DOES BEETROOT JUICE SUPPLEMENTATION AFFECT TO SPORT PERFORMANCE IN HIGH PERFORMANCE FEMALE VOLLEYBALL PLAYERS?

¿AFECTA LA SUPLEMENTACIÓN CON ZUMO DE REMOLACHA AL RENDIMIENTO DEPORTIVO EN JUGADORAS DE VOLEIBOL DE ALTO RENDIMIENTO?

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Abstract

It has been suggested that the response to beetroot juice supplementation (BRJ) may be mediated by sex, but research is limited on studies focused on the effect of this supplement on explosive efforts in female athletes. Thus, the aim of this study was to analyze the effect of BRJ on sports performance in female volleyball players. Twelve female (22.9 \pm 3.6 years) high-performance volleyball players (Spain Superliga 2) randomly ingested an enriched nitrate BRJ (12.8 mmol NO3-) or a depleted NO3- placebo (PLA) in two experimental sessions. In each experimental session, participants performed different sports performance tests such as countermovement jump (CMJ), block jump (BJ), attack jump (AJ), a modified T-test, and a 5-0-5 test. The results reported a statistically significant enhancement in CMJ after BRJ compared to PLA (BRJ: 31.89 \pm 2.84 cm vs. PLA: 31.48 \pm 2.37 cm; *p* = .044; *d* = 0.66), but no significant differences were reported for the other tests used. The findings suggest that BRJ supplementation may enhance vertical jump performance (CMJ) in female volleyball players, but it did not lead to significant improvements in volleyball-specific actions (i.e., BJ and AJ), agility, or acceleration.

Keywords: Ergogenic aids, sport nutrition, team sports, physical performance, women athletes.

Resumen

Se ha sugerido que la respuesta a la suplementación con zumo de remolacha (BRJ) puede estar mediada por el sexo, pero la investigación sobre el efecto de este suplemento en esfuerzos explosivos en atletas femeninas es limitada. Por lo tanto, el objetivo de este estudio fue analizar el efecto del BRJ en el rendimiento deportivo de jugadoras de voleibol. Doce jugadoras de voleibol de alto rendimiento (22.9 \pm 3.6 años) de la Superliga 2 de España ingirieron aleatoriamente un BRJ en el rendimiento (21.9 \pm 3.6 años) de la Superliga 2 de España ingirieron aleatoriamente un BRJ en el rendimiento (22.9 \pm 3.6 años) de la Superliga 2 de España ingirieron aleatoriamente un BRJ encipatoria de voleibol con NO3- reducido (PLA) en dos sesiones experimentales. En cada sesión experimental, las participantes realizaron diferentes pruebas de rendimiento deportivo como el salto con contramovimiento (CMJ), el salto de bloqueo (BJ), el salto de ataque (AJ), un T-test modificado y una prueba 5-0-5. Los resultados reportaron una mejora estadísticamente significativa en el CMJ después del BRJ en comparación con el PLA (BR]: 31.89 \pm 2.84 cm vs. PLA: 31.48 \pm 2.37 cm; p = .044; d = 0.66), pero no se reportaron diferencias significativas en las otras pruebas utilizadas. Los resultados de este estudio sugieren que la suplementación con BRJ podría mejorar el rendimiento en el salto vertical (CMJ) en jugadoras de voleibol, pero no conllevó mejoras significativas en acciones específicas de voleibol (i.e., BJ y AJ), en su agilidad o en su aceleración.

Palabras Clave: Ayudas ergogénicas, nutrición deportiva, deportes de equipo, rendimiento físico, atletas femeninas.

Introduction

According to the International Olympic Committee (IOC), a dietary supplement comprises foods, food components, nutrients, or non-food compounds deliberately consumed in addition to one's habitual diet to attain specific health and/or performance benefits (Maughan et al., 2018). Dietary supplement intake is a common practice among athletes of different sport modalities, including team sports, regardless of sex and competitive level (Günalan et al., 2022; Muñoz et al., 2020). Various international expert institutions such as the Australian Institute of Sport (AIS) and IOC (Maughan et al., 2018) have classified B-alanine, caffeine, creatine, NO3-, and sodium bicarbonate as sport supplements based on high scientific

evidence. Specifically, the prevalence of NO3- supplementation has been demonstrated to be very low (less than 10%) (Günalan et al., 2022; Muñoz et al., 2020).

The ergogenic effects of NO3- supplements are mediated by their capacity to increase nitric oxide (NO) via the NO3- – nitrite (NO2-) – NO pathway (Vanhatalo et al., 2018). The NO3- ingested is reduced to NO2- in the oral cavity by nitrate reductase produced by anaerobic microorganisms on the dorsal side of the tongue (Potter et al., 2001). This NO2- is subsequently reduced to NO by the actions of stomach acids, which is then absorbed in the gut (Raat et al., 2009). When passing into the systemic circulation, part of the NO2- is reduced to NO in acidosis (Modin et al., 2001) or in low oxygen bioavailability situations (Lundberg & Govoni, 2004). Muscle fibres are a NO2- reservoir (Ortiz de Zevallos et al., 2012), and the skeletal muscle experiences acidic and hypoxic conditions during muscle contractions (Rimer et al., 2016). Therefore, the NO3- – NO2- – NO pathway is potentiated during exercise, especially during high-intensity efforts (Domínguez et al., 2018).

BRJ is a dietary source rich in NO3- commonly used by athletes since it causes higher increases in NO levels compared to the same amount of NO3- provided in other types of dietary supplements such as salts (Bailey et al., 2017). The ergogenic effects of increased NO bioavailability are mediated by physiological mechanisms such as vasodilatory function, which enhances blood circulation to muscle tissue (Erzurum et al., 2007), gas exchange in muscle (Jones et al., 2018), mitochondrial efficiency (Stamler & Meissner, 2001), and muscle force production (Hernández et al., 2012; Ramos Álvarez et al., 2020).

Different studies have examined the effects of BRJ on sports performance, reporting positive effects on endurance (d'Unienville et al., 2021; Evangelista et al., 2024; Silva et al., 2022; Zamani et al., 2021) and high-intensity efforts (Aucouturier et al., 2015), including power and muscular strength (Bender et al., 2018; Mosher et al., 2016; Rodríguez-Fernández et al., 2021). However, the use of BRJ supplementation in team sports and sport-specific actions has been understudied.

In team sports, CMJ has been the most used test (Tingelstad et al., 2023), although sport-specific actions have been poorly studied. One of the most practiced team sports is volleyball, which is characterized by high demands on acceleration, agility, and jumping ability (Lidor & Ziv, 2010), specifically in sport-specific jumps like BJ and AJ (Goranovic et al., 2022).

Although sports nutrition guidelines promote evidence-based practice, women are usually underrepresented in randomized clinical trials. Smith et al. (2022) analyzed 1,826 studies with a total of 34,889 participants in studies that analyzed the effects of supported evidence-based supplements. The results found that only 23% of the participants were women. Specifically, in the case of BRJ, only 20.56% of the total sample were women, which could be considered an underrepresentation of female athletes in studies with acute and chronic NO3- supplementation (Wickham & Spriet, 2019).

Both men and women exhibit sex-specific physiological, morphological, and anthropometric characteristics that impact endurance and fatigue during exercise (Ansdell et al., 2019). These differences could lead to distinct responses to supplementation between sexes (Wickham & Spriet, 2019). Specifically, several factors suggest a potential attenuated effect of NO3- supplementation on exercise performance in women compared to men. Women experience greater endotheliumdependent dilation (Stanhewicz et al., 2018), potentially resulting in higher plasma NO2- concentrations due to increased oral activity of nitrate-reducing bacteria (Kapil et al., 2018). However, they also present a more oxidative muscle phenotype compared to men (Wickham et al., 2019), which could reduce the conversion of NO2- to NO and thus limit the benefits of NO3- supplementation. Moreover, women show a higher proportion of slow-twitch muscle fibres (Haizlip et al., 2015), which may attenuate the ergogenic response of BRJ because the ergogenic effect of this supplement has been related only to the fast-twitch muscle fibres (Jones, 2014). The ergogenic properties of BRJ reported in different meta-analyses on endurance performance variables (efforts with demands depending mainly on an oxidative metabolism) such as economy (d'Unienville et al., 2021) have not been found in any of the four studies carried out in samples formed only by women (Forbes & Spriet, 2021; Hogwood et al., 2023; Rienks et al., 2015; Wickham et al., 2019). However, studies focused on physical efforts with high glycolytic energy demands present contradictory results. For example, it has been reported a reduced time for covering a 500 m test in kayakers (Peeling et al., 2015) or repeated sprint in swimmers (Leitão et al., 2024), but not in a single 168 m swimming test (Lowings et al., 2017). Regarding the performance of power and explosive efforts characterized by a predominant metabolism of the high energy phosphagen system (Chamari & Padulo, 2015), some studies have reported an improved vertical jump ability (Jurado-Castro et al., 2022; López-Samanes et al., 2022), while others could not replicate those positive effects in female athletes (Hemmatinafar et al., 2023; López-Samanes et al., 2023). Volleyball athletes present a high proportion of type II muscle fibers (Hopwood et al., 2023) and are possibly more sensitive to the ergogenic effects of BRJ.

Despite the increasing research on the effects of beetroot juice (BRJ) supplementation on sports performance (Jones et al., 2018), the specific impact on team sports and sport-specific actions, such as volleyball-specific jumps, remains underexplored, especially in female athletes. Given that volleyball demands high-intensity efforts reliant on type II muscle fibers, which are particularly responsive to BRJ supplementation, this study aims to fill this research gap by focusing on high-performance female volleyball players. Furthermore, women are often underrepresented in studies on ergogenic aids like BRJ, leading to inconsistent findings regarding their physiological responses to such supplementation (Sims et al., 2023).

This underrepresentation underscores the need to investigate the specific effects of BRJ in women, particularly in sports that require explosive power and agility, to better understand the potential sex-specific responses to this supplementation (Larrosa et al., 2025). Therefore, the objective of this study was to analyze the effect of BRJ supplementation on the main variables of physical condition in high-performance female volleyball players (jumping ability, acceleration, and agility) and on specific jumping tests such as the BJ and AJ.

Materials and Methods

Participants

The sample of the present study consisted of 12 female competitive volleyball players (22.9 ± 3.62 years; 72.02 ± 7.22 kg; 175.64 \pm 6.7 cm; 23.33 \pm 1.96 kg/m² body mass index) from the second national division (Superliga 2) of Spain. The training and competitive experience was 7.1 \pm 2.69 years, and all participants trained at least five sessions weekly (including two hours of specific volleyball training from Monday to Friday and two strength training sessions on Tuesdays and Thursdays, which was mainly focused on core strength, lower limbs strength, and shoulder strength). The intervention was carried out during the competitive period of the 2021/2022 sports season in February 2022. Inclusion criteria were: i) training and competing uninterruptedly for six months before the intervention; ii) not taking any dietary supplements or medication during the previous six months; and iii) not having any cardiovascular, neuromuscular, or orthopedic disease that could alter the performance of the tests. All participants were informed of the study protocol and signed a consent form to participate in the study.

Experimental Design

A randomized cross-over double-blind placebo-controlled experimental design was used. Two familiarization sessions were performed at the same hour of the test. Then, two experimental sessions were conducted with seven days of washout (James et al., 2015). In each experimental session, 50% of volleyball players ingested a BRJ supplement (12.8 mmol of NO3-) and 50% of them ingested a depleted NO3- supplement as placebo (PLA) 2.5 hours before testing. Randomization was carried out using Research Randomizer software (www.randomizer.org). A vertical jump battery, agility, and acceleration test were performed after a specific warm-up. Figure 1 shows the experimental protocol.

Figure 1 Experimental Protocol

RANDOMIZED CROSS-OVER, DOUBLE-BLIND, PLACEBO-CONTROLLED EXPERIMENTAL DESIGN Randomized PLA (n=12) BRJ (n=12) <--2.5 hours before B 10 min: Warm-up (run + articular mobility + isometric exercise + jump + submaximal test) Vertical jump battery Acceleration Test Agility test Counter Movement Jump Block Jump Attack Jump Modified T-test 5-0-5 test Dav c 8 D 8 Test CMJ < 1min > CMJ BJ < 1min > BJ < 3min > AJ < 1min > AJ T-test < 1min > T-test < 3min > 5-0-5 < 1min > 5-0-5 < 3min > < 3min >

Note. PLA: Placebo; BRJ: Beetroot juice; CMJ: Countermovement jump; BJ: Block jump; AJ: Attack jump.

Beetroot Juice Supplementation and Dietary Control

On testing days, each participant ingested 140 ml of BRJ (12.8 mmol NO3- Beet-It-Pro Elite Shot; James White Drinks Ltd., Ipswich, UK) or 140 ml of PLA (0.01 mmol NO3-) (Domínguez et al., 2017). Twenty-four hours before the tests, participants

followed recommendations for a structured diet with a similar macronutrient distribution (60% carbohydrates, 10% proteins, and 30% fats) without NO3- dietary sources, caffeine, and alcohol. Additionally, participants were instructed to avoid brushing their teeth, using antibacterial mouthwashes (Casado et al., 2021), or chewing gum to avoid NO3- reduction by oral bacteria (López-Samanes et al., 2023).

Instruments and Procedure

Vertical Jump

Three vertical jump tests were used for assessing jump ability: CMJ, BJ, and AJ. An infrared system (Optogait Modular System, Microgate, Bolzano, Italy; software version 1.13.24) with excellent validity and reproducibility was used (Glatthorn et al., 2011). Two jump attempts were performed, separated by one minute of recovery between repetitions and three minutes between the different types of jumps. The highest value in centimetres of the two attempts was registered.

For CMJ, we instructed the participants to stand upright with their hands on their hips. Participants executed a controlled leg flexion until a 90° angle followed by a synchronized and explosive movement of leg extension (concentric action) with the aim of reaching the maximum height possible. During the flight phase, the knees should be fully extended and start the ground contact with the toes while hand were kept in their hands to avoid any sideways forward/backward movements (Garnacho-Castaño et al., 2015). For the BJ, participants initiated the jump with their hands in front of their chest and leg extended before performing an eccentric phase in the legs similar to CMJ, however with the hand in front of their chest. Then, participants executed an explosive concentric contraction of the legs with the intention of reaching the maximum height possible. Therefore, in the maximal height, the hands were stretched up and forwards at the peak height, resembling a volleyball block motion (Amasay, 2008; Sattler et al., 2015). Similarly to BJ, AJ mimic a specific volleyball action as attack. For that, participants executed a series of three preceding steps and they were instructed for using an arm swing as they perform for the spiking actions during a real game context; however the aim of the AJ was to reach the maximum vertical jump avoiding sideways forward/backward movements (Pérez-López et al., 2015; Sattler et al., 2015).

Acceleration and Agility

For the agility assessment, a modified T-test was used. Participants began the test 30 cm behind the start line to prevent early triggering of the timing gates (Pleša et al., 2022). For assessing acceleration, a 5-0-5 test was performed using the modified (standing start) version (Gallo-Salazar et al., 2017). The time was recorded using a photoelectric cell gate (Witty Gate, Microgate, Bolzano, Italy) placed 0.9 m above the ground level, placed at the start and finish line in both tests. The athletes' timing data were obtained using the wireless stopwatch integrated with the photoelectric cells and subsequently recorded manually on paper.

