detectable at 370 nm using spectrophotometry. Results were expressed as nmol of carbonyl per milligram of protein.

#### ***Standardization and quality control***

Each assay was conducted in triplicate to ensure precision and inter-assay variability was maintained below 5%. Calibration curves were prepared for each assay using appropriate standards and validated before sample analysis. All spectrophotometric readings were obtained using a BioTek Synergy HT Plate Reader (BioTek Instruments), ensuring consistent and reliable results.

#### ***Dietary assessment***

Participants completed a validated food frequency questionnaire during the last week of the intervention in order to assess their dietary intake. The food frequency questionnaire captured information on the frequency and portion sizes of consumed foods, allowing for the calculation of total energy intake, as well as macronutrient and micronutrient distribution. Data were analyzed using EasyDiet software (Mettler-Toledo S.A.E., online version), which has been validated for use in sports nutrition research. Macronutrient and micronutrient intake were normalized to body mass (kg) to account for individual differences among participants (Mielgo-Ayuso et al., 2015; Mielgo-Ayuso et al., 2020).

#### ***Statistical analysis***

Data are presented as mean ± standard deviation and differences are expressed as percentage change (delta, Δ). Data normality was assessed using the Shapiro–Wilk test, which is appropriate for small sample sizes ( $n < 30$ ). Homogeneity of variances was tested using Levene test. All variables met the assumptions of normality ( $P > 0.05$ ), thus parametric tests were applied. Respectively standardized differences were calculated using Hedges'  $g$  ES to evaluate the practical significance of changes, with ESs interpreted based on Cohen criteria: trivial ( $< 0.2$ ), small (0.2–0.5), moderate (0.5–0.8) and large ( $> 0.8$ ). Additionally, qualitative probabilistic mech-

anistic inference was applied to assess the practical relevance of the observed changes, using 90% confidence intervals for each outcome. To evaluate whether these changes were meaningful, the smallest worthwhile change was defined as 0.2 times the baseline standard deviation (SD). Based on the likelihood that the true effect exceeded this threshold, qualitative descriptors such as very unlikely, possibly, likely, very likely, or most likely were assigned, in accordance with published guidelines (Hopkins, 2007). This approach allows for a more nuanced interpretation of the results by combining statistical uncertainty with practical significance.

Two-way repeated measures ANOVA was conducted separately for male and female participants to analyze within- and between-group differences. Time (pre- and postintervention) was considered the within-subject factor and group (FRG vs. CG) was the between-subject factor. Significant interaction effects were further explored using Bonferroni, post-hoc tests to determine the source of differences.

A stepwise multiple regression analyses were performed separately for males and females to identify key oxidative stress biomarkers predicting performance changes. For each sex, the dependent variables included changes in specific performance tests ( $\Delta$ 20-m sprint swim,  $\Delta$ Throwing speed and  $\Delta$ In-Water Boost), while independent variables included changes in oxidative stress biomarkers such as  $\Delta$ T proteins,  $\Delta$ total thiols,  $\Delta$ AC by FRAP,  $\Delta$ GSH, and  $\Delta$ ODP by carbonyls. Adjusted  $R^2$  was used in these models to account for the number of predictors, ensuring an accurate assessment of model fit and explanatory power. This approach aimed to identify sex-specific predictors of performance improvements or declines associated with changes in oxidative stress biomarkers. Data analyses were conducted using IBM SPSS Statistics ver. 25.0 (IBM Co.), with the significance level set at  $P < 0.05$ .

## RESULTS

Any significant differences between groups or sexes were noted and analyzed within the context of the study outcomes. The dietary analysis revealed key findings regarding energy, macronutrients, and antioxidant vitamin intake among male and female WP players (Table 2). Both sexes met protein recommendations, with males consuming  $22.68\% \pm 4.07\%$  of energy from protein and females  $21.29\% \pm 3.53\%$ , supporting recovery. Lipid intake was high in both groups (males,  $36.74\% \pm 8.27\%$ ; females,  $36.35\% \pm 4.94\%$ ). Carbohydrate intake was for males:  $37.41\% \pm 4.41\%$  and for females:  $40.95\% \pm 7.40\%$ . Vitamin A intake varied widely, with males in the FRG consuming more ( $3,243.85 \pm 4,333.62 \mu\text{g}$ ) than CG males ( $1,126.52 \pm 521.94 \mu\text{g}$ ),  $P < 0.05$ . Females had lower average intakes (CG:  $1,556.82 \pm 1,029.86 \mu\text{g}$ , FRG:  $1,342.33 \pm 804.15 \mu\text{g}$ ). While intakes exceeded the recommended dietary allowance (PDA) ( $900 \mu\text{g}$  for males and  $700 \mu\text{g}$  for females), variability indicates inconsistent consumption of vitamin A-rich foods. Vitamin E intake was sufficient across most groups, with males averaging  $19.49 \pm 12.12 \text{ mg}$  and females  $18.37 \pm 9.86 \text{ mg}$ , exceeding the RDA of  $15 \text{ mg/day}$ . Vitamin C Both sexes had adequate intake, with males consuming  $310.07 \pm 167.86 \text{ mg}$  on average and females  $366.28 \pm 192.33 \text{ mg}$ , far exceeding the RDA of  $90 \text{ mg/day}$  for males and  $75 \text{ mg/day}$  for females. Regarding specific performance measurements, the changes for FRG and CG between male and female WP players are shown in Table 3.

### 20-m sprint swim

In the FRG group, males showed a slight improvement in their speed swimming performance with an increase of  $1.3\%$ , whereas in the CG there was little change with a variation of  $-0.57\%$ . However, the two-way repeated measures ANOVA analysis showed no

**Table 2.** Food frequency values in male (n = 13) and female (n = 14) water polo players

Variable	Male			Female		
	CG	FRG	Total	CG	FRG	Total
Energy (kcal)	2,776 ± 908	3,141 ± 839	2,972 ± 855	2,972 ± 1071	2,559 ± 754	2,736 ± 890
Proteins (%)	21.54 ± 3.67	23.66 ± 4.42	22.68 ± 4.07	19.90 ± 3.45	22.32 ± 3.43	21.29 ± 3.53
Lipids (%)	39.58 ± 5.93	34.31 ± 9.63	36.74 ± 8.27	35.54 ± 4.81	36.95 ± 5.27	36.35 ± 4.94
Carbohydrates (%)	37.61 ± 5.21	37.25 ± 4.03	37.41 ± 4.41	43.49 ± 8.08	39.05 ± 6.75	40.95 ± 7.40
Vitamin A (μg)	1,127 ± 521	3,244 ± 4,333	2,267 ± 3,273	1,557 ± 1,030	1,342 ± 804	1,434 ± 877
Vitamin E (mg)	22.08 ± 16.39	17.28 ± 7.60	19.49 ± 12.12	18.42 ± 8.60	18.34 ± 11.30	18.37 ± 9.86
Vitamin C (mg)	377.9 ± 151.9	251.9 ± 169.1	310.1 ± 167.9	407.3 ± 192.8	335.5 ± 199.1	366.3 ± 192.3

Values are presented as mean ± standard deviation.  
CG, control group; FRG, foam roller group.

**Table 3.** Specific performance values for the control and experimental group in male (n = 13) and female (n = 14) water polo players

Variable	CG				FRG				Interaction								
	Pre	Post	Δ (% Change)	P-value	ES (95% CI)	ES M	PI	Pre	Post	Δ (% Change)	P-value	ES (95% CI)	ES M	PI	Sig.	η <sup>2</sup>	ES
<b>Male</b>																	
20-m sprint swim (sec)	11.78±0.33	11.71±0.52	-0.57±3.27	0.688	0.15 (+0.98; 1.28)	Tr	P	11.23±0.50	11.37±0.42	1.30±2.91	0.310	-0.28 (-1.33; 0.77)	S	P	0.723	0.012	N
Throwing speed (m/sec)	63.62±4.44	64.09±4.02	0.8±2.55	0.506	-0.10 (-1.23; 1.03)	Tr	P	63.55±3.66	65.29±3.48	2.77±2.38	0.021	-0.45 (-1.51; 0.61)	S	P	0.026	0.377	Mo
In-water boost (cm)	122.67±5.99	121.10±4.72	-1.21±2.76	0.307	0.27 (-0.87; 1.41)	S	P	118.50±8.50	118.29±6.45	-0.04±3.25	0.876	0.03 (-1.02; 1.07)	T	P	0.375	0.072	Mi
<b>Female</b>																	
20-m sprint swim (sec)	12.79±0.58	12.64±0.85	-1.18±4.22	0.528	0.18 (-0.96; 1.31)	Tr	P	12.94±0.56	12.86±0.49	-0.57±3.07	0.579	0.14 (-0.94; 1.12)	T	P	0.373	0.067	Mi
Throwing speed (m/sec)	51.89±4.44	51.91±4.17	0.11±3.14	0.972	-0.01 (-1.14; 1.13)	Tr	P	51.36±3.90	51.77±4.05	0.82±2.92	0.441	-0.10 (-1.08; 0.88)	T	P	0.606	0.023	N
In-water boost (cm)	115.57±6.12	111.83±5.18	-3.19±2.05	0.012	0.61 (-0.55; 1.77)	Mo	L	114.89±6.66	111.21±5.89	-3.14±2.75	0.015	0.55 (-0.45; 1.55)	Mo	L	0.001	0.649	St

Values are presented as mean ± standard deviation.

GC, control group; FRG, foam roller group; ES, effect size; CI, confidence interval; ES M, effect size magnitude; PI, probabilistic inference; Tr, trivial; S, small; Mo, moderate; P, possibly; L, likely or probably; N, no effect; Mi, minimum; St, strong.

**Table 4.** Oxidative stress biomarkers for the control and experimental group in male (n = 13) and female (n = 14) water polo players

Variable	CG				FRG				Interaction								
	Pre	Post	Δ (% Change)	P-value	ES (95% CI)	ES M	PI	Pre	Post	Δ (% Change)	P-value	ES (95% CI)	ES M	PI	Sig.	η <sup>2</sup>	ES
<b>Male</b>																	
Total proteins (g/dL)	5.75±0.05	5.87±0.14	2.05±1.79	0.038	-1.05 (-2.26; 0.15)	La	L	5.71±0.15	5.89±0.12	3.31±3.36	0.041	-1.23 (-2.38; -0.09)	La	V	0.005	0.526	Mo
Total thiols (μmol/mg protein)	0.46±0.03	0.43±0.03	-203.20±488.40	0.151	0.92 (-0.27; 2.11)	La	L	0.45±0.18	0.42±0.03	-7.09±9.51	0.093	0.22 (+0.83; 1.27)	S	P	0.026	0.377	Mo
AC by FRAP (μmol Trolox/mL)	0.42±0.00	0.47±0.03	10.67±7.78	0.020	-2.18 (-3.60; -0.75)	La	M	0.42±0.00	0.46±0.03	9.42±6.06	0.006	-1.75 (-2.99; -0.52)	La	M	<0.001	0.106	St
GSH (μg/mg protein)	8.30±1.80	10.22±1.59	27.56±29.73	0.080	-1.04 (-2.25; 0.16)	La	L	8.06±4.39	10.20±4.79	53.21±86.64	0.486	-0.43 (-1.49; 0.63)	S	P	0.237	0.124	St
ODP: carbonyls (nmol/mg protein)	42.26±7.97	48.52±9.24	19.15±34.87	0.310	-0.67 (-1.83; 0.49)	Mo	L	41.54±3.51	51.78±4.70	25.38±15.72	0.002	-2.30 (-3.65; -0.95)	La	M	0.013	0.446	Mo
<b>Female</b>																	
Total proteins (g/dL)	5.52±0.18	5.85±0.08	6.12±4.33	0.015	-2.19 (-3.62; -0.76)	La	M	5.57±0.13	5.90±0.13	5.99±4.18	0.005	-2.40 (-3.68; -1.11)	La	M	<0.001	0.706	St
Total thiols (μmol/mg protein)	0.44±0.02	0.39±0.02	-10.86±6.14	0.008	2.31 (0.85; 3.77)	La	M	0.42±0.02	0.39±0.02	-6.44±8.30	0.061	1.42 (0.32; 2.51)	La	V	0.001	0.607	Mo
AC by FRAP (μmol Trolox/mL)	0.42±0.00	0.43±0.01	1.19±1.99	0.203	-1.31 (-2.55; -0.06)	La	V	0.42±0.01	0.42±0.03	0.60±7.13	0.818	0.00 (-0.98; 0.98)	T	P	0.562	0.029	Mo
GSH (μg/mg protein)	9.75±1.26	13.50±4.62	42.83±58.99	0.145	-1.02 (-2.23; 0.18)	La	L	10.31±3.59	11.28±4.65	28.86±74.00	0.632	-0.22 (-1.20; 0.76)	S	P	0.132	0.178	St
ODP: carbonyls (nmol/mg protein)	34.06±4.69	56.41±4.23	68.12±24.81	<0.001	-4.62 (-6.78; -2.45)	La	M	35.17±3.82	51.10±4.12	47.51±25.69	<0.001	-3.79 (-5.43; -2.15)	La	M	<0.001	0.921	St

