ACUTE EFFECTS OF TERERÉ (ilex paraguariensis) INGESTION ON HYDRATION PARAMETERS IN RUNNERS

EFEITOS AGUDOS DA INGESTÃO DE TERERÉ (*ilex paraguariensis*) SOBRE PARÂMETROS DE HIDRATAÇÃO EM PRATICANTES DE CORRIDA

Carolina Rocha Diniz ¹, Raphael de Jesus Brittes¹, Fabiane La Flor Ziegler Sanches¹, Cássio Pinho dos Reis¹, Christianne de Faria Coelho-Ravagnani¹

¹ Federal University of Mato Grosso do Sul, Campo Grande-MS, Brazil.

ABSTRACT

Objectives: To evaluate the effects of pre-exercise Tereré consumption on hydration indicators in runners. **Methodology:** This randomized, crossover, double-blind study included 19 street runners (10 M: 31.3 ± 8 years, 15.9 ± 4.2 % body fat, and 9 F: 32.8 ± 5.1 years, 20.5 ± 7 % body fat), divided into an Experimental Tereré Group (TrEX), which consumed traditional Tereré (50g of yerba mate infused in 6ml/kg of body weight of cold water at $\pm 10^{\circ}$ C), and a Placebo Group (TrPL), which received the same amount of water but with decaffeinated yerba mate. Anthropometric measurements (body mass and height) were collected at baseline. Subsequently, 45 minutes after ingesting the drinks, body mass, total body water (TBW), intracellular water (ICW) and extracellular water (ECW) (by electrical bioimpedance), urine specific gravity (UEG), and urine color (UC) measurements were taken. At the end of the run, the thirst scale and thirst sensation questionnaires were applied, and the UEG and UC were collected again. **Results:** The consumption of Tereré did not alter TBW, ICW, ECW, UEG, or UC compared to the TrPL in either sex (p>0.05). All athletes remained hydrated after the run. **Conclusion:** Tereré did not change the hydration parameters of the runners when compared to decaffeinated Tereré, and both drinks were able to maintain a good hydration status in the runners after the physical effort. Tereré could be offered as a nutritional strategy for hydrating street runners in races lasting under 30 minutes.

Key-words: Ilex Paraguariensis. Yerba Mate. Hydration. Street Running.

RESUMO

Objetivo: Avaliar os efeitos do consumo de Tereré sobre a hidratação de praticantes de corrida. Metodologia: Neste estudo randomizado, cruzado, duplo cego, foram selecionados 19 corredores de rua, (10 homens: 31.3 ± 8 anos, 15.9 ± 4.2 % de gordura e 9 mulheres: 32.8 ± 5.1 anos, 20.5 ± 7 % de gordura), divididos em Tereré Experimental (TrEX) que consumiu 50g de erva-mate (EM) infundida em 6mL/kg do peso corporal de água a $\pm 10^{\circ}$ C e Tereré Placebo (TrPL), que recebeu a mesma quantidade de água em descafeinada. No baseline, os participantes realizaram medidas de massa corporal e estatura. Após 45 minutos da ingestão das bebidas, foram feitas medidas de massa corporal, água corporal total (ACT), intracelular (AI) e extracelular (AE) (por bioimpedância elétrica), gravidade específica da urina (GEU) e coloração da urina (UC). Ao término da corrida (20 a 27 minutos), foram analisadas escalas de sede, GEU e UC. Resultados: A ingestão de Tereré não alterou a ACT, AI, AE, GEU e a UC, em relação ao TrPL em ambos os sexos (p>0.05). Após a corrida, todos os atletas permaneceram em bom estado de hidratação. Conclusão: O Tereré não alterou os parâmetros de hidratação dos corredores quando comparado ao Tereré descafeinado, sendo que ambas as bebidas foram capazes de manter um bom estado de hidratação dos atletas após o esforço físico. O Tereré pode ser uma estratégia nutricional para a hidratação de corredores de rua em corridas inferiores a 30 minutos.

Palavras-chaves: Ilex Paraguariensis. Erva-Mate. Hidratação. Corrida De Rua.

Introduction

Hydration has a great influence on the physical performance of runners and should be encouraged in different periods of exercise, including pre, during, and post-training/competition. Hydration guidelines suggest the intake of 3 to 7 mL/kg of body weight, between 2 to 4 hours before exercise¹, however, runners generally consume less fluids than recommended.

Losses of 2% of body mass are already enough to cause impairments in physical performance² and there is some evidence that even low levels of dehydration may impair performance, including during relatively short-duration exercises³. Hypohydration impairs exercise through a series of mechanisms, that include reduced plasma/blood volume, cardiovascular function, muscle blood flow, thermoregulatory capacity, and neuromuscular



Page 2 of 14 Diniz et al.

function, and increased psychological tension⁴. Therefore, preventing dehydration during exercise is one of the most effective ways to maintain good physical performance.

The most important ergogenic aid for athletes is water; however, the addition of substances to water (e.g., carbohydrates and caffeine) may provide additional benefits to physical performance, depending on factors such as the type and duration of the exercise. Carbohydrate-containing drinks, for example, are mainly indicated during exercises lasting longer than 1 hour, while caffeine-containing drinks are considered to be potentially ergogenic if consumed pre-exercise. Although ergogenic, caffeine has a dose-dependent diuretic action, that is, while doses between 300–500 mg do not modify diuresis⁵, doses above 500 mg seem to promote significant diuresis⁶, which could lead to the risk of fluid deficits in runners. Thus, considering the concentration of caffeinated beverages is important.

Tereré is a beverage based on yerba mate (Ilex paraguariensis), that is widely consumed in Paraguay and in the Central-West region of Brazil. Tereré contains caffeine, purine alkaloids, phenolic compounds, and saponins, and presents potential ergogenic, anti-inflammatory, and thermogenic effects^{7,9}. In previous studies, yerba mate supplementation was able to alter the kinetics of energy metabolism during exercise testing, reduce body weight and fat mass¹⁰, improve lipid parameters¹¹, reduce perceived exertion and improve satiety^{12,13}, and improve performance in time-trial tests^{14,15}.

Unlike chimarrão, which is consumed with hot water, Tereré is consumed with cold water, through a straw, and served gradually, in approximately 6 to 10 servings, with about 50 to 100 mL of water in each portion. It is worth mentioning that both the temperature and the amount of water usually consumed in Tereré are consistent with the pre-training hydration guidelines proposed by Med Sci Sports Exerc¹, which recommend the intake of approximately 500 mL of fluids, adjusted to body weight, approximately 2 hours before physical exercise. In addition, the hydrating and thermoregulatory potential of this beverage for runners is highlighted. However, surprisingly, no studies have yet investigated the effects of Tereré on hydration status, especially in athletes. A survey conducted with 240 athletes (including 102 street runners) from Mato Grosso do Sul showed that 60% of respondents consumed Tereré, and 50% reported consuming Tereré at some point during training; 17.4% before training, 17.4% during training, and 65.2% after training. In addition, a large portion of the athletes (43%) reported hydration as the reason for consumption (data not yet published).

Due to its vasodilatory^{16,17} and hydrating effects (since it is consumed with cold water), it is speculated that yerba mate in the form of Tereré could act positively on total and intracellular body water content. This, in turn, could facilitate blood flow, body temperature control, reactions related to energy production, and the elimination of metabolic by-products during physical exercise. However, due to its caffeine content, it is also speculated that Tereré may influence hydration status.

In view of the above, the following question was raised: does the consumption of Tereré before physical effort improve the hydration indicators of runners compared to the consumption of decaffeinated Tereré?

The hypothesis of the current study is that Tereré consumption may be as effective as decaffeinated Tereré in promoting hydration, since the beverage contains moderate amounts of caffeine, which would not impair hydration. Therefore, the objective of the current research is to evaluate the effects of Tereré consumption before physical effort on hydration indicators in runners.

Methods

Participants

Street runners from training teams in the city of Campo Grande, Brazil, were initially invited to participate in this study through their coaches or directly via phone call, text message,

social media, and/or in person. One participant was excluded for not attending the second day of testing. Thus, 19 street runners, ten men and nine women, were included in the final analysis. Inclusion criteria for the study were: a) age between 20 and 50 years, b) running for at least one year, and c) not consuming caffeine-containing foods or supplements in the 24 hours prior to testing. Exclusion criteria were: a) smoking, b) musculoskeletal injuries in the six months prior to the end of testing, c) history of heart disease, and d) not complying with pre-test protocols and/or not attending the second data collection within the stipulated period (7 to 14 days). Participants' characteristics are described in Table 2.

The athletes were fully informed about the study's objectives, risks, and benefits. Those who agreed to participate signed two copies of the Informed Consent Form (ICF); one copy was retained by the participant and the other by the researcher. The study was approved by the Ethics Committee of the Federal University of Mato Grosso do Sul, under opinion No. 6.010.210.

Procedures

In this double-blind, randomized, crossover study, all participants attended two identical test days, separated by a 7 to 14-day interval, with only the test beverage differing. Tests were conducted between April and May 2024, always on Saturday mornings, at scheduled times, in the Exercise Physiology Laboratory of the Federal University of Mato Grosso do Sul, Campo Grande, Brazil.

Participants were randomly assigned to ingest Tereré (TrEX) or placebo (TrPL). The TrEX contained 50g of yerba mate, and was served in an aluminum cup, with 60 to 100mL of cold filtered water (10°C) infused for each round of sips, through a straw, until reaching the amount of 6mL of water/kg of the athlete's body mass. The same amount of cold water was provided in the placebo condition, but using 50g of washed and decaffeinated yerba mate, with similar visual and organoleptic properties to the traditional herb. Sips were taken immediately after each cold water infusion into the ready-to-consume yerba mate. Considering the total water provided to the group, approximately 4 to 7 infusions per athlete were performed. Participants were instructed to consume the beverages within 10 minutes and not to comment on the taste or perceived physiological effects with other participants.

The preparation and delivery of the study beverages and the randomization of conditions were carried out by a researcher not involved in data collection, using the software www.sealedenvelope.com, which generated random codes with a 50% chance of ingestion of TrEX or TrPL. In addition to the athletes, both the evaluators and the statistician were blinded to the two experimental conditions.

Before visiting the laboratory, participants were informed about the tests and instructed to maintain their normal dietary routine but avoid consuming any caffeine- or yerba mate-based stimulant foods or supplements and not to perform training or physical exercise for 24 hours before the first test day. On the test day, they signed the ICF and answered the training questionnaire, after which they were randomly assigned to the experimental conditions (TrEX or TrPL). Athletes consumed a standardized meal (cookies and juice) containing 0.75g to 1g of carbohydrate/kg of body mass and then remained in total fasting (no food or water) for 2 hours before testing.

After the 2-hour fast, weight and height were measured using a scale with 100g precision and 200kg maximum capacity and a stadiometer from the LIDER® brand, model P200C, blood pressure, with an automatic monitor from Omron®, and body composition and hydration (total, intra- and extracellular water) using an octapolar analyzer (InBody® S10). After baseline measurements, participants ingested one of the beverages (TrEX or TrPL) and, while waiting for the physical test, answered a 24-hour dietary recall and an adapted caffeine consumption questionnaire to assess energy and nutrient intake on the days before the test.

Page 4 of 14 Diniz et al.

After 45 minutes of Tereré consumption, new measurements of body water were taken, along with urine collection for specific gravity and color analysis. At the end of the run, athletes were weighed again, answered thirst and thirst sensation questionnaires, and another urine collection was performed. Figure 1 presents the procedures adopted in the study.

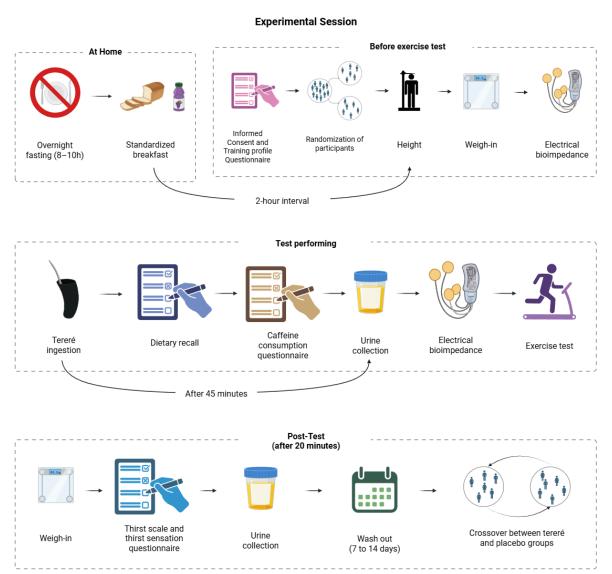


Figure 1. Overview of the study procedures **Source: authors.**

Acquisition and analysis of yerba mate

The yerba mate used in this study was donated for research purposes by the company "BARÃO INDÚSTRIA E COMÉRCIO DE ERVA-MATE S/A", batch No. 080922, from the region of Rio Grande do Sul, Brazil. The amount of caffeine in the yerba mate was analyzed at the laboratories of the Faculty of Pharmaceutical Sciences, Food and Nutrition - FACFAN of UFMS. To verify the caffeine content in Tereré, the High-Performance Liquid Chromatography (HPLC) technique was used, adapted from Vieira (2015). The extraction method was performed cold, based on the solubility of caffeine in water. The caffeine solution was prepared from 50g of yerba mate, which was transferred to a 500 mL volumetric flask and filled with cold water (10°C). Subsequently, a 1 mL aliquot was filtered using a syringe filter with a cellulose acetate membrane and a pore diameter of 0.22 μ m. Aliquots of 20 μ L were injected into the Ultimate 3000 liquid chromatograph, from Thermo Scientific®, equipped with a Supelco Discovery®

column (10 cm x 4.6 mm). The mobile phase consisted of a mixture of acetonitrile and water (32:68, v/v) with pH 7.5. The elution was performed isocratically, with a flow rate of 1.0 mL/min, and detection occurred at 273 nm. For caffeine quantification, a calibration curve was used with a caffeine standard (99.8% purity). The final concentration was determined by the average of three consecutive injections, and the values were expressed in μ g/mL. Thus, a caffeine content of approximately 44 mg/50 g of yerba mate/Tereré was obtained.

Anthropometric measurements

Body mass and height measurements were taken using a scale with a stadiometer from the LIDER® brand, model P200C, with a precision of 100g and maximum capacity of 200kg. For body mass measurements, participants wore minimal clothing (shorts for men, top and shorts for women), and were barefoot. The measurements were performed after physical effort, when the athletes wiped off the excess sweat from their skin with a towel provided by the research team and were instructed to remove all objects containing metal or relevant weights¹⁹.

Electrical Bioimpedance

Electrical bioimpedance (InBody® S10) was used to assess the athletes' body composition, including intracellular water (ICW), extracellular water (ECW), and total body water (TBW), at a frequency of 50 kHz of electric current. Before the procedure, participants were instructed to empty the bladder, remove any objects containing metal or weight, and wear minimal clothing (shorts for men, top and shorts for women), with bare feet. After removing excess sweat from the skin with a towel, they were instructed to lie on their backs, with legs extended along the midline of the body and hands facing down. After cleaning the skin and tactile electrodes with alcohol, the electrodes were placed on the right and left hands (on the middle finger and thumb), and on the inner and outer sides of the right and left ankles, according to the procedures described by the manufacturer²⁰.

Questionnaires (Thirst scale and thirst sensation)

Thirst was measured using a Likert scale ranging from 1 to 9, where 1 means "not thirsty at all," 3 "a little thirsty," 5 "moderately thirsty," 7 "very thirsty," and 9 "extremely thirsty" 21.22.

Thirst sensation was measured using a visual analog scale that displays a 10 cm horizontal line with the following anchored questions at the ends: How thirsty are you right now? (not thirsty – extremely thirsty); How pleasant would it be to drink some water right now? (very unpleasant – very pleasant); How dry is your mouth right now? (not dry at all – extremely dry); How would you describe the taste in your mouth? (normal – very unpleasant); How full is your stomach right now? (not full at all – very full)²³.

Urine measurements

Urine samples were classified according to color and specific gravity. Each athlete received a disposable, sterilized plastic container. Participants were instructed to collect approximately 30 mL of urine for analysis and place the containers in a paper bag. Urine analysis was performed by a single evaluator immediately after collection.

Urine Specific Gravity (USG) was analyzed using a portable refractometer (RTP-20ATC, INSTRUTHERM, BRAZIL), and the collection was carried out according to procedures described in the literature²⁴. Evaluation was based on the following measures: values between 1.002 to $1.010\mu G$ were considered normal or hydrated, values between 1.011 to $1.020\mu G$ as minimal dehydration, 1.021 to $1.029\mu G$ as moderate dehydration, and $>1.030\mu G$ as severe dehydration¹⁹,²⁵.

Urine color was classified using a chart with eight shades of urine, where tone 1 is the lightest and tone 8 the darkest. The analysis was performed by comparing the sample (urine)

Page 6 of 14 Diniz et al.

with the colors on the chart²⁶. To determine each athlete's hydration status, the cut-off points proposed by Casa et al.²⁶ were adopted, as shown in Table 1.

Table 1. Hydration status indices.

Hydration status	Body weight variation (%)	Urine color	Urine Specific Gravity (USG)
Hydrated	+1 to -1	1 or 2	<1010
Minimal dehydration	-1 to -3	3 or 4	1010 - 1020
Moderate dehydration	-3 to -5	5 or 6	1021 - 1030
Severe dehydration	>-5	>6	>1030

Source: National Athletic Trainer's Association – NATA - CASA et al²⁶

Sweat rate

The sweat rate was calculated using the formula developed by Hamouti et al.²⁷, where Sweat rate (liters.h⁻¹) = (Change in body mass in kg + Fluid intake in liters) / duration of activity (h).

Exercise test

The total exercise session lasted between 25 and 30 minutes. The run was performed on a treadmill from the Inbramed brand, model Top18. Participants began with a light warm-up of 3 minutes, which included stretching and other exercises the runners were familiar with. The experimental protocol was then applied in three stages: the first consisted of a 3-minute walk at 5 km/h; the second stage involved a steady-state run for 10 minutes at 85% of the comfortable long-run pace reported by the athlete; and in the third and final stage, after the initial 10 minutes, the speed was increased by 1 km/h every 120 seconds until the athlete reached exhaustion or met at least two of the criteria described by the National Ergometrics Consensus guidelines²⁸, including: 1) Sustained increase in RQ above 1.18; 2) Heart rate above 105% of the value predicted by the formula (220 - age); 3) Sustained oxygen consumption plateau; 4) Chest discomfort or dyspnea disproportionate to the effort intensity; 5) Monitoring system failure; or 6) Treadmill operational limit.

Statistical analysis

The statistical analysis of the data was performed using R software, version 4.4.1. Descriptive analyses were calculated and the data are expressed as mean and standard deviation. The comparison between the TrEX and TrPL groups was performed using the paired Student's t-test for parametric data (thirst sensation questionnaire – question 3). Normality was assessed by the Shapiro-Wilk test (W). Non-parametric data (thirst scale, thirst sensation questionnaire – questions 1, 2, 4, 5) were compared using the Wilcoxon test. A two-way repeated measures ANOVA was used to assess interactions between conditions and time points for normally distributed variables, in this case, body mass, TBW, ICW, ECW, and urine color (for females), followed by the Bonferroni post hoc test. The Friedman test was applied to compare urine color between groups, as these data were considered non-parametric. The significance level for all analyses was set at p < 0.05. G*Power software was used to calculate the required sample size considering an alpha level of 0.05 and power of 0.8, with an a priori effect size of 0.9 for a significant difference in a two-way repeated measures ANOVA, resulting in 10 athletes.

Results

Table 2 presents the physical and sports characteristics of the evaluated group, including age, anthropometric measurements, clinical parameters, and training data.

Table 2. Characteristics of the Athletes

Variables	Male ± SD	Female ± SD	
N	10	9	
Age (years)	31.3 ± 8	32.8 ± 5.1	
Weight (kg)	74.4 ± 10.2	56.8 ± 6.4	
Height (cm)	173.5 ± 5.8	163.2 ± 4.5	
Resting HR (bpm)	58.8 ± 14.84	61.12 ± 11.09	
Resting SBP (mmHg)	126 ± 13.08	114.65 ± 13.89	
Resting DBP (mmHg)	70.15 ± 17.45	68.11 ± 10.15	
Body Fat (%)	15.94 ± 4.24	20.55 ± 7	
Practice time (years)	2 ± 1.17*	3 ± 0.87*	
Weekly training frequency (days)	3.5 ± 0.45 *	4 ± 1.22	
Daily training duration (hours)	1 ± 0.36*	1.32 ± 0.56	

Note: *Median ± Standard error

Source: authors

Table 3 presents the hydration parameters of the athletes. There was no statistical interaction between the effects of beverage ingestion and time for body mass in the female athletes (F(1, 16) = 4.72, p = 0.45). Furthermore, neither beverage ingestion (F(1, 16) = 0.74, p = 0.85) nor time (F(1, 16) = 2.78, p = 0.75) showed statistically significant effects on changes in body mass. The same behavior was observed for men, with no statistical differences in body mass for the ingestion \times time interaction (F(1, 16) = 0.63, p = 0.56), beverage ingestion (F(1, 16) = 0.00, p = 0.97), or baseline compared to post-exercise (F(1, 16) = 2.87, p = 0.34).

Regarding total body water, no statistically significant interaction was found between Tereré ingestion and time for either women (F(1, 16) = 0.12, p = 0.73) or men (F(1, 18) = 1.89, p = 0.69). Neither time (F(1, 16) = 3.50, p = 0.08 for women; F(1, 18) = 0.68, p = 0.18 for men) nor beverage ingestion had a statistically significant effect on total body water (F(1, 16) = 0.03, p = 0.95 for women; F(1, 18) = 0.01, p = 0.97 for men).

The ingestion \times time interaction was also not significant for intracellular water in women (F(1, 16) = 0.08, p = 0.93), neither were the isolated effects of beverages (F(1, 16) = 0.05, p = 0.94) nor time (F(1, 16) = 1.48, p = 0.24). The same occurred for men, for the ingestion \times time interaction (F(1, 18) = 0.17, p = 0.68), beverage ingestion (F(1, 18) = 0.07, p = 0.98), and time points baseline and 45 minutes after beverage ingestion (F(1, 18) = 0.65, p = 0.20).

The ingestion \times time interaction was not significant for extracellular water in women (F(1, 16) = 0.96, p = 0.34), and neither were the isolated effects of beverages (F(1, 16) = 0.01, p = 0.98) nor time (F(1, 16) = 0.52, p = 0.36). The same occurred for men: ingestion \times time

Page 8 of 14 Diniz et al.

interaction (F(1, 18) = 0.16, p = 0.70), beverage ingestion (F(1, 18) = 0.00, p = 0.96), and time (F(1, 18) = 0.50, p = 0.38).

Regarding urinary hydration indicators, Table 3 shows that the beverage ingestion \times time interaction was not statistically significant (F(1, 16) = 0.52, p = 0.48) for USG in women. The condition in which the participant was allocated was also not significant (F(1, 16) = 0.53, p = 0.48); however, time had a significant effect (F(1, 16) = 10.01, p < 0.01), i.e., there was a reduction in USG between post-beverage and post-exercise time points in both conditions (TrEX and TrPL). The same behavior was observed in men: beverage ingestion \times time interaction (F(1, 16) = 3.24, p = 0.86), condition effect (F(1, 16) = 1.70, p = 0.2), but the time effect was significant (F(1, 16) = 15.70, p < 0.01).

Urine color in the female runners showed no statistically significant changes for the ingestion \times time interaction (F(1, 16) = 0.53, p = 0.48), beverage ingestion (F(1, 16) = 0.04, p = 0.85), or time (F(1, 16) = 1.47, p = 0.24). For men, there was no difference between the two time points (p = 0.13). Moreover, in both sexes and conditions (TrEX and TrPL), the athletes can be classified as having "minimal dehydration".

Table 3. Changes in body mass, total, intra and extracellular body water, urine specific gravity, and urine color of runners after the consumption of Tereré and placebo Tereré (Females, N = 9; Males, N = 10).

Variables	Condition	Baseline	Post beverage	Post exercise	p	F
Weight(kg)	TrEX Fem	57.0±7.0 ^{Aa}	-	56.6±6.5 ^{Aa}	0.5	4.7
	TrPL Fem	56.8±6.6 ^{Aa}	-	56.7±6.6 ^{Aa}		
	TrEX Masc	74.5±10.6 ^{Aa}	-	74.1 ±10.4 ^{Aa}	0.6	0.7
	TrPL Masc	74.3±10.4 ^{Aa}	-	$74.0\pm10.3^{\mathrm{Aa}}$		
Total Body Water (L)	TrEX Fem	32.6±2.1 ^{Aa}	32.4±2.1 ^{Aa}	-	0.7	0.1
	TrPL Fem	32.5±2.4 ^{Aa}	32.4±2.3 ^{Aa}	-		
	TrEX Masc	45.4±5.8 ^{Aa}	45.2±5.8 ^{Aa}	-	0.7	1.9
	TrPL Masc	45.6±5.7 ^{Aa}	45.2±5.9 ^{Aa}	-		
Intracellular Water (L)	TrEX Fem	20.4±1.4 ^{Aa}	20.3±1.3 ^{Aa}	-	0.9	0.1
	TrPL Fem	20.3±1.5 ^{Aa}	20.2±1.4 ^{Aa}	-		
	TrEX Masc	28.5±3.7 ^{Aa}	28.3±3.6 ^{Aa}	-	0.7	0.2
	TrPL Masc	28.6±3.5 ^{Aa}	28.4±3.6 ^{Aa}	-		
Extracellular Water (L)	TrEX Fem	12.3±0.8 ^{Aa}	12.1±0.8 ^{Aa}	-	0.3	1.0
	TrPL Fem	12.2±0.9 ^{Aa}	12.2±0.9 ^{Aa}	-		

	TrEX Masc	16.9±2.1 ^{Aa}	16.8±2.2 ^{Aa}	-	0.7	0.2
	TrPL Masc	17.0±2.2 ^{Aa}	16.9±2.3 ^{Aa}	-		
USG	TrEX Fem	-	1014.2±7.6 ^A	1010.4±5.6 ^A		
	TrPL Fem	-	1013.6±7.7 ^A	1007.6±1.7 ^A		
	TrEX Masc	-	1013.6±7.3 ^A	1009.4 ± 6.5^{A}	0.1	
	TrPL Masc	-	1010.8 ±5.7 ^{Aa}	1006.2±1.8 ^A		
Urine Color	TrEX Fem	-	$2.0{\pm}1.0^{Aa}$	1.9±0.6 ^{Aa}	0.1	
	TrPL Fem	-	2.1±0.9 ^{Aa}	1.7±0.5 ^{Aa}		
	TrEX Masc	-	$2.4{\pm}0.8^{\mathrm{Aa}}$	1.8±0.8 ^{Aa}	0.1	
	TrPL Masc	-	2.0±1.1 ^{Aa}	2.2±2.1 ^{Aa}		

Nota: TrEX = Tereré; TrPL = Decaffeinated Tereré; USG = Urine Specific Gravity; Post-beverage = 45 min after beverage ingestion (TrEX or TrPL); Post-exercise = Immediately after 20 to 27 min of physical effort. Lowercase letters indicate equal means between time points (p > 0.05); Uppercase letters indicate equal means between groups (p > 0.05).

Fonte: os autores.

Table 4 presents the results of Thirst Sensation and Thirst Scale. No significant differences were found between the TrEX and TrPL conditions in the responses to all questions of the thirst sensation questionnaire (p>0.05) and in the values reported by the athletes on the thirst scale (p>0.05).

Page 10 of 14 Diniz et al.

Table 4. Influence of Tereré (TrEX) or placebo Tereré (TrPL) consumption on perception and thirst scale in runners.

Instrument	Questions	Conditions	Post exercise	p
Thirst sensation questionnaire		TrEX	5.9 ± 2.8	0.8
4	1- How thirsty are you right now?	TrPL	5.6 ± 2.3	
	2- How pleasant would it be to	TrEX	7.4 ± 2.3	0.5
	drink some water right now?	TrPL	6.9 ± 2.5	
	-	TrEX	5.7 ± 2.6	0.3
	3-How dry is your mouth right now?	TrPL	5 ± 2.3	
	4-How would you describe the	TrEX	4.3 ± 2.6	0.5
	taste in your mouth?	TrPL	3.7 ± 3.1	
	F. Harrifull days	TrEX	2.3 ± 1.7	0.6
	5 -How full does your stomach feel right now?	TrPL	2.3 ± 2.3	
Thirst scale	1 to 9	TrEX	5.1 ± 2.2	0.8
		TrPL	4.9 ± 1.9	

Source: authors.

Discussion

The findings of the current study indicate that the ingestion of Tereré, when compared to decaffeinated Tereré, did not result in statistical differences in the hydration parameters of well-trained female and male street runners. The body water content, whether total, or intra- or extracellular, remained constant 45 minutes after the ingestion of both beverages. Body mass and urinary measures (USG and color) were also not altered by the ingestion of TrEX and TrPL after physical effort. Only USG decreased due to physical effort in both conditions and sexes.

Our study is the first to analyze the effects of EM consumption in the traditional manner (i.e., with cold water, straw, and gourd) on hydration indicators in runners. In a similar study²⁹, it was observed that compared to water intake, Tereré intake (50g of yerba mate infused in 6 mL/kg of water) did not result in changes in body mass and water content (total, intra- and extracellular) in judo athletes, either 60 minutes after beverage ingestion or after a 90-minute judo training session. It is noted in our study that female and male runners in both experimental conditions showed variable behavior in total body water content after beverage ingestion. Some

athletes showed losses of 100 to 600 mL, while others demonstrated gains of 100 to 400 mL, and 3 athletes showed no change in body water content.

It is worth highlighting that water losses were expected, since participants emptied their bladders before performing the BIA test, and average urinary losses are approximately 245 mL per urination. The amount of water ingested by our athletes in both conditions ranged from 271 to 403 mL in women and 318 to 539 mL in men. We believe that this amount was sufficient to keep the athletes' average body mass relatively constant in both the TrEX and TrPL conditions, even after 20 to 27 minutes of physical effort. It is noted that the sweat rate of our athletes ranged from 0.01 to 0.03 L/min, which corresponds to sweat losses of 230 to 900 mL in women and 320 to 1300 mL in men, considering the time of effort performed.

The analysis of urinary parameters reinforces our assumption. The presence of light yellow urine and the low USG of the athletes, both after beverage ingestion and after physical effort, indicate that the athletes maintained a good hydration status throughout the experimental session, despite temporal variations in USG. Interestingly, there was a reduction in USG values after physical effort in both sexes and beverages, contradicting what would be expected since the athletes performed intense physical effort. We suppose that besides the adequate volume of water consumed (sufficient to ensure hydration), the duration of effort performed by the athletes (20 to 27 minutes) may not have been enough to promote changes in urine solute concentration. In addition, the favorable temperature conditions (approx. 22°C) and humidity (between 53 and 59%) in the laboratory supported the good hydration status of the athletes.

The joint analysis of our results leads us to presume that the amount of water (matched between beverages), and not the substances present in yerba mate (especially caffeine), was the main determining factor for the hydration of our athletes. Although caffeine has diuretic properties, which generally increase the body's water excretion, its impact on hydration may not be so direct. The study by Killer et al.⁶ revealed that moderate coffee intake, about 3–4 cups per day, does not cause significant water losses, especially in individuals who are accustomed to caffeine, indicating that, in moderate amounts, caffeine does not exert a diuretic effect sufficient to cause dehydration. In our study, caffeine intake by athletes ranged from 0.59 mg/kg in men to 0.77 mg/kg in women. Considering the caffeine concentration in our yerba mate (44 mg), we believe that the dosage was far below that necessary to produce changes in the hydration status of the athletes.

Previous studies by Armstrong et al.³⁰ and Maughan & Griffin³¹ indicate that moderate doses of caffeine do not significantly affect water balance in individuals accustomed to its consumption. In our study, the water used to prepare the Tereré directly contributed to fluid intake, which may increase total body water. Therefore, even with the presence of caffeine, regular consumption of Tereré may aid hydration.

The results obtained regarding the sensation of thirst are as expected. After physical effort, many athletes reported feeling significant thirst, as shown by the responses to the questionnaire. However, there were no statistical differences between the groups that consumed Tereré and those that consumed the placebo. The increase in thirst sensation in both groups may be seen as a natural body response, trying to replenish lost fluids and weight during training. Studies indicate that we usually feel thirsty even with discrete body mass losses (less than 1%)³². In the current study, not all athletes lost body mass after performing the treadmill run. Among those who did, body mass loss in women in the placebo condition corresponded to 0.27%, and in the Tereré condition to 0.51% of baseline body mass. Among men, the losses were 0.51% in the placebo condition and 0.42% in the Tereré condition.

The current study presents some limitations. The absence of studies involving similar protocols and populations reduces the possibility of greater comparisons with our findings. In the majority of previous studies on this theme, yerba mate was administered in capsule or tea form³³. Additionally, beverages were mainly tested in clinical populations (e.g., people with

Page 12 of 14 Diniz et al.

diabetes, obesity)³⁴, and the outcomes were different from those analyzed in the current study, with emphasis on biochemical and metabolic markers, and physical performance measures. Only one study analyzed hydration parameters. Among the strengths of the study, we highlight the inclusion of women and the experimental design adopted (randomized double-blind clinical trial), considered the gold standard design, as it allows us to reduce the influence of some confounding variables. Furthermore, in our study, yerba mate was administered in the culturally common form (with water, a straw, and a gourd), increasing the external validity of our results.

Conclusions

Our findings indicate that the effects of Tereré were similar to those of decaffeinated Tereré on the hydration parameters of the athletes. Therefore, both beverages were effective in ensuring the hydration of runners, representing a viable option for pre-training or precompetition use in races lasting less than 30 minutes.

References

- Sawka MN, Burke LM, Eichner ER, Maughan RJ, Montain SJ, Stachenfeld NS. Exercise and fluid replacement. Med Sci Sports Exerc. 2007;39(2):377-90.DOI: https://doi.org/10.1249/mss.0b013e31802ca597.
- 2. Kerksick CM, Wilborn CD, Roberts MD, Smith-Ryan A, Kleiner SM, Jäger R, et al. ISSN exercise & sports nutrition review update: research & recommendations. J Int Soc Sports Nutr. 2018;15:38.DOI: https://doi.org/10.1186/s12970-018-0242-y.
- 3. Sawka MN, Noakes TD. Does dehydration impair exercise performance? Med Sci Sports Exerc. 2007;39(8):1209-17. DOI: https://doi.org/10.1249/mss.0b013e318038ad4e
- 4. Cheuvront SN, Kenefick RW. Dehydration: Physiology, assessment, and performance effects. Compr Physiol. 2014;4(1):257-85. DOI: https://doi.org/10.1002/cphy.c130017.
- Zhang Y, Coca A, Casa DJ, Antonio J, Green JM, Bishop PA. Caffeine and diuresis during rest and exercise: a meta-analysis. J Sci Med Sport. 2015;18(5):569-74. DOI: https://doi.org/10.1016/j.jsams.2014.07.017.
- 6. Killer SC, Blannin AK, Jeukendrup AE. No evidence of dehydration with moderate daily coffee intake: a counterbalanced cross-over study in a free-living population. PLoS One. 2014;9(1):e84154. DOI: https://doi.org/10.1371/journal.pone.0084154.
- 7. Alikaridis F. Natural constituents of Ilex species. J Ethnopharmacol. 1987;20(2):121-44. DOI: https://doi.org/10.1016/0378-8741(87)90144-1.
- 8. Bastos DHM, de Oliveira DM, Matsumoto RT, Carvalho PDO, Ribeiro ML. Yerba maté: Pharmacological Properties, Research and Biotechnology [Internet]. Medicinal and Aromatic Plant Science and Biotechnology. 2007[cited 2025 Apr 30]. Available from: www.isiknowledge.com
- Chaicouski A, Silva JE, Trindade JLF, Canteri MHG. Determinação da quantidade de compostos fenólicos totais presentes em extratos líquido e seco de erva-mate (Ilex paraguariensis). Rev Bras Prod Agroind. 2014[cited 2025 Apr 30];16(1):33-41. Available from: https://www.researchgate.net/publication/277926322_DETERMINACAO_DA_QUANTIDADE_DE_COMPOSTOS_FENOLICOS_TOTAIS_PRESENTES_EM_EXTRATOS_LIQUIDO_E_SECO_DE_E_RVA-MATE_Ilex_paraguariensis.
- 10. Kim SY, Oh MR, Kim MG, Chae HJ, Chae SW. Anti-obesity effects of Yerba Mate (Ilex paraguariensis): a randomized, double-blind, placebo-controlled clinical trial. BMC Complement Altern Med. 2015;15:338. DOI: https://doi.org/10.1186/s12906-015-0873-1.
- 11. de Morais EC, Stefanuto A, Klein GA, Boaventura BCB, de Andrade F, Wazlawik E, et al. Consumption of Yerba Mate (Ilex paraguariensis) improves serum lipid parameters in healthy dyslipidemic subjects and provides an additional LDL-cholesterol reduction in individuals on statin therapy. J Agric Food Chem. 2009;57(18):8316-24. DOI: https://doi.org/10.1021/jf9013072.
- 12. Alkhatib A, Seijo M, Larumbe E, Naclerio F. Acute effectiveness of a "fat-loss" product on substrate utilization, perception of hunger, mood state and rate of perceived exertion at rest and during exercise. J Int Soc Sports Nutr. 2015;12:40. DOI: https://doi.org/10.1186/s12970-015-0101-0
- 13. Alkhatib A, Atcheson R. Yerba Maté (Ilex paraguariensis) metabolic, satiety, and mood state effects at rest and during prolonged exercise. Nutrients. 2017;9(8):882. DOI: https://doi.org/10.3390/nu9080882.

- Areta JL, Austarheim I, Wangensteen H, Capelli C. Metabolic and performance effects of Yerba Mate on well-trained cyclists. Med Sci Sports Exerc. 2018;50(4):817-26. DOI: https://doi.org/10.1249/MSS.0000000000001492.
- 15. Krolikowski TC, Borszcz FK, Panza VP, Bevilacqua LM, Nichele S, da Silva EL, et al. The impact of pre-exercise carbohydrate meal on the effects of Yerba Mate drink on metabolism, performance, and antioxidant status in trained male cyclists. Sports Med Open. 2022;8:93. DOI: https://doi.org/10.1186/s40798-022-00461-y.
- 16. Baisch ALM, Johnston KB, Stein FLP. Endothelium-dependent vasorelaxing activity of aqueous extracts of Ilex paraguariensis on mesenteric arterial bed of rats. J Ethnopharmacol. 1998;60(2):133-9. DOI: https://doi.org/10.1016/S0378-8741(97)00148-0.
- 17. Stein FLP, Schmidt B, Furlong EB, Soares LAS, Soares MCF, Vaz MRC, et al. Vascular responses to extractable fractions of Ilex paraguariensis in rats fed standard and high-cholesterol diets. Biol Res Nurs. 2005;7(2):146-56. DOI: https://doi.org/10.1177/1099800405280922.
- 18. Monte MM. Elaboração, validação e reprodutibilidade de um questionário para avaliação do consumo de cafeína em adultos [dissertação]. Maceió: Universidade Federal de Alagoas; 2021[cited 2025 Apr 30]. Available from:

 <a href="https://www.repositorio.ufal.br/bitstream/123456789/11775/1/Elabora%C3%A7%C3%A3o%2C%20valida%C3%A7%C3%A3o%20e%20reprodutibilidade%20de%20um%20question%C3%A1rio%20para %20avalia%C3%A7%C3%A3o%20do%20consumo%20de%20cafe%C3%ADna%20em%20adultos.pdf
- Minton DM, Eberman LE. Best practice for clinical hydration measurement. Athl Ther Today. 2009[cited2025 Apr 30;14(1):9-11. Available from: https://www.researchgate.net/publication/270569844 Best Practice for Clinical Hydration Measurm ent.
- 20. Inbody10. Manual de Operação. Ottoboni; 2020[cited 2025 Apr 30]. Available from: https://www.ottoboni.com.br/produtos/inbodys10/.
- 21. Armstrong LE, Ganio MS, Klau JF, Johnson EC, Casa DJ, Maresh CM. Novel hydration assessment techniques employing thirst and a water intake challenge in healthy men. Appl Physiol Nutr Metab. 2014;39(2):138-44. DOI: https://doi.org/10.1139/apnm-2013-0360.
- 22. Engell DB, Maller O, Sawka MN, Francesconi RN, Drolet L, Young AJ. Thirst and fluid intake following graded hypohydration levels in humans. Physiol Behav. 1987;40(2):229-36. DOI: https://doi.org/10.1016/0031-9384(87)90179-1.
- 23. Rolls BJ, Wood RJ, Rolls ET, Lind H, Lind W, Ledingham JG. Thirst following water deprivation in humans. Am J Physiol Regul Integr Comp Physiol. 1980;239(5):R476-82. DOI: https://doi.org/10.1152/ajpregu.1980.239.5.R476.
- 24. Armstrong LE. Hydration assessment techniques. Nutr Rev. 2005;63(6 Pt 2):S40-54. DOI: https://doi.org/10.1111/j.1753-4887.2005.tb00153.x.
- 25. Kostelnik SB, Davy KP, Hedrick VE, Thomas DT, Davy BM. The validity of urine color as a hydration biomarker within the general adult population and athletes: a systematic review. J Am Coll Nutr. 2021;40(2):172-9. DOI: https://doi.org/10.1080/07315724.2020.1734987.
- 26. Casa DJ, Armstrong LE, Hillman SK, Montain SJ, Reiff RV, Rich BS, et al. National Athletic Trainers' Association position statement: fluid replacement for athletes. J Athl Train. 2000[cited 2025 Apr 30].;35(2):212-24. Available from: https://www.nata.org/sites/default/files/FluidReplacementsForAthletes.pdf
- 27. Hamouti N, Del Coso J, Estevez E, Mora-Rodriguez R. Dehydration and sodium deficit during indoor practice in elite European male team players. Eur J Sport Sci. 2010;10(5):329-36. DOI: https://doi.org/10.1080/17461391003699157.
- 28. Conselho Nacional de Ergometria. Consenso Nacional de Ergometria. Arq Bras Cardiol. 1995[cited 2025 Apr 30];65(2):165-71. Available from: https://derc.org.br/wp-content/uploads/2021/05/20.pdf .
- 29. Denarde FEF. Ciências do movimento humano [tese]. Campo Grande: Universidade Federal de Mato Grosso do Sul; 2023[cited 2025 Apr 30]. Available from: https://repositorio.ufms.br/handle/123456789/6637.
- 30. Armstrong LE, Casa DJ, Maresh CM, Ganio MS. Caffeine, fluid-electrolyte balance, temperature regulation, and exercise-heat tolerance. Exerc Sport Sci Rev. 2007;35(3):135-40. DOI: https://doi.org/10.1097/jes.0b013e3180a02fa.
- 31. Maughan RJ, Griffin J. Caffeine ingestion and fluid balance: a review. J Hum Nutr Diet. 2003;16(6):411-20. DOI: https://doi.org/10.1046/j.1365-277x.2003.00477.x.
- 32. Armstrong LE, Maresh CM. The induction and decay of caffeine tolerance in athletes. J Appl Physiol. 1999;87(2):1220-7. DOI: https://doi.org/10.1152/jappl.1999.87.2.1220.

Page 14 of 14 Diniz et al.

33. Alkhatib A. Yerba Maté (Ilex paraguariensis) ingestion augments fat oxidation and energy expenditure during exercise at various submaximal intensities. Nutr Metab (Lond). 2014;11:42. DOI: https://doi.org/10.1186/1743-7075-11-42.

34. Gawron-Gzella A, Chanaj-Kaczmarek J, Cielecka-Piontek J. Yerba Mate—A long but current history. Nutrients. 2021;13(11):3706. DOI: https://doi.org/10.3390/nu13113706.

Acknowledgements: Federal University of Mato Grosso do Sul (UFMS), Coordination for the Improvement of Higher Education Personnel (CAPES), National Council for Scientific and Technological Development (CNPq), Erva-mate Barão de Cotegipe.

CRediT author statement

Carolina Rocha Diniz1:Conceptualization, Data curation, Methodology, Project administration, Writing – original draft, Writing – review & editing; Raphael de Jesus Brittes 2:Methodology, Project administration, Writing review; Fabiane La Flor Ziegler Sanches 3: Validation, Writing review; Cássio Pinho dos Reis 4: Data curation, Software, Formal analysis; Christianne de Faria Coelho-Ravagnani 5:Funding acquisition, conceptualization, Project administration, Supervision, Writing – draft, review & editing.

ORCID dos autores:

Carolina Rocha Diniz: https://orcid.org/0009-0003-2965-5270
Raphael de Jesus Brittes: https://orcid.org/0000-0002-9162-1798
Fabiane La Flor Ziegler Sanches: https://orcid.org/0000-0002-0096-3697

Cássio Pinho dos Reis: https://orcid.org/0000-0002-2211-2295

Christianne de Faria Coelho-Ravagnani: https://orcid.org/0000-0002-9082-6521

Editor: Carlos Herold Junior. Received om Aug 14, 2024. Reviewed on Nov 28, 2024. Accepted on Jan 07, 2025.

Corresponding author: Carolina Rocha Diniz. E-mail: rochacarolinad@gmail.com