Statistical Analysis

A Shapiro-Wilk test was performed to check that all the variables were adjusted to the pattern of the normal distribution. Therefore, for comparing the possible effect of BRJ, a t-test for paired samples was performed for each variable assessed. As a complementary analysis, effect size (ES) using Cohen's d (*d*) was calculated with the interval of confidence (95%). ES was considered as trivial (d < 0.19), small (d = 0.2 to 0.49), moderate (d = 0.5 to 0.79), and large ES (d > 0.8) (Cohen, 1992). Statistical differences were fixed as p < .05. Jamovi software (version 2.3.28) was used for calculating all the statistical treatments.

Results

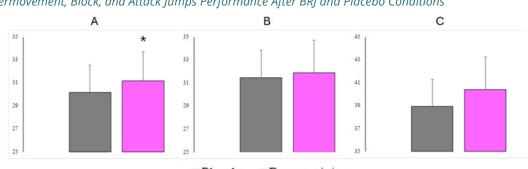


Figure 2 Countermovement, Block, and Attack Jumps Performance After BRJ and Placebo Conditions

Note. Data presented as mean \pm standard deviation. A: CMJ (cm); B: BJ (cm); C: AJ (cm); *: statistical differences (p < .05) between experimental conditions.

Placebo Beetroot juice

Vertical Jump

The results reported an improved CMJ performance after BRJ compared to PLA (BRJ: 31.18 ± 2.53 cm vs. PLA: 30.16 ± 2.38 cm; t = 2.273; p = .044; d = 0.66 [from 0.02 to 1.27]) (see Figure 2A). Regarding the specific volleyball jumps, no effect was found for BJ (BRJ: 31.89 ± 2.84 cm vs. PLA: 31.48 ± 2.37 cm; t = 0.811; p = .435; d = 0.23 [from -0.35 to 0.80]) (see Figure 2B), while a trend towards significant differences with a moderate ES was found in AJ (BRJ: 40.40 ± 4.0 cm vs. PLA: 38.92 ± 3.77 cm; t = 1.971; p = .074; d = 0.57 [from -0.05 to 1.17]) (see Figure 2C).

Acceleration and Agility

In the assessment of acceleration capability, no differences were found between BRJ and PLA (BRJ: 2.92 ± 0.15 s vs. PLA: 2.90 ± 0.21 s; t = 0.794; p = .469; d = 0.21 [from -0.63 to 0.78]). Neither was any effect of BRJ reported in the T-test (BRJ: 6.36 \pm 0.36 s vs. PLA: 6.36 ± 0.35 s; t = 0.037; p = .972; d = 0.01 [from -0.55 to 0.58]).

Discussion

The goal of the present study was to analyze the effect of BRJ supplementation on jumping ability, acceleration, and agility in high-performance female volleyball players. The main findings are that BRJ does not affect acceleration capability and agility in female volleyball players but could act as an ergogenic aid in vertical jump ability. Therefore, BRJ has demonstrated improvement in performance in a non-specific volleyball jump such as CMJ but not in specific volleyball jumps such as BJ and AJ.

The positive effects of BRJ supplementation on CMJ found in female volleyball players in this study (3%) are in line with the results of previous research in female sports (Jurado-Castro et al., 2022; López-Samanes et al., 2022). In a sample composed of women highly trained in resistance exercises (four years of training experience), Jurado-Castro et al. (2022) reported a 6% improvement in CMJ, while similar enhancements after 140 ml BRJ (12.8 mmol of NO3-) in CMJ (7%) were obtained in a sample of female semi-professional rugby players (López-Samanes et al., 2022). In opposition, López-Samanes et al. (2023) could not detect any effect of BRJ (6.5 mmol NO3-) in female elite hockey players. These differences could be attributed to two possible factors: i) the sample analyzed; ii) the posology used. Hockey is a sport modality that combines explosive efforts with prolonged endurance and involves a mixed metabolism (Reilly & Borrie, 1992), similar to other sport modalities such as soccer. At a histological level, semi-professional rugby players are characterized by a significantly higher proportion of type II motor units (54.6%) compared to soccer players (27.5%–55%) (Hopwood et al., 2023). Considering the specific ergogenic effects of BRJ on type II muscle fibers (Jones, 2014), an ergogenic effect on CMJ could be appreciated in female athletes in sport modalities characterized by a predominance of type II muscle fibers such as volleyball, rugby, or resistance training but not in endurance or sport modalities that involve a mixed metabolism. Another factor that could explain the different results of López-Samanes et al. (2023) is the posology used. Firstly, Lundberg et al. (2008) observed a proportional response of increased NO levels with the NO3- ingested; secondly, Wylie et al. (2013) established a relationship between the amount of NO3- ingested and the ergogenic effect of BRJ. They found positive effects on physical performance after ingesting 8.4 and 16.8 mmol of NO3- but not 4.2 mmol. This response suggests a possible threshold for detecting ergogenic properties of BRJ supplementation. Therefore, the dosage of NO3- used by López-Samanes et al. (2023) (6.4 mmol) could be insufficient for detecting any effect on CMJ.

In contrast with CMJ, BRJ supplementation was not associated with any effect in sport-specific volleyball jumps (BJ and AJ). An enhancement of CMJ is associated with an increase in power production of lower limbs (Rodacki et al., 2002). Therefore, the assessment of CMJ is frequently used as an indirect parameter of sport performance (Jiménez-Reyes et al., 2014). Nevertheless, in a sample formed by competitive female volleyball players, Ahmadi et al. (2021) detected a positive effect of a plyometric training program on CMJ but not on AJ. Similarly, in female trained swimmers, an increased CMJ performance in response to a resistance training program had no effect on a specific sport action such as a swimming start (Breed & Young, 2003). These results suggest that CMJ could be more sensitive to detecting changes in neuromuscular performance compared to specific actions of sports in trained female athletes. However, an alternative explanation could be attributed to the sensitivity of different tests for detecting statistical differences. Thus, CMJ involves only the power production of lower limbs (Rodacki et al., 2002), while specific volleyball jumps such as AJ include the contribution of the mechanical impulse of upper limbs and trunk to the jumps, requiring complex intersegmental coordination (Fuchs et al., 2019). This more complex intersegmental coordination increases the coefficient of variation in AJ compared to CMJ in our study (> 20% in AJ vs CMJ). Based on the lower coefficient of variation found in our and other studies, CMJ could be a more suitable test for detecting small changes in neuromuscular function (Klavora, 2000). In fact, in the only study that has analyzed the effect of BRJ in volleyball players, no effect was detected in vertical jump (Sargent test) with a very high (17%) coefficient of variation (Hemmatinafar et al., 2023).

In agreement with previous studies, BRJ did not impact agility and acceleration. Regarding agility, López-Samanes et al. (2022) did not detect any effect in a modified T-test in female rugby players. Regarding acceleration, no effect of BRJ could be detected in the 10 m, 20 m, or 30 m test in either female rugby (López-Samanes et al., 2022) and hockey players

(López-Samanes et al., 2023) on repeated sprint ability in female team sport modalities (Buck et al., 2015), or on intermittent swimming sprints (Jonvik et al., 2018). The results of the present study strengthen the absence of any effect of BRJ in agility and acceleration capability in female athletes.

This study responds to a need for carrying out studies that analyze the effects of BRJ in female athletes. Wickham and Spriet (2019) reported that only 20.56% of the participants in studies of evidence-based BRJ supplementation were female. This need is more pronounced in explosive efforts because women were only 7% of the total sample included in a systematic review (Tan et al., 2022). Based on the results of a meta-analysis, BRJ has been proposed as ergogenic for enhancing economy (d'Unienville et al., 2021), although four of the five studies (Forbes & Spriet, 2021; Hogwood et al., 2023; Ortiz de Zevallos et al., 2023; Wickham et al., 2019) carried out in samples formed exclusively by women failed to detect any ergogenic effect. This sex-mediated different response to BRJ supplementation could be attributed to a more oxidative muscle phenotype in females compared to males (Wickham et al., 2019), which could reduce the conversion of NO2- to NO and thus limit the benefits of NO3- supplementation. In addition, the higher proportion of type I muscle fibers (Haizlip et al., 2015), improved ATPase efficiency (Harmer et al., 2014), mitochondrial content (Miotto et al., 2018), and tissue perfusion (Ortiz de Zevallos et al., 2023) could limit the sensitivity of female endurance athletes to the ergogenic properties of BRJ. Nevertheless, the present study showed that BRJ could act as an ergogenic dietary supplement for enhancing neuromuscular performance in female athletes of sport modalities characterized by a predominance of type II muscle fibers such as volleyball.

While this study provides valuable insights into the effects of BRJ supplementation on female volleyball players, several limitations must be acknowledged. Firstly, the small sample size may limit the generalizability of the findings to a broader population of female athletes. Secondly, the study did not control for the participants' menstrual cycle, which is particularly important given that hormonal fluctuations throughout the cycle can influence factors such as muscle contractility, fatigue, and vascular function. These physiological changes may have impacted the response to BRJ supplementation and could account for some variability in the results. Future studies should consider controlling for the menstrual cycle to better understand the potential sex-specific effects of nitrate supplementation (Larrosa et al., 2025; Sims et al., 2023). Lastly, while this study focused on explosive efforts, further research is needed to examine the effects of BRJ supplementation on other performance variables, including endurance and recovery, across different phases of the training cycle.

Conclusions

The present study contributes to the literature on the effects of BRJ supplementation in female athletes, providing insight into its potential to improve neuromuscular performance in non-specific tests. The limited previous research on BRJ in the female population suggests a limited effect of this supplement on sport performance in female athletes, with a possible sex-mediated different response. Despite the limited evidence from studies focused on explosive efforts in female athletes, the results of the present study indicate ergogenic properties of BRJ in non-specific jumping ability in athletes with a predominance of type II muscle fibers. These ergogenic effects could be blunted in sport-specific actions.

These findings underscore the complexity of the relationship between enhancements in motor performance and sportspecific actions, particularly in highly trained athletes. Further research is needed to fully understand the impact of BRJ supplementation on sport-specific actions in female athletes, as well as the possible underlying physiological mechanisms.

Ethics Committee Statement

The study was conducted in accordance with the Declaration of Helsinki and was approved by the Ethics Committee of Universidad Isabel I (code UI1-PI002; approval date November 12th, 2019).

Conflict of Interest Statement

The authors declare no conflicts of interest. The funding entities or institutions had no influence on the design of the study, data analysis, or interpretation of the results.

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Authors' Contribution

Conceptualization I.L.L., J.M.L., A.J.S.O. & R.D.; Methodology A.J.S.O., A.M.Lo. & R.D.; Software A.M.Lo., A.M.LI. & R.D.; Validation A.J.S.O., A.M.Lo. & R.D.; Formal Analysis I.L.L., J.M.L., E.R.S. & R.D.; Investigation I.L.L., J.M.L., A.J.S.O., E.R.S., A.M.Lo., A.M.LI. & R.D.; Resources I.L.L., J.M.L. A.M.Lo. & A.M.LI.; Data Curation A.M.Lo. & R.D.; Writing – Original Draft I.L.L., J.M.L.,

A.J.S.O. & R.D.; Writing – Review & Editing A.J.S.O., A.M.Lo. & A.M.Ll.; Visualization A.J.S.O., A.M.Lo. & R.D.; Supervision A.J.S.O., A.M.Ll. & R.D.; Project Administration A.M.Ll.; Funding Acquisition A.M.Ll. All authors have read and agreed to this published version of the manuscript.

Data Availability Statement

Data available upon reasonable request to corresponding author (amllerena@us.es).

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