Values are presented as mean ± standard deviation.

GC, control group; FRG, foam roller group; ES, effect size; CI, confidence interval; ES M, effect size magnitude; PI, probabilistic inference; AC, antioxidant capacity; FRAP, ferric reducing ability of plasma; GSH, glutathione; ODP, oxidative damage in proteins; La, large; M, moderate; P, possibly; L, likely or probably; M, most likely or almost certainly; V, very likely; T, trivial; S, small; Mo, moderate; P, possibly; L, likely or probably; N, no effect; Mi, minimum; St, strong.

significant interaction between group and time for the men ( $P > 0.05$ ). The situation was similar for the female participants: those in the FRG group showed a slight decrease in performance (-0.57%), whereas a slight improvement (-1.18%) was observed in the CG. However, the ANOVA did not reveal any significant interaction between group and time ( $P > 0.05$ ).

### Throwing speed

Male participants in the FRG group showed a significant increase in throwing speed (+2.77%), as revealed by the two-way repeated measures ANOVA (group × time interaction:  $P = 0.021$ ), outperforming CG males who only improved by +0.8%. Among females, both groups displayed negligible changes, with FRG showing a slight improvement (+0.82%) compared to CG (+0.11%).

### In-water boost

Reductions were observed across all groups in in-water boost performance. Among females, both FRG (-3.14%) and CG (-3.19%) showed significant decreases, as revealed by the two-way repeated measures ANOVA (group × time interaction:  $P = 0.015$  and  $P = 0.012$ , respectively). In contrast, males in both groups showed minimal changes, and the interaction effect was not statistically significant ( $P > 0.05$ ). Concerning oxidative stress biomarkers, Table 4 presents changes for FRG and CG players separated by sex.

### Total proteins

Significant increases were observed in both groups for males (+2.05% CG, +3.31% FRG) and females (+6.12% CG, +5.93% FRG) with FRG showing more pronounced improvements ( $P < 0.05$ ; large ESs).

### Total thiols

A decrease in total thiol levels was observed in both sexes. In

men, the CG showed a more pronounced, although not significant, decrease (-203.2%,  $P = 0.151$ ) compared to the FRG group, whose decrease was much smaller (-7.09%,  $P = 0.093$ ). In the case of women, a significant interaction between group and time was found ( $P = 0.001$ ;  $\eta^2 = 0.607$ ): the CG showed a significant reduction in thiols (-10.86%,  $P = 0.008$ ), whereas in the FRG group, the reduction was more moderate and close to the threshold of significance (-6.44%,  $P = 0.061$ ).

### AC by FRAP

Although both groups of men improved their antioxidant capacity, there was a significant interaction between group and time ( $P < 0.001$ ;  $\eta^2 = 0.106$ ). Participants in the FRG group showed an increase of 9.42% ( $P = 0.006$ ), while those in the CG showed a slightly greater improvement of 10.67% ( $P = 0.020$ ). For women, there were no significant changes in either group, and no interaction effect was found ( $P = 0.562$ ).

### Glutathione

Both groups exhibited increases in GSH levels with FRG males showing a higher rise (+53.21%) compared to CG males (+27.56%). In females, CG participants had a substantial increase (+42.83%), though not statistically significant ( $P > 0.05$ ).

### Oxidative damage in proteins

Significant increases in oxidative damage in proteins (ODPs) were observed in all groups. In men, there was a significant interaction between group and time ( $P = 0.013$ ;  $\eta^2 = 0.446$ ): players in the FRG group showed a remarkable increase of 25.38% ( $P = 0.002$ ), while those in the CG also improved, although to a lesser extent and without statistical significance (+19.15%,  $P = 0.310$ ). In women, the interaction effect was even more pronounced ( $P < 0.001$ ;  $\eta^2 = 0.921$ ): participants in the CG had the highest increase (+68.12%,

**Table 5.** Associated variables to estimate specific performance values (dependent variable) in female water polo players ( $n = 14$ )

Model	Nonstandardized coefficients		Standardized coefficients	<i>t</i>	Significance	Summary of the model	
	<i>B</i>	Deviation error	Beta			<i>R</i> <sup>2</sup>	Adjusted <i>R</i> <sup>2</sup>
Δ20-m sprint swim							
(Constant)	-1.153	0.790		-1.460	0.170	0.347	0.293
ΔAC by FRAP	0.380	0.150	0.589	2.526	0.027		
ΔIn-water boost							
(Constant)	-0.319	1.325		-0.241	0.814	0.317	0.260
ΔODP: carbonyls	-0.051	0.021	-0.563	-2.358	0.036		

Independent variables: oxidative stress biomarkers

ΔAC by FRAP, change in antioxidant capacity by ferric reducing ability of plasma; ΔODP, change in oxidative damage to proteins.

**Table 6.** Associated variables to estimate specific performance values (dependent variable) in male water polo players (n= 13)

Model	Nonstandardized coefficients		Standardized coefficients	t	Significance	Summary of the model	
	B	Deviation error	Beta			R <sup>2</sup>	Adjusted R <sup>2</sup>
Δ20-m Sprint swim							
(Constant)	-1.124	0.731		-1.538	0.137	0.154	0.120
ΔAC by FRAP	0.172	0.081	0.392	2.129	0.043		
Independent variables: oxidative stress biomarkers							

ΔAC by FRAP, change in antioxidant capacity by ferric reducing ability of plasma.

$P < 0.001$ ), followed by those in the FRG group, with an equally significant increase of 47.51% ( $P < 0.001$ ).

The association between oxidative stress biomarkers and specific performance metrics in female WP players were analyzed (Table 5). For the 20-m sprint swim, the model explained 34.7% of the variance ( $R^2 = 0.347$ , adjusted  $R^2 = 0.293$ ). The ΔAC by FRAP was identified as a significant positive predictor of sprint performance ( $B = 0.380$ ;  $t = 2.526$ ;  $P = 0.027$ ), indicating that increases in AC were associated with improved sprint performance. The constant term was not significant ( $P = 0.170$ ).

In the case of the in-water boost, the model accounted for 31.7% of the variance ( $R^2 = 0.317$ , adjusted  $R^2 = 0.260$ ). The ΔODP emerged as a significant negative predictor of performance in this explosive movement metric ( $B = -0.051$ ;  $t = -2.358$ ;  $P = 0.036$ ), suggesting that higher levels of ODP were linked to reduce in-water boost performance. The constant term for this model was also not significant ( $P = 0.814$ ).

The relationship between oxidative stress biomarkers and the 20-m sprint swim performance in male WP players was analyzed (Table 6). The model explained 15.4% of the variance ( $R^2 = 0.154$ ; adjusted  $R^2 = 0.120$ ). The ΔAC by FRAP was identified as a significant positive predictor of performance in the sprint swim ( $B = 0.172$ ,  $t = 2.129$ ,  $P = 0.043$ ), indicating that improvements in AC were associated with enhanced sprint swim performance.

## DISCUSSION

The primary main aim of this study was to evaluate the effects of a FR intervention on oxidative stress biomarkers and sport-specific performance metrics in male and female WP players. The results demonstrated that FR positively influenced certain performance metrics and oxidative stress recovery-management, particularly in male players. Improvements in throwing speed and AC by FRAP were observed, while declines in in-water boost performance and increases in ODP were noted across groups, with sex-specific differences. Regression analyses further emphasized signif-

icant relationships between oxidative stress biomarkers and physical performance, highlighting the role of FR as recovery tool in managing oxidative stress to optimize performance outcomes.

Our data described that the improvement in 20-m sprint swim performance observed in male FRG participants was +1.3%. This phenomenon has been described in scientific literature given that is aligned with FR's ability to reduce muscle stiffness and enhance recovery (Wiewelhove et al., 2019). These potential effects, coupled with improved blood flow, support sustained performance during repeated sprint activities (Cheatham et al., 2015). However, females demonstrated nonsignificant changes, potentially due to hormonal influences such as estrogen. These changes could be linked to antioxidative benefits that may modulate recovery processes (Arias-Loza et al., 2013), although hormonal levels were not assessed in the present study. Besides, other potential explanation could be the differences in muscle fiber type composition and baseline fitness levels to contribute to these sex-based discrepancies (Nuzzo, 2024).

On the other hand, the significant improvement in throwing speed among male FRG players (+2.77%,  $P = 0.021$ ) highlights the potential of FR to enhance power-based movements (Wiewelhove et al., 2019). Previous studies suggest that FR may reduce muscle adhesions and promote neuromuscular efficiency (Beardsley and Škarabot, 2015), which could help explain the improvements in throwing speed observed in our male participants. However, in females, the lack of significant improvement may reflect lower baseline power output or differences in biomechanical efficiency during throwing. Unfortunately, no previous evidence has been reported for the best of our knowledge.

The decline in in-water boost performance across all groups underscores the complexity of this explosive movement, which heavily relies on rapid energy turnover and neuromuscular coordination (Martin et al., 2021). Regression analyses of our data revealed a negative relationship between ΔODP and Δin-water boost in females ( $B = -0.051$ ,  $P = 0.036$ ), suggesting that increased ODP impairs power-based performance. The ROS has been shown to dis-

rupt mitochondrial function and ATP availability, critical for explosive activities (Powers et al., 2020). This finding underscores the need for recovery strategies that specifically target oxidative damage (O'Connor et al., 2022).

In contrast, although both male groups improved in AC by FRAP, a significant group  $\times$  time interaction suggests that FR influenced the pattern of antioxidant adaptation. The increase in the FRG group (+9.42%,  $P = 0.006$ ) supports its potential role in enhancing antioxidant defenses during the competitive period. This improvement was a significant predictor of 20-m sprint swim performance, explaining 34.7% of the variance in females and 15.4% in males. Enhanced antioxidant capacity supports both aerobic and anaerobic energy systems during high-intensity efforts, aligning with findings from Margaritelis et al. (2015) and Clemente-Suárez et al. (2023). One more time, the gender improvement is different between both genders. Although the changes in total thiol levels in the women in the FRG group were not statistically significant, the decrease was smaller than in the CG. This trend suggests that FR may have a possible protective effect on thiol-based antioxidant defenses, which deserves further investigation.

Increased GSH levels, particularly in male FRG participants (+53.21%), reflect the ability of FR to support redox balance through glutathione recycling, critical for neutralizing ROS during physical exertion. However, the substantial increases in ODP observed in females, particularly in the CG (+68.12%), highlight the vulnerability of proteins to oxidative damage when recovery strategies are inadequate. Notably, the FRG also experienced a significant increase in ODP (+47.51%), suggesting that FR may attenuate, but not fully prevent, oxidative protein damage. This supports previous findings that oxidative stress impairs muscle recovery and performance (Clemente-Suárez et al., 2023).

Besides, the relationships identified in regression analyses emphasize the interplay between oxidative stress biomarkers and physical performance. These biomarkers collectively provide a comprehensive view of the oxidative stress status and recovery in WP players, capturing antioxidant defenses (FRAP-GSH- thiols). The ODP and metabolic stress (total proteins). This multi-marker approach allows for a detailed evaluation of how FR could influence the physiological balance between oxidative stress and recovery mechanisms in athletes. These improvements in  $\Delta$ AC by FRAP positively impacted sprint swim performance, while increases in  $\Delta$ ODP negatively influenced explosive power in females.

These findings partially align with biochemical evidence that managing oxidative stress during recovery is critical for optimizing performance (Nikolaidis et al., 2012), as some oxidative stress

markers (e.g., FRAP) improved with FR use, whereas others like ODP increased despite the intervention. The FR appears to be a partially effective intervention for enhancing antioxidant defenses. However, its capacity to mitigate oxidative damage, such as ODP, may be limited and warrants further investigation.

The constant term was not significant ( $P = 0.137$ ), suggesting that the relationship between oxidative stress biomarkers and sprint swim performance is predominantly driven by the changes in FRAP. These results emphasize the importance of antioxidant capacity in supporting speed-based performance in male WP athletes, highlighting its role in managing oxidative stress for optimal athletic output (Park and Kwak, 2016). Therefore, these results highlight the role of oxidative stress biomarkers in predicting performance outcomes as well. Improvements in  $\Delta$ AC by FRAP positively impacted speed-based performance in the sprint swim while increases in  $\Delta$ ODP negatively affected power-based movements such as the in-water boost. This underscores the importance of managing oxidative stress to optimize specific performance metrics in female WP players.

Finally, it is worth mentioning that in our previous study (Barrenetxea-García et al., 2024a) that analyzed the effect of FR on WP performance, the results coincided in that there was no clear evidence of the potential effect on the use of this tool in a sport such as WP. It has been shown that water reduces pressure on the musculoskeletal system and stress on the extremities due to its buoyancy properties, decreasing the risk of injury in athletes (Biswas and Ghosh, 2022). However, the data from the present study show the exact opposite. This reality may be because the results of the study by Barrenetxea-García et al. (2024a) focused on general measurements and subjective perceptions, unlike the physiological mechanisms analyzed in the current study. In addition, the number of participants was smaller compared to that previously published (Barrenetxea-García et al., 2024a) due to participant mortality. Even so, to highlight these or other results, more research is needed that combines physiological measurements (oxidative stress) with performance tests and subjective scales.

This study has several limitations. The relatively small sample size may restrict the generalizability of the findings, and the 7-week intervention period might not fully capture long-term adaptations to FR. Hormonal levels were not measured, limiting insights into sex-specific differences. It should be noted that it is difficult to obtain larger samples in athletes, as not many have the availability to comply with the training and supplementation instructions required by the study. Moreover, sampling using a convenient, non-probabilistic sampling procedure may produce results that are not

representative of the rest of the population. These limitations may underrepresent the results and may affect study outcomes. Nevertheless, the purpose of this study is not to transfer information to the general population. However, the study's strengths include its integration of performance metrics with oxidative stress biomarkers, the inclusion of both male and female athletes, and its ecological validity, ensuring applicability to real-world training scenarios. Future research should investigate the long-term effects of FR, explore its interactions with other recovery strategies such as nutrition, and examine its effects on hormonal responses. Future studies with larger sample sizes and direct measurements of hormonal status could provide further insights into the sex-specific mechanisms underlying recovery responses to FR interventions.

In summary, the study demonstrates that FR positively influences oxidative stress biomarkers and certain sport-specific performance metrics, particularly in male WP players. The observed improvements in AC by FRAP and their correlation with sprint swim performance underscore the importance of managing oxidative stress during recovery. Conversely, increases in ODP negatively impacted power-based movements, emphasizing the need for targeted recovery strategies. Although FR may reduce the extent of oxidative protein damage, the increases observed suggest that additional or complementary recovery interventions may be necessary. These findings provide a foundation for integrating FR into recovery protocols to enhance performance and mitigate oxidative stress in high-intensity sports like WP.

## CONFLICT OF INTEREST

No potential conflict of interest relevant to this article was reported.

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