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# Effects of Yerba Mate Consumption (*Ilex paraguariensis*) on Cardiac Remodeling in Rats

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## Abstract

This study investigated the effects of yerba mate consumption, a South American beverage, on cardiac remodeling in rats. For this purpose, 24 male Wistar rats were divided into Control Group (CG) which received filtered water and a standard diet, and Yerba Mate Group (YM), 6 g of *Ilex paraguariensis* in 100 ml water and the same diet, for 30 days. The YM group showed a reduction in final body weight and food consumption without altering weight gain. Regarding cardiac remodeling, the YM group exhibited a decrease in the right ventricle weight/final body weight ratio, suggesting cardiac atrophy, without affecting the atria and left ventricle. There was no change in cardiomyocyte area or nuclear fractal dimension in both groups. However, animals that consumed yerba mate showed increased collagen deposition and a smaller fractal dimension in the left ventricle. The consumption of yerba mate at room temperature for 30 days induced changes in cardiac remodeling, as evidenced by increased collagen deposition and alterations in fractal dimension in the left ventricle.

## Keywords

Tereré, *Ilex paraguariensis*, Yerba Mate, Cardiac Remodeling, Fractal Dimension, Hypertrophy

## 1. Introduction

The plant *Ilex paraguariensis* A. St.-Hil, commonly known as yerba mate, belongs to the Aquifoliaceae family and is indigenous to South American countries, including Brazil, Paraguay, Argentina, and Uruguay. Predominantly consumed as an infusion, yerba mate has been the subject of various studies highlighting its therapeutic effects [1]-[3]. These effects encompass antioxidative, anti-inflammatory, antihypertensive, and anti-atherosclerotic properties, contributing to its potential as a beneficial botanical agent [4]-[10].

*Ilex paraguariensis* is rich in phenolic compounds, with caffeic acid being the major constituent, followed by saponins and methylxanthines. Its noteworthy feature lies in the antioxidant effects attributed to these compounds, capable of neutralizing reactive species and inducing the expression of antioxidant enzymes [2] [4] [11]-[15]. Furthermore, chlorogenic acid, found abundantly in the plant, exhibits therapeutic potential for cardiovascular diseases [4] [16].

Although the caffeine concentration in yerba mate is not as abundant as caffeic acid and chlorogenic acids, concerns persist regarding the risk of cardiovascular disorders due to caffeine present in yerba mate [4]. This includes the potential to induce arrhythmias, elevate blood pressure, and cause coronary vasospasm, in addition to affecting the metabolism of vitamin B-6, leading to an increase in plasma homocysteine concentration and subsequently elevating the risk of cardiovascular diseases [4] [17] [18]. The cardiovascular diseases are the leading cause of death globally and they also generate a great impact on direct hospitalization costs and indirect costs due to reduced productivity due to absence from work. So, the cardiovascular diseases are not only a health problem but also an economic problem [19]. The heart diseases have a common pathological process the cardiac remodeling (CR).

Due to these divergent results in relation to yerba mate, a comprehensive approach to aspects related to cardiac remodeling is essential. Cardiac remodeling (CR) can be conceptualized as a process involving molecular, cellular, biochemical, and interstitial modifications in cardiac tissue due to genomic alterations triggered by mechanical, biochemical, genetic, and humoral factors affecting the organ [20]. These factors are responsible for instigating and regulating changes in the heart, such as alterations in mass, volume, shape, composition, and cardiac function. Initially, CR may contribute to maintaining organ stability in cases of injury, assuming an adaptive character. However, the persistence of this process leads to progressive ventricular functional deterioration, cardiac hypertrophy, arrhythmias, and heart failure, resulting in a poor prognosis for the patient and potential mortality. Recognizing the consequences of CR, it is crucial to identify this process early on [20] [21].

There is a scarcity of studies evaluating yerba mate consumption in vivo regarding cardiac remodeling aspects. Therefore, this research investigated the potential effects of daily yerba mate consumption for 30 days on anatomical and histological aspects of cardiac remodeling in rats.

## 2. Materials and Methods

### 2.1. Animal Model and Experimental Design

For the experiment, 24 male Wistar rats (8 weeks old) with an average weight of 200 g were obtained from the Laboratory of Animal Experimentation at the University of Western São Paulo (UNOESTE) in Presidente Prudente, SP. The study was conducted following the ARRIVE guidelines [22].

The animals were housed in polypropylene plastic cages lined with wood shavings (2 rats per cage), with a temperature maintained between 20˚C to 24˚C and relative humidity between 45% and 65%. Light cycles consisted of 12 hours of darkness (from 7:00 pm to 7:00 am) and 12 hours of light (from 7:00 am to 7:00 pm). This study was approved by the Animal Use Ethics Committee of the University of Western São Paulo (Protocol 5300) and followed all ethical standards governed by the Brazilian College of Animal Experimentation.

All animals underwent a 7-day adaptation process before the experiment. After this period, they were randomly divided into two groups, each consisting of 12 animals: Control Group (CG), where animals received only filtered water and ad libitum balanced diet, and Yerba Mate Group (YM), where animals received a commercial balanced diet (Supralab®, Alisul, Rio Grande do Sul, Brazil) and filtered water at room temperature with *Ilex paraguariensis* ad libitum for a period of 30 days. The herb ingestion protocol can be observed in other studies [15] [23]. A schematic representation of the experimental design is shown in **Figure 1**.

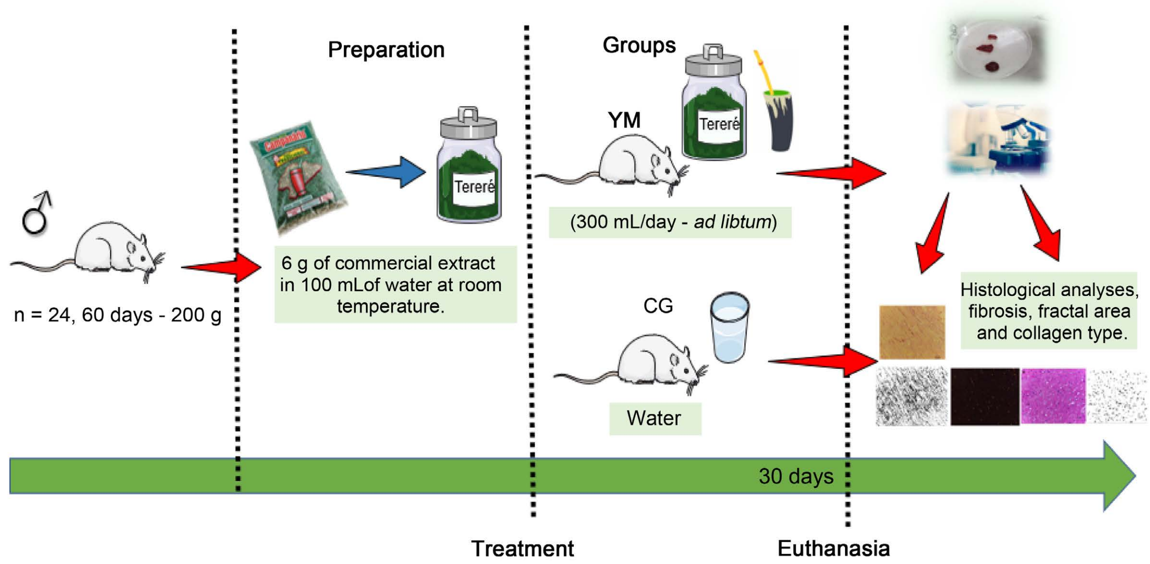


**Figure 1.** Experimental design. Groups: YM: Yerba Mate; CG: Control Group.

### 2.2. Yerba Mate Preparation

For the extraction process, commercial yerba mate from the brand Campanário Tradicional (Elervin Ind e Com Alimentos LTDA, MS, Brazil) containing different parts of the plant, from leaves to stem pieces, was used (**Table 1**). The aqueous extract was prepared at a ratio of 100 mL of room temperature water for every 6 g of *Ilex paraguariensis*, with these measurements performed using an analytical digital scale. Additionally, following the recommendations of Fioroto *et al.* (2022), this ratio aims to mimic traditional yerba mate consumption24. After preparation, the mixture rested for 15 minutes, allowing the substances present in the herb to dissolve into the water. Subsequently, it was filtered through a cloth filter, with 300 mL of the aqueous extract distributed per water dispenser. The preparation was changed daily, and the consumed volumes were recorded, along with the average food intake.

**Table 1.** Nutritional information of the commercial yerba mate.

| Amount per 200 mL Serving of Yerba Mate | | % DV |
|---|---|---|
| Caloric Value | 4.8 Kcal | 0.4% |
| Carbohydrates | <0.1 g | 0.0% |
| Proteins | 1.2 g | 1.2% |
| Total Fat | <0.5 g | 0% |
| Dietary Fiber | 6.24 g | 23% |
| Sodium | 0.50 g | 0% |

% DV: Daily Values based on a 2500-calorie diet.

### 2.3. Sample Collection

Following a 30-day period and a 10-hour fasting period, all rats underwent weighing and subsequent anesthesia via intraperitoneal administration of xylazine hydrochloride (10 mg/kg body weight at a concentration of 20 mg/mL) and ketamine hydrochloride (solution of 75 mg/kg body weight at a concentration of 100 mg/mL). Anesthesia was carefully induced, and vital signs confirming death (absence of cardiac activity, respiratory movements, and loss of neural reflexes) were meticulously assessed and verified, establishing the demise of the specimens. The heart was then extracted, dissected into atria, right ventricle, and left ventricle, and precisely weighed. Subsequently, the ventricular specimens were meticulously preserved in plastic containers immersed in a 10% formalin solution for subsequent histological analyses.

### 2.4. Phytochemical Screening of Yerba Mate

#### 2.4.1. Total Phenolic Content

The total phenolic content was determined using the Folin-Ciocalteu colorimetric method, as described by Ryu *et al.*, 2017, with modifications [24]. A small quantity (250 μL) of the crude tea preparation method and 150 μL of Folin-Ciocalteu reagent (Sigma) were thoroughly mixed. Then, 1 mL of $Na_2CO_3$ (10%) was added, followed by dilution to 5 mL with water. The mixture was kept in the dark at room temperature for 60 minutes. Absorbance was measured at 760 nm using a UV spectrophotometer (UV-1800, Shimadzu, Kyoto, Japan). The phenolic content was then calculated using a gallic acid calibration curve (mg GAE/g).

#### 2.4.2. Total Flavonoid Content

The total flavonoid content of the extracts was determined as described by Ryu *et al.*, 2017 [24]. A tea sample (200 µL) was added to 4 mL of distilled water and 300 µL of 5% $NaNO_2$ in a vial. The samples were allowed to stand for 5 minutes, and then 300 µL of 10% $AlCl_3$ was added. After 6 minutes, 2 mL of NaOH was added, and the volume was adjusted to 10 mL with distilled water. Absorbance was then measured at 510 nm. The total flavonoid content was calculated using a quercetin equivalents calibration curve (mg QE/g).

#### 2.4.3. Vibrational Spectroscopy in the Infrared Region (FTIR)

Infrared Vibrational Absorption Spectroscopy measurements with Fourier Transform were performed using a Bruker Tensor 27 instrument equipped with support for KBr pellets and a KBr window cell for liquid sample measurements. The measurements were conducted in the range of 400 to 4000 $cm^{-1}$ using 120 scans at a resolution of 4.0 $cm^{-1}$.

### 2.5. Antioxidant Activity *in Vitro*

The antioxidant activity of DPPH was tested using the method described by Rząsa-Duran *et al.*, (2022), with the DPPH radical (2,2-diphenyl-1-picrylhydrazyl) [25]. A volume of 0.1 mL of yerba mate infusion was analyzed, along with negative control (saline) and positive control (Quercetin 300 µg/mL). The respective concentrations (0.1 mL) were mixed with 4.9 mL of 0.1 mM DPPH dissolved in 80% methanol, in 15 mL Falcon tubes covered with aluminum foil, in triplicate. The reaction mixture was shaken and then incubated in the dark at room temperature for 15 minutes. The solution's absorbance was measured at 517 nm against the blank using a UV/VIS spectrophotometer. The antioxidant activity was calculated as DPPH [%] = $[(A_0 - A_1)/A_0] \times 100$, where $A_0$ and $A_1$ are the absorbance of reference and test solutions, respectively.

### 2.6. Analysis of Anatomical Parameters

After euthanasia, the hearts of the animals were removed and individually weighed. The weights of the right and left ventricles, as well as the atria, were normalized to their final body weights, used as an index of ventricular hypertrophy [26].

### 2.7. Histological Analysis of Cardiomyocytes

Cardiac tissue samples were fixed in a 10% formalin solution and then stored in paraffin blocks, allowing for histological analysis of coronal sections with a thickness of 4 micrometers. Hematoxylin-Eosin (HE) staining was used for the slides, enabling the measurement of cross-sectional areas of cardiomyocytes. Slide analysis was conducted using a LEICA DMLS microscope (DM750, Leica Microsystems, Wetzlar, Germany) at 100× magnification. Cuts were captured in videos, and digital images were transferred to computers using the Image-Pro-Plus image analysis software. For each animal, fifty transverse cardiomyocytes were selected, and their areas were measured [27].

## 2.8. Fibrosis Indicators

After fixation in a 10% formalin solution, tissue samples were stored in paraffin blocks, allowing for histological analysis of coronal sections with a thickness of 4 micrometers. Picrosirius Red (PSR) dye was used for staining, following the protocol established by the laboratory responsible for histological analysis. PSR staining, along with the use of ImageJ software, enabled fibrosis assessment through collagen quantification [28].

## 2.9. Fractal Dimension

For the analysis of the fractal dimension of the left ventricle (LV), hematoxylin and eosin-stained slides, as well as picrosirius-stained slides, were photographed and subjected to binarization for reading. Fractal dimension estimation was performed using the box-counting method, utilizing Image J software (National Institutes of Health, United States—NIH), freely available on the internet (http://rsbweb.nih.gov/ij/).

The software employs box-counting in two dimensions, allowing the quantification of pixel distribution in space without considering image texture. This means that two images with the same pixel distribution, one binarized and the other in grayscale, will exhibit the same fractal dimension. Thus, fractal analysis of histological slides is based on the relationship between resolution and the evaluated scale. The result is quantitatively expressed as the object's fractal dimension, calculated by the formula DF = (log Nr/log r−1), where Nr represents the quantity of elements needed to fill the original object, and r is the scale applied to the object (**Figure 1**).

Therefore, the fractal dimension calculated using Image J software varies between 0 and 2, making no distinction between different textures [26] [28].

## 2.10. Statistical Analyses

The normality of the data was assessed using the Shapiro-Wilk test. For parametric data, the unpaired Student's t-test will be employed. For non-parametric data, the Mann-Whitney test was utilized, followed by the Dunn post-test. Data were expressed as mean ± standard deviation, median, minimum, and maximum. DPPH data were expressed as mean ± standard deviation. One-way ANOVA followed by Tukey's post-hoc test was employed. GraphPad Prism software was used for analysis. The significance level for consideration was set at

$p < 0.05$.

## 3. Results

### 3.1. Phytochemical Analysis of Yerba Mate

The mean spectrum of the sample is depicted in **Figure 2**. The prominent bands observed in the spectral range of 900 to 1700 cm$^{-1}$ are assignable to the vibrational modes of OH groups, acids, and alcohols inherent in compounds such as caffeine [29], phenols [30], and saponins [31], among others. The band situated in the 1658 to 1700 cm$^{-1}$ region arises from the stretching vibration of CH, NO, C-H, and C=C groups [32]. Simultaneously, the spectral region spanning from 923 to 1391 cm$^{-1}$ is attributed to the carbonyl group in esters, amides, acids, and other compounds like xanthines and saponins found in yerba mate [33].

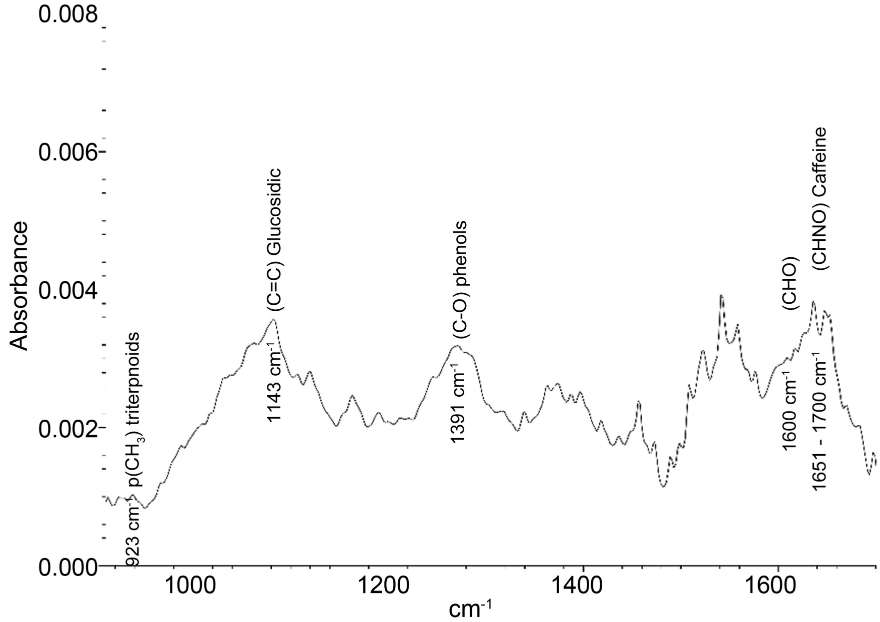


**Figure 2.** Reflectance spectra of commercial yerba mate.

It is evident from the phytochemical screening that a substantial amount of phenolic compounds was detected, approximately 86.14 ± 0.35 mg of gallic acid equivalent per gram of the herb. Additionally, the presence of flavonoids in the sample was confirmed, with a content of 0.53 ± 0.21 mg of quercetin equivalent per gram of the herb (**Table 2**).

**Table 2.** Phitochemical screening (mg/g).

| Total Phenols (mgEAG/g) | Total Flavonoids (mgEQ/g) |
|---|---|
| 86.14 ± 0.35 | 0.53 ± 0.21 |

EAG: Equivalent in Galic Acid; EQ: Equivalent in Quercetin.

Concerning the in vitro antioxidant assay (DPPH), it is possible to observe that the ability of Yerba Mate (EM) to scavenge the DPPH radical and reduce it was relatively high, with a mean of 76.59% ± 0.43% (Figure 3).

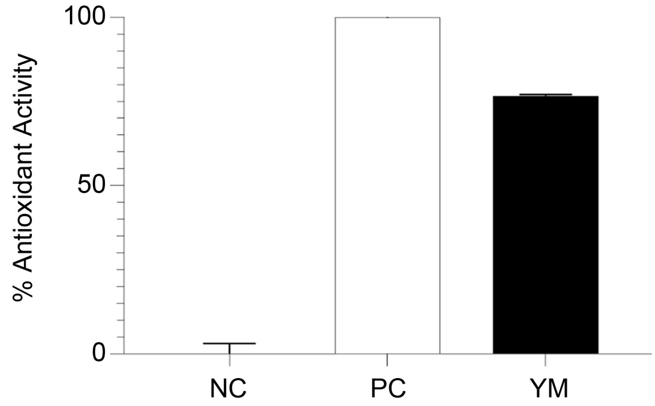


**Figure 3.** In vitro Antioxidant Activity (DPPH [%]). NC: negative control; PC: positive control; YM: Yerba Mate. One-way ANOVA followed by Tukey's post-hoc test $p < 0.05$.

## 3.2. Anatomical Data

Regarding the final body weight (Final BW), the CG group exhibited a mean of 435 ± 45.80 g, while the YM group showed a reduction to 400 ± 21.45 g ($p < 0.05$), with no alteration in weight gain. Although the animals consumed the same amount of liquids, yerba mate consumption led to a decrease in food intake compared to the control group (CG = 33.80 ± 1.80 g, YM = 31.83 ± 1.09 g, $p < 0.05$) (Table 3).

**Table 3.** Comparative analysis between Control Groups (CG) and experimental groups (YM) on body weight, weight gain, water consumption, and food intake variables.

| | Experimental Groups | | |
|---|---|---|---|
| **Variables** | | | |
| | **CG (n = 12)** | **YM (n = 12)** | ***p* value** |
| IBW (g) | 373 ± 37.79 | 352.1 ± 21.71 | 0.11 |
| FBW (g) | 435 ± 45.80 | 400 ± 21.45 | 0.02 |
| % Weight Gain | 17.26 ± 11.52 | 13.98 ± 7.49 | 0.06 |
| Liquid Consumption (mL) | 52.10 ± 4.72 | 59.73 ± 3.44 | 0.71 |
| Food Intake (g) | 33.80 ± 1.80 | 31.83 ± 1.09 | 0.006 |

IBW: Initial Body Weight; FBW: Final Body Weight. Unpaired Student's t-test.

Concerning hypertrophy indices, a notable reduction in the RV/FBW ratio is evident in rats subjected to yerba mate (CG = 0.64 ± 0.03 vs. YM = 0.56 ± 0.08 mg/g). The weights of the left ventricle (LV) and atria normalized by the final body weight remained unchanged (Figure 4).

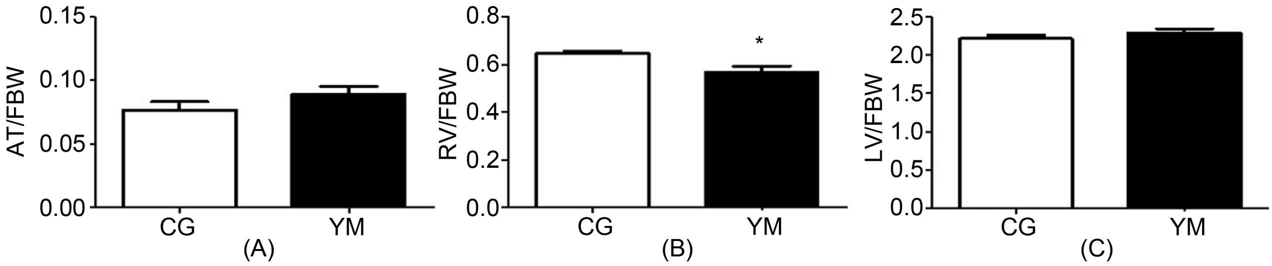


**Figure 4.** Cardiac anatomical data. (A) Atria-to-Final Body Weight Ratio; (B) Right Ventricle-to-Final Body Weight Ratio; (C) Left Ventricle-to-Final Body Weight Ratio. Unpaired t-test. *p < 0.05.

### 3.3. Histological Data

In the histological analyses conducted on the left ventricle, it is evident that there were no changes in the cardiomyocyte area (CG: 312.7 ± 77.6 μm$^2$ vs. YM: 314.9 ± 56.75 μm$^2$, p = 0.95) (**Figure 5**). Similarly, for fractal dimension analyses, no alterations were observed in both groups (CG: 1.52 ± 0.10 au; YM: 1.49 ± 0.10 au, p = 0.48) (**Figure 6**).

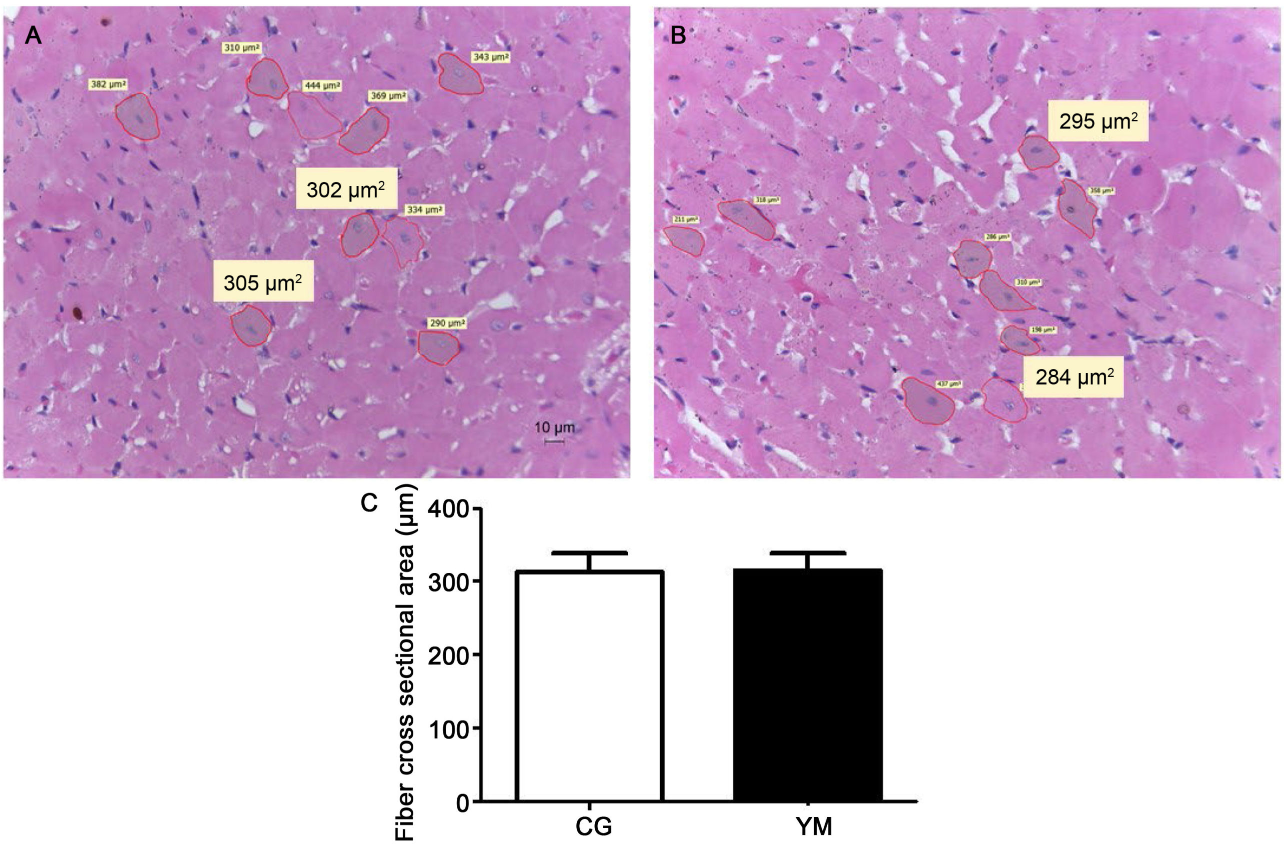


**Figure 5.** Area of cardiomyocytes located in the epicardial region of the left ventricle stained with hematoxylin-eosin at 40× magnification on the objective lens. (A) CT (n = 9): Control Group; (B) YM (n = 7): Yerba Mate Group; (C) Quantitative analysis of cardiomyocyte areas. Unpaired t-test.

There was an increase in collagen deposition in the left ventricle of animals subjected to yerba mate (CG: 3.53% ± 0.56%; YM: 4.59% ± 0.64%, p = 0.003), along with a reduction in fractal dimension (CG: 1.62 {1.51 - 1.73}; YM: 1.42 {1.30 - 1.55}; p = 0.001). Regarding collagen types, yerba mate consumption did not alter collagen type I (CG: 31.73 ± 10.39 au, YM: 26.90 ± 1.04 au) and type III (CG: 16.89 ± 8.7 vs. YM: 12.21 ± 1.09 au) (Figure 7).

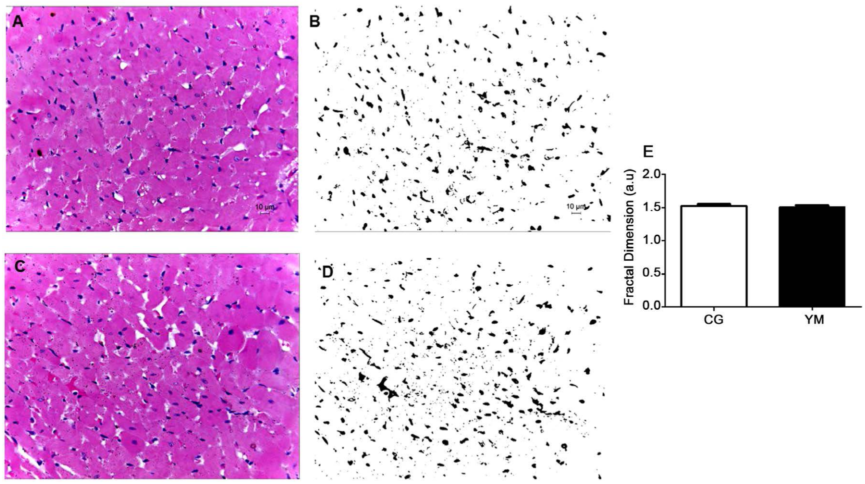


Figure 6. Histological sections of the left ventricle from the CG Group and YM Group treated with yerba mate, stained with H&E ((A) and (C)). Images in H&E after binarization process are shown in ((B) and (D)), where the nucleus appears in black, while the rest of the cell appears in white. In both groups, there were no changes in fractability (E). Unpaired t-test.

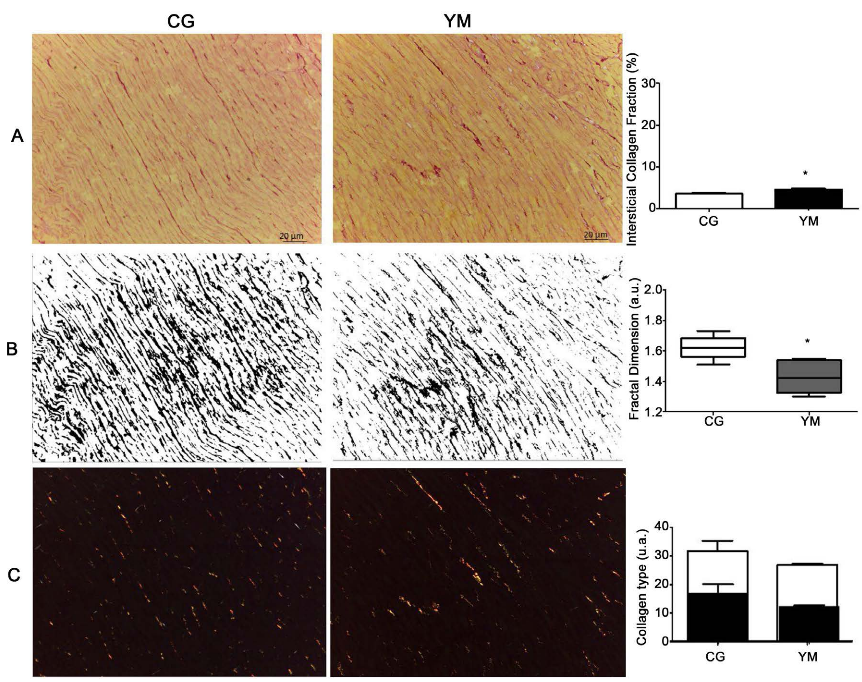


Figure 7. Analysis of fibrosis by identification of collagen deposition in cardiac tissue in CG and YM groups, stained with PSR (A). Images in PSR after the binarization process are shown in (B), where collagen appears in black, while the rest of the tissue appears in white. Additionally, (C) illustrates the distribution of collagen type I (red) and type III (green) in cardiac tissue. Unpaired t-test or Mann-Whitney test, *p = 0.001.

## 4. Discussion

In recent years, there has been a growing interest in research aimed at investigating the physiological and metabolic effects of natural products such as yerba mate. This product, highly prevalent in the culture of South American populations, contains bioactive compounds and stimulant substances like caffeine, as well as possessing antioxidant, anti-inflammatory, vasodilatory, antimutagenic, and hypolipidemic Properties [1] [22] [23] [34]-[40]. These positive effects are largely attributed to the high concentration of polyphenols in the plant [41]-[43].

The objective of the present study was to investigate the impact of yerba mate on the anatomical and histological structure of the heart following one month of consumption of the plant extract. To our knowledge, this study represents the first demonstration that yerba mate intake resulted in an increased deposition of cardiac collagen, accompanied by a reduced fractal dimension and atrophy of the right ventricle. No significant changes were observed in weight gain, left ventricle hypertrophy, or fractal dimension of the cellular nuclei.

In our study, despite a lower feed intake observed in the yerba mate group, there was no significant difference in weight gain. Other research findings diverge from our results, suggesting potential weight reduction benefits of yerba mate, particularly in obese rats. This disparity may be attributed to the presence of obesity and the duration of yerba mate usage [4] [44]-[47].

Regarding left ventricle hypertrophy, yerba mate did not induce any significant alterations. Consistent with our findings, other studies have reported no increase in left ventricle mass and have documented protective cardiovascular effects, including a reduction in systemic blood pressure and inhibition of apoptosis in cardiomyocytes [4] [48]. These beneficial effects have been attributed to the activation of the antioxidant system and the action of substances such as chlorogenic acid and other phenolic compounds, as observed in our study. Similar antioxidant effects were also demonstrated in vitro through the capture and reduction of the DPPH compound, aligning with findings from other studies [25]. However, in the right ventricle, the reduction in weight may indicate cardiomyocyte atrophy, potentially accompanied by the activation of catabolic pathways. Further studies are required to analyze this alteration, and morphometric analyses are deemed necessary.

We observed an increase in cardiac collagen with a reduced fractal dimension, without modifications in the types of collagens. The augmented collagen deposition in the heart has been previously demonstrated in studies and is directly associated with various alterations such as ischemia, infarction, hypertension, and heart failure [49]-[51]. These alterations may limit the contractile and relaxation capacity of cardiomyocytes, interfere with electrical conductivity, and impede nutrient distribution [52]. The types of collagens found in the heart include types I, III, IV, V, and VI, with types I (approximately 85%) and III (11%) being the most abundant in cardiac tissue. However, collagens type IV (in the basement membrane), V, and VI (in the middle and adventitia layers of muscular arteries and in the thin septa of connective tissue) can also be present but in smaller quantities [53] [54].

As our evaluation focused on the most prevalent types, namely I and III, this increase may be related to the types not assessed in this study. Further studies evaluating yerba mate and its cardiac repercussions are necessary, with a focus on functional analyses addressing systolic and diastolic function associated with collagen deposition.

Regarding fractal dimension, it was assessed using an innovative, simple, and cost-effective method, providing a precise analysis independent of observer bias of cellular organization. Fractal dimension has been employed to characterize cardiac phenotypic changes in pulmonary hypertension and has shown promise in detecting post-transplant cardiac alterations and differentiating between physiological cardiac hypertrophy and cardiomyopathy in athletes [26] [55] [56]. In cases of cardiac tissue hypertrophy, whether stimulated by a pathological condition or a natural physiological process, an increase in cardiomyocyte fractal dimension has been observed, indicating possible cardiac remodeling. In the present study, when evaluating fractal dimension focusing on cardiomyocyte nuclei, no nuclear irregularities were identified [57] [58]. However, when collagen deposition structure was assessed using picrosirius staining, a reduced fractal dimension was observed, indicating greater extracellular matrix disorganization.

In our study, we used crushed yerba mate prepared as an infusion with ambient temperature water, commonly known as tereré. The yerba mate production method (ground or crushed) and the preparation form (hot or ambient temperature water) directly influence phenolic compound concentrations, which, in turn, are associated with biological effects. Mate tea, prepared with hot water, showed higher concentrations of these phenolic compounds compared to the preparation with ambient temperature water [4]. However, it is important to note that in the present study, a semi-quantitative evaluation was conducted to assess the chemical composition of the infusion regarding total phenolic compounds. The total phenolic compounds found were similar to those in mate tea described by Rzasa-Duran *et al.* (2022), differing in the amount of flavonoids present [25]. Flavonoids are secondary metabolites whose pharmacological effects are well-described in the literature. When compared to an extraction using hot water, the lower amount of flavonoids found may be directly associated with the remodeling effects found in this study, as previously described cardioprotective effects may not be acting effectively.

These results emphasize the importance of considering the diversity in the chemical composition of yerba mate and the implications that these variations may have on the pharmacological and biological effects associated with its consumption. Such findings reinforce the need for in-depth investigations to fully understand the mechanisms of action and potential benefits of this plant in the cardiovascular context, given its widespread consumption, reported by approximately 70% of the South American population [11].

It is worth mentioning that numerous studies have been conducted to test and validate the cardioprotective effects of yerba mate in different preparations, supporting evidence suggesting a potential cardioprotective effect of the herb [1] [9] [11] [59]. However, our data suggest an increase in collagen deposition in cardiac tissue, indicating a possible adaptive response of the heart to yerba mate consumption [60]. It is important to note that despite the increase in collagen, its distribution was uniform, with no significant differences in the distribution of collagen in cardiac tissue or differences between types I and III, which are abundant in the fibrotic matrix of cardiac tissue. Therefore, this aspect needs further elucidation.

In the present study, it is possible that processes of repair or remodeling in cardiac tissue occurred in response to some stimulus present in the beverage. One compound commonly associated with minor changes in cardiac tissue, such as cardiomyocyte hypertrophy and arrhythmias, is caffeine [61]. Although the assay used in this study is an analytical method, high peaks of caffeine were detected, indicating a strong presence of the compound in the sample. In their analyses, Butt *et al.* (2019) observed similar caffeine characterization and peaks to those found in our study, with the analysis of their synthetic caffeine [32]. Marcelo *et al.* (2015) conducted a study characterizing yerba mate and its compounds, reporting the presence of caffeine and phenolic compounds directly in the 650 to 4000 $cm^{-1}$ region using reflectance [33]. de Lima *et al.* (2019) in their benchtop spectroscopy analyses provided a model with strong caffeine prediction, also similar to our analysis [62].

Research on the effects of yerba mate on the cardiovascular system is still limited due to cultural and regional differences in the consumption of these substances. Therefore, it is essential to encourage additional research to gain a better understanding of the mechanisms of action and impacts of yerba mate in the cardiovascular context. Studies evaluating the extracellular matrix associated with cardiac functionality and clinical trials involving yerba mate consumption are necessary.

## 5. Conclusion

The consumption of yerba mate for 30 days induced changes in cardiac remodeling, as evidenced by increased collagen deposition and alterations in fractal dimension in the left ventricle.

## Funding

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## Authors' Contributions

All authors had an essential role in formulation of the research questions; CEBP, LPG, FLP and TBM wrote the first draft of the paper, TBBP, DSMS and SCG analyzed the data; RCC, MFSW, RS, FLP and JARF were involved in interpretation of the data and revision of the manuscript. All authors have read and approved the final paper.

## Conflicts of Interest

The authors declare no conflicts of interest.

## References

[1] Cardozo Junior, E.L. and Morand, C. (2016) Interest of Mate (*Ilex paraguariensis* A. St.-Hil.) as a New Natural Functional Food to Preserve Human Cardiovascular Health—A Review. *Journal of Functional Foods*, **21**, 440-454. https://doi.org/10.1016/j.jff.2015.12.010

[2] Nagatomo, A., Inoue, N., Konno, T., Xu, Y., Sakamoto, C., Sone, M., *et al.* (2022) Ursane-Type Triterpene Oligoglycosides with Anti-Hepatosteatosis and Anti-Hyperlipidemic Activity from the Leaves of *Ilex paraguariensis* A. St.-Hil. *Journal of Natural Medicines*, **76**, 654-669. https://doi.org/10.1007/s11418-022-01614-5

[3] de Moraes Pontilho MPH, P., Mariana Nunes da Costa Teixeira MPH, A., Yuan BSc, C., Alves Luzia, L., Helena Markowicz Bastos, D. and Helen Rondó, P. (2015) Yerba Mate (*Ilex paraguariensis* A. St. Hil) and Risk Factors for Cardiovascular Diseases. *Journal of Food and Nutrition Research*, **3**, 182-190. https://doi.org/10.12691/jfnr-3-3-9

[4] Mesquita, M., Santos, E., Kassuya, C.A. and Salvador, M.J. (2021) Chimarrão, Terere and Mate-Tea in Legitimate Technology Modes of Preparation and Consume: A Comparative Study of Chemical Composition, Antioxidant, Anti-Inflammatory and Anti-Anxiety Properties of the Mostly Consumed Beverages of *Ilex paraguariensis* St. Hil. *Journal of Ethnopharmacology*, **279**, Article ID: 114401. https://doi.org/10.1016/j.jep.2021.114401

[5] Arçari, D.P., Bartchewsky, W., dos Santos, T.W., Oliveira, K.A., DeOliveira, C.C., Gotardo, É.M., *et al.* (2011) Anti-inflammatory Effects of Yerba Maté Extract (*Ilex paraguariensis*) Ameliorate Insulin Resistance in Mice with High Fat Diet-Induced Obesity. *Molecular and Cellular Endocrinology*, **335**, 110-115. https://doi.org/10.1016/j.mce.2011.01.003

[6] Clement, T.M., Savenkova, M.I., Settles, M., Anway, M.D. and Skinner, M.K. (2010) Alterations in the Developing Testis Transcriptome Following Embryonic Vinclozolin Exposure. *Reproductive Toxicology*, **30**, 353-364. https://doi.org/10.1016/j.reprotox.2010.05.086

[7] de Oliveira, E., Lima, N.S., Conceição, E.P.S., Peixoto-Silva, N., Moura, E.G. and Lisboa, P.C. (2018) Treatment with *Ilex paraguariensis* (Yerba Mate) Aqueous Solution Prevents Hepatic Redox Imbalance, Elevated Triglycerides, and Microsteatosis in Overweight Adult Rats That Were Precociously Weaned. *Brazilian Journal of Medical and Biological Research*, **51**, e7342. https://doi.org/10.1590/1414-431x20187342

[8] Farias, I.V., Fratoni, E., Theindl, L.C., de Campos, A.M., Dalmarco, E.M. and Reginatto, F.H. (2021) In Vitro Free Radical Scavenging Properties and Anti-Inflammatory Activity of *Ilex paraguariensis* (Maté) and the Ability of Its Major Chemical Markers to Inhibit the Production of Proinflammatory Mediators. *Mediators of Inflammation*, **2021**, Article ID: 7688153. https://doi.org/10.1155/2021/7688153

[9] Santa-Helena, E., Castro, M., Victoria, F.N., Rodrigues, J.S. and Gonçalves, C.A.N. (2022) Consumption of Mate *Ilex paