



# DOES GREEN TEA ENHANCE THE EFFECTS OF PHYSICAL TRAINING ON HEART RATE VARIABILITY?

O CHÁ VERDE POTENCIALIZA EFEITOS DO TREINAMENTO FÍSICO NA VARIABILIDADE DA FREQUÊNCIA CARDÍACA?


¿AUMENTA EL TÉ VERDE LOS EFECTOS DEL ENTRENAMIENTO FÍSICO SOBRE LA VARIABILIDAD DEL RITMO CARDÍACO?

Wellington Lourenço Mendes dos Santos<sup>1</sup> 


(Physical Education Professional)

José Martins Juliano Eustáquio<sup>1,2</sup> 

(Sports Physician)

Isabella Cecilio Resende Ferreira<sup>2</sup> 


(Medical Student)

Júlia Oliveira Perez<sup>2</sup> 

(Medical Student)

Rafael Pacheco Policarpo<sup>2</sup> 

(Medical Student)

Rosângela Soares Chriquer<sup>3</sup> 

(Biological Sciences)

Octávio Barbosa Neto<sup>1</sup> 

(Physical Education Professional)

1. Universidade Federal do Triângulo Mineiro, Graduate Program in Physical Education, Sports and Exercise Cardiology Research Group, Uberaba, MG, Brazil.

2. Universidade de Uberaba, Hospital Mário Palmério, Uberaba, MG, Brazil.

3. Universidade Federal de São Paulo, Department of Biosciences, São Paulo, SP, Brazil.

## Correspondence:

Octávio Barbosa Neto  
Universidade Federal do Triângulo Mineiro, Programa de Pós-Graduação em Educação Física, Grupo de Pesquisa em Cardiologia do Esporte e do Exercício.  
Av. Tutunas, 490, Uberaba, Brasil.  
38061-500.  
zemartinsjuliano@hotmail.com/  
octavio.neto@uftm.edu.br

## ABSTRACT

**Introduction:** Green tea is associated with a series of health benefits, as is physical training. However, in combination they present little known chronotropic and autonomic cardiac effects. **Objective:** To evaluate the effect of the association of chronic administration of green tea and physical training on basal heart rate (HR) and heart rate variability (HRV) in Wistar rats. **Methods:** Forty-three Wistar rats (*Rattus norvegicus*, var. *albinus*), paired by weight and age, were distributed among four experimental groups, titled sedentary control (CON<sub>sed</sub>, n = 10), trained control (CON<sub>tr</sub>, n = 08), sedentary tea (CHÁ<sub>sed</sub>, n = 16) and trained tea (CHÁ<sub>tr</sub>, n = 09). Ingestion of the tea was *ad libitum*. The physical training protocol lasted for eight weeks and consisted of sessions of swimming with incremental loads. At the end of the training, basal heart rate and heart rate variability (HRV) in both time and frequency domains were determined. The level of significance adopted was 5% ( $p < 0.05$ ). **Results:** Both physical training and consumption of green tea caused higher resting bradycardia than that of the CON<sub>sed</sub> group animals. Regarding HRV, the CON<sub>tr</sub>, CHÁ<sub>sed</sub>, and CHÁ<sub>tr</sub> groups presented significantly higher values than the CON<sub>sed</sub> group. Supplementation with green tea caused an increase in the variance, high frequency (HF) component, and sympathovagal balance as compared to the CON<sub>sed</sub> group. Physical training (PT) did not enhance any of the parameters evaluated. **Conclusions:** There was no significant optimization of the hemodynamic or autonomic cardiovascular parameters resulting from the association between the administration of green tea and physical training in Wistar rats. **Level of Evidence IV; Case Series.**

**Keywords:** Tea; Exercise; Sports medicine; Cardiovascular system; Heart rate.

## RESUMO

**Introdução:** O chá verde está associado a uma série de benefícios à saúde, assim como o treinamento físico. Porém, a associação de ambos apresenta efeitos cronotrópicos e autonômicos cardíacos pouco conhecidos. **Objetivo:** Avaliar o efeito da associação da administração crônica de chá verde e do treinamento físico sobre a frequência cardíaca (FC) basal e a variabilidade da frequência cardíaca (VFC) em ratos Wistar. **Métodos:** Quarenta e três ratos Wistar (*Rattus norvegicus*, var. *albinus*), pareados pelo peso e por idade, foram distribuídos em quatro grupos experimentais, intitulados de controle sedentário (CON<sub>sed</sub>, n = 10), controle treinado (CON<sub>tr</sub>, n = 08), chá sedentário (CHÁ<sub>sed</sub>, n = 16) e chá treinado (CHÁ<sub>tr</sub>, n = 09). A ingestão de chá foi "ad libitum". O protocolo de treinamento físico teve duração de oito semanas e foi realizado através de sessões de natação com incrementos de carga. Ao final, foram determinadas a frequência cardíaca basal e a variabilidade da frequência cardíaca (VFC) nos domínios do tempo e da frequência. O nível de significância adotado foi de 5% ( $p < 0,05$ ). **Resultados:** Tanto o treinamento físico quanto o consumo de chá verde promoveram bradicardia de repouso maior, quando comparados com os animais do grupo CON<sub>sed</sub>. Com relação à VFC, os grupos CON<sub>tr</sub>, CHÁ<sub>sed</sub> e CHÁ<sub>tr</sub> apresentaram valores significativamente maiores que o grupo CON<sub>sed</sub>. A suplementação do chá verde ocasionou aumento da variância, do componente de alta frequência (HF) e do balanço simpato-vagal com relação ao grupo CON<sub>sed</sub>. O treinamento físico (TF) não potencializou nenhum dos parâmetros avaliados. **Conclusões:** Não houve otimização significativa nos parâmetros hemodinâmicos e autonômicos cardiovasculares em decorrência da associação entre administração de chá verde e treinamento físico em ratos Wistar. **Nível de Evidência IV; Série de casos.**

**Descritores:** Chá; Treinamento físico; Medicina esportiva; Sistema cardiovascular; Frequência cardíaca.

## RESUMEN

**Introducción:** El té verde está asociado a una serie de beneficios para la salud, así como el entrenamiento físico. Sin embargo, la asociación de ambos presenta efectos cronotrópicos y autonómicos cardíacos poco conocidos. **Objetivo:** Evaluar el efecto de la asociación de la administración crónica de té verde y el entrenamiento físico sobre la frecuencia cardíaca (FC) basal y la variabilidad de la frecuencia cardíaca (VFC) en ratos Wistar. **Métodos:** Cuarenta y tres ratos Wistar (*Rattus norvegicus*, var. *albinus*), clasificadas por peso y edad, fueron distribuidas en cuatro grupos



experimentales denominados control sedentario (CONsed, n = 10), control entrenado (CONtre, n = 08), té sedentario (CHÁsed, n = 16) y té entrenado (CHÁtre, n = 09). La ingesta de té fue "ad libitum". El protocolo de entrenamiento físico duró ocho semanas y se realizó mediante sesiones de natación con incrementos de carga. Al final, se determinó la frecuencia cardíaca basal y la variabilidad de la frecuencia cardíaca (VFC) en los dominios del tiempo y la frecuencia. El nivel de significancia adoptado fue del 5% ( $p < 0,05$ ). Resultados: Tanto el entrenamiento físico como el consumo de té verde promovieron una mayor bradicardia en reposo en comparación con los animales del grupo CONsed. En cuanto a la VFC, los grupos CONtre, CHÁsed y CHÁtre presentaron valores significativamente más altos que el grupo CONsed. La suplementación con té verde provocó un aumento de la varianza, del componente de alta frecuencia (HF) y del balance simpático-vagal en relación con el grupo CONsed. El entrenamiento físico (TF) no potenció ninguno de los parámetros evaluados. Conclusiones: No hubo una optimización significativa de los parámetros hemodinámicos y autonómicos cardiovasculares derivada de la asociación entre la administración de té verde y el entrenamiento de ejercicio en ratas Wistar. **Nivel de Evidencia IV; Serie de casos.**

**Descriptor:** Té; Ejercicio físico; Medicina deportiva; Sistema cardiovascular; Frecuencia cardíaca.

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## INTRODUCTION

Better known for its thermogenic effects, green tea is associated with a series of health benefits<sup>1,2</sup> attributed mainly to the presence of catechins, polyphenols among which epigallocatechin-3-gallate (EGCG) is the one with the greatest biological power.

In the cardiovascular system, these substances have protective effects,<sup>2</sup> possibly due to their anti-inflammatory power, with a fundamental reduction of oxidative stress, in addition to important repercussions on the autonomous nervous system, mainly by prolonging and intensifying parasympathetic stimulation.<sup>3</sup>

Similar benefits are also provided by physical training. It is known that moderate-intensity aerobic physical training reduces sympathetic nervous activity in several populations,<sup>4,5</sup> with significant impact on cardiovascular physiology and a consequent decrease in mortality rates.<sup>6</sup>

When combined, it is known, for example, that the thermogenic effect of green tea can be enhanced,<sup>7</sup> since the energy expenditure becomes greater during physical training, which favors the metabolism and, thus, weight loss.<sup>8</sup> However, as regards the cardiovascular system, to date there are limited studies that report the long-term effects of the combination of green tea and physical exercise.

Thus, it is proposed that the application of green tea combined with physical training for an eight-week period will influence chronotropic and autonomic cardiac patterns. The objective of the present study was to evaluate the effect of the association of the administration of green tea and physical training on basal heart rate (HR) and heart rate variability (HRV) in Wistar rats.

## MATERIALS AND METHODS

Forty-three Wistar rats (*Rattus norvegicus*, var. *albinus*), paired by weight (356.23 grams) and age ( $\pm 12$  weeks), were kept in 414 x 344 x 168 mm autoclavable polypropylene collective boxes (ZOOTECH 375), with galvanized steel grated lids and stainless steel dividers, lined with wood shavings, at a room temperature of 22- 23°C, at 40-70% humidity, and a photoperiod of 12 hours light/dark, with free access to water or tea and feed (Nuvilab CR1, Nuvital Nutrientes Ltda, Curitiba, Paraná, Brazil).

The animals were distributed, according to the treatment to be received, into four experimental groups named sedentary control (CON<sub>sedr</sub>, n=10), trained control (CON<sub>tre</sub>, n=08), sedentary tea (CHÁ<sub>sedr</sub>, n=16), and trained tea (CHÁ<sub>tre</sub>, n=09).

This study was approved by the Ethics Committee on Animal Use (CEUA) of the Universidade Federal do Triângulo Mineiro (UFTM) (protocol 338) and all experimental procedures used are in accordance with the standards for the use and care of laboratory animals in research.<sup>9,10</sup>

## Physical Training

The animals of the trained groups were submitted to physical training (PT) in swimming sessions five times a week for eight weeks. The PT protocol adopted was adapted from a previous study<sup>11</sup> with a similar methodology, and characterized as moderate intensity, sufficient to optimize muscle oxidative capacity (Figure 1). The sedentary groups were placed in the tank for ten minutes, five times a week, without weight on their tails, in order to rule out possible physiological changes that could occur due to water stress.

The PT, started daily at 1 pm, was conducted in a glass tank measuring 100 x 50 x 60 cm, filled with water heated to 30 °C  $\pm$  2 °C at a depth of 50 cm, sufficient to prevent the animals from touching their tails on the bottom of the tank and resting.

After the PT sessions, the animals were dried with paper toweling, placed in a dry environment without shavings for five minutes, and then in an environment with shavings under incandescent light (60 watts) for fifteen minutes to warm up before being returned to the boxes.

## Preparation and administration of the green tea

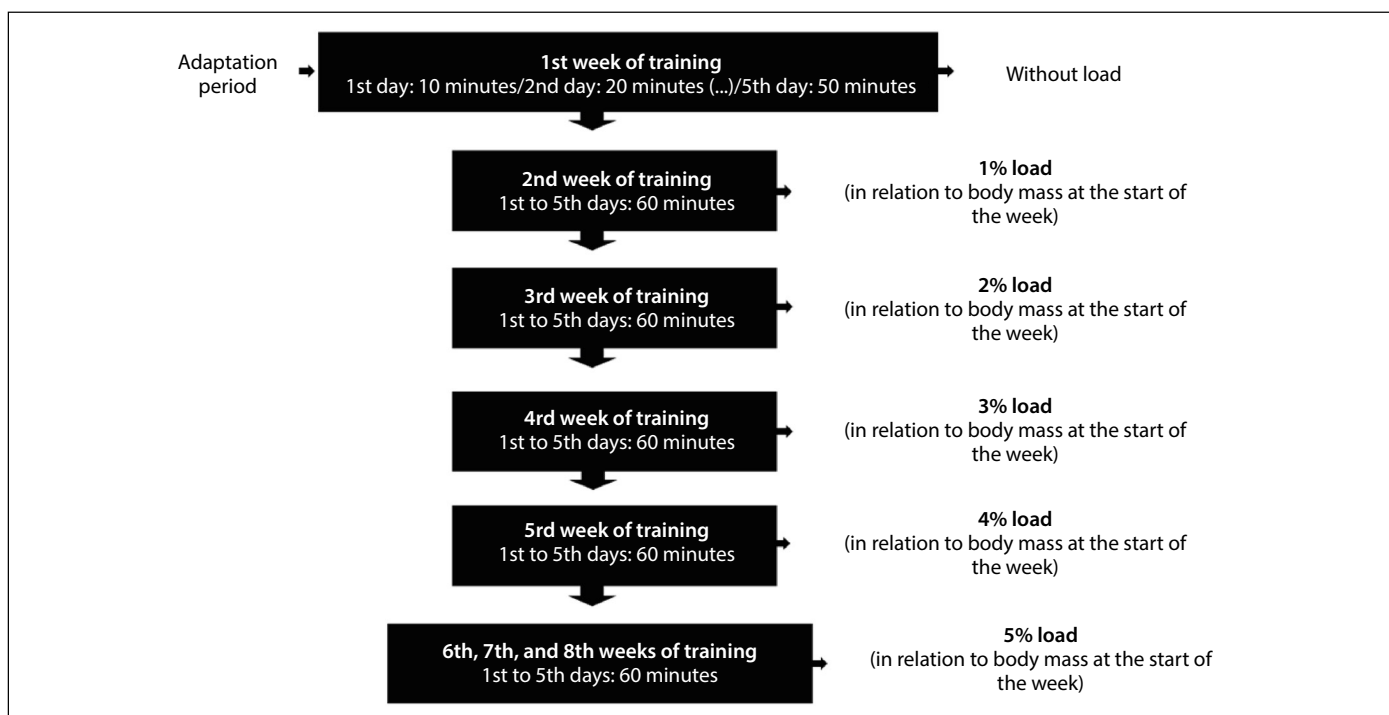
To prepare the green tea, a total of two grams of leaves of the herb *Camellia Sinensis*, were added to 1000 milliliters of boiling water (infusion method). After 20 minutes, the mixture was filtered, and a determined volume put into opaque drinking dispensers.

The tea was prepared daily at the same time as the physical training period. The animals of the tea ingestion groups were offered tea *ad libitum* from the 12th week of life onwards. The tea was always prepared at 08:00 am. An aliquot of this tea was separated to confirm the lyophilization of the green tea mass to estimate the amount of green tea ingested by each animal.

## Chronotropic analysis

Twenty-four hours after the last swimming protocol session, the animals of both trained groups underwent a surgical procedure for cannulation of the right femoral artery. Then, the catheters were externalized to the dorsal region to position them properly for direct recording of the hemodynamic parameters. The animals remained lodged in individual boxes for postoperative recovery with water or tea and feed *ad libitum*.

On the day after the surgery, the arterial catheter was connected to a blood pressure (BP) transducer and a signal amplifier (Model 8805A®, Hewlett-Packard, USA), where the signal was converted by an analog-digital board with a sampling frequency of 1000 Hz. Then, the signal was transferred to a computerized data acquisition system (Aqdados®, Lynx Tec. Eletron. SA, São Paulo, Brazil) and subsequently stored in the computer.



**Figure 1.** Swimming protocol adopted by the trained groups.

At the time of recording, all the animals were kept in a noise- and temperature-controlled (27 °C) room. After positioning the animals and connecting them to the equipment, they underwent an adaptation period of 15 minutes before the recordings were performed. After the period of animal adaptation and confirmation of the adequacy of signal capture, the continuous 30-minute recording of the pulsatile BP was started in order to obtain the heart rate (HR) values. These basal values were subsequently used to analyze the heart rate variability (HRV) in the time domain (TD), through the variance in the pulse interval (PI), and the frequency domain (FD), through autoregressive spectral analysis.<sup>12</sup>

### Heart rate variability

The PI time series, collected during the 30 basal minutes, were divided into serial segments of 300 beats, for which spectral decomposition was performed. The procedure allowed automatic quantification of the central frequency and power of each relevant spectral component in absolute units, as well as in normalized units. The normalization of values minimizes the interference of total power on the components.

The normalization procedure was performed by dividing the power of the low-frequency (LF; 0.15-0.4 Hz) or high-frequency (HF; 0.04 –0.15 Hz) component by the total spectral power, from which the power of the very-low-frequency band (VLF, 0.01-0.20 Hz) was subtracted, and multiplying the result by 100.<sup>12</sup>

The spectral parameters obtained for each individual stationary 300-beat segment were measured and the resulting mean values for the 30 basal minutes were collected for each animal. The quotient of LF by HF (ratio of LF to HF; LF/HF) was used to express sympathovagal balance. The normalized power of the components LF and HF was calculated in normalized units (nu), using the equations mentioned above.<sup>12</sup>

### Statistical analysis

Data normality was tested by means of the Kolmogorov-Smirnov test. For normal data, the two-way ANOVA test was performed, followed by the Tukey's post hoc test or the Mann-Whitney test, according to normality. The significance level adopted was 5% ( $p < 0.05$ ). The analyses were conducted using SigmaStat 8.0 software (Jandel Scientific Software, SPSS, Chicago, IL).

## RESULTS

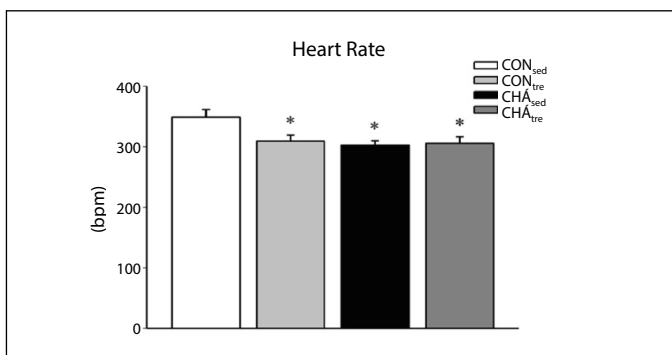
Mean tea consumption by the animals was 207.1 mg during the day without a significant difference between the groups.

Both physical training ( $CON_{tre}$  309.2 ± 9.9 bpm) and green tea consumption ( $CH\dot{A}_{sed}$  302.5 ± 7.13 bpm and  $CH\dot{A}_{tre}$  305.6 ± 10.8 bpm) promoted higher resting bradycardia, when compared to the animals in the  $CON_{sed}$  (348 ± 12.56;  $p < 0.05$ ) group. On the other hand, supplementation with green tea combined with PT did not enhance bradycardia among the  $CON_{tre}$ ,  $CH\dot{A}_{sed}$ , and  $CH\dot{A}_{tre}$  groups (Figure 2).

In relation to HRV, the  $CON_{tre}$ ,  $CH\dot{A}_{sed}$ , and  $CH\dot{A}_{tre}$  groups had significantly higher values than the  $CON_{sed}$  group. Supplementation with green tea caused increases in variance, the HF component, and the sympathovagal balance in the  $CON_{sed}$  group, and PT did not enhance this increase. The  $CON_{tre}$ ,  $CH\dot{A}_{sed}$ , and  $CH\dot{A}_{tre}$  groups presented a significantly higher LF than the  $CON_{sed}$  group. The  $CH\dot{A}_{tre}$  group also had an increase in LF in relation to the  $CH\dot{A}_{sed}$  group. No significant differences between the other HRV data were observed (Table 1).

## DISCUSSION

The main findings of this study were that both supplementation with green tea and PT promoted better heart rhythm, but the combination of these factors did not evolve with potentiation of the effects.



**Figure 2.** Basal heart rate of the study groups (presented as mean and standard error of mean).

**Table 1.** HRV component values in the time and frequency domains.

	CONsed	CONtre	CHÁsed	CHÁtre
PI (ms <sup>2</sup> )	174.1 ± 6.6	195.2 ± 6.3*	199.5 ± 4.5*	198.0 ± 6.8*
Variance (ms <sup>2</sup> )	13.2 ± 2.7	20.0 ± 4.1	43.0 ± 7.8*	44.3 ± 27.3*
VLF (ms <sup>2</sup> )	8.3 ± 1.9	9.1 ± 2.7	43.0 ± 7.8	44.3 ± 9.6
LF (ms <sup>2</sup> )	0.6 ± 0.2	1.1 ± 0.2*	0.9 ± 0.1*	1.3 ± 0.3* <sup>ns</sup>
LF (nu)	9.1 ± 2.3	9.8 ± 1.8	10.5 ± 1.2	12.4 ± 1.9
HF (ms <sup>2</sup> )	4.5 ± 0.6	9.7 ± 2.6	14.2 ± 1.3*	15.6 ± 2.6*
HF (nu)	85.7 ± 2.2	89.8 ± 2.1	83.8 ± 1.8	85.0 ± 1.4
LF/HF	0.15 ± 0.04	0.13 ± 0.03	0.06 ± 0.01*	0.08 ± 0.02*

Data expressed as mean ± standard error of mean (sem). PI = pulse interval, LF = low frequency band, HF = high frequency band, nu = normalized units, ms = milliseconds, ms<sup>2</sup> = milliseconds squared, CON<sub>sed</sub> = Sedentary control, CON<sub>tre</sub> = Trained control, CHÁ<sub>sed</sub> = Sedentary tea, CHÁ<sub>tre</sub> = Trained tea. \*p < 0.05 versus CON<sub>sed</sub>, <sup>ns</sup>p < 0.05 versus CHÁ<sub>sed</sub>.

The PT and green tea supplementation used in the study caused significant resting bradycardia. Regarding the effect of training, we observed data similar to those reported by previous studies conducted using similar PT protocols.<sup>11,13,14</sup> It is known that aerobic exercise performed regularly reduces resting HR values both in humans<sup>15</sup> and in animal models,<sup>16</sup> and this is considered an important marker of cardiovascular adaptation to this type of training.<sup>17</sup> This finding shows, therefore, the effectiveness of the protocol used in the current study.

Green tea was also effective in reducing resting HR, an outcome similar to that observed in other studies.<sup>18,19</sup> This resting bradycardia is possibly associated with the mechanisms that justify the findings about the other cardioprotective effects of green tea extract, such as an improved diastolic pattern in patients with myocardial pathology,<sup>20</sup> and control of blood pressure levels in hypertensive patients,<sup>3</sup> among others.<sup>21,22</sup> Currently, the best known of these mechanisms is its potential to act in controlling oxidative stress, both at the vascular and systemic levels.<sup>2,3,23</sup>

It is known that resting bradycardia is more pronounced in people who practice increasingly intensive physical exercise, with the prime example being low HR among professional athletes.<sup>15</sup> However, as for the association between supplementation with increasing doses of green tea and resting bradycardia, the current findings are conflicting.<sup>19,24</sup>

The resting bradycardia results evidenced by PT and by the administration of green tea can also be corroborated by the increase in HR variance, which corresponds to greater estimated vagal activity on the sinus node in the time domain.<sup>12</sup> In addition, an increase in the high

frequency (HF) band was also observed in the animals in the CHÁ<sub>sed</sub> and CHÁ<sub>tre</sub> groups, which is the spectral component modulated by the parasympathetic nervous system. In a way, these results show that the chronic use of green tea caused better adjustment of the cardiac rhythm at different moments and in response to different stimuli, such as PT.

Little potentiation effect was observed, both in relation to resting bradycardia and HRV, from the combination of green tea and PT. The absence of this intensification of effects was also verified in a double-blind, randomized study that evaluated the HRV parameters during energy drink supplementation (caffeine and green tea) and cycling training.<sup>25</sup> Therefore, to date, studies suggest that there is no increase in these hemodynamic variables from the association.

As for other factors, the findings in the literature are conflicting. A recent meta-analysis observed that green tea supplementation associated with physical exercise did not show greater potentiation in improving the lipid profile in humans.<sup>26</sup> However, studies in obese humans reported evidence of enhanced effects on anti-inflammatory and anthropometric parameters from the association between green tea supplementation and aerobic training.<sup>8,27</sup>

The main limitations of our study are the absence of green tea ingestion using the gavage method and of supplementation using increasing doses among the groups. In general, the variety of results in the literature that addresses this topic suggests that, initially, a better understanding of the parameters involved in supplementation, such as concentration, dilution, and time and form of administration, is needed so we can determine the best way to obtain the results sought by this intervention. However, when considered together with the current knowledge, our study suggests that the combination of green tea and PT may not result in autonomic cardiovascular effects.

## CONCLUSION

Our data shows that there was no significant optimization of the hemodynamic and autonomic cardiovascular parameters in Wistar rats as a result of the association of the administration of green tea and physical training.

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All authors declare no potential conflict of interest related to this article

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**AUTHORS' CONTRIBUTIONS:** Each author made significant individual contributions to this manuscript. WLMS: writing, review, and conducting the evaluations; JMJE: data analysis and writing; ICRF: data analysis and writing; JOP: statistical analysis and bibliographical review; RPP: intellectual concept, review, and conducting the evaluations; RSC: review and conducting the evaluations; OBN: writing, review, and preparation of the entire research project. All authors reviewed and approved the final version of the manuscript.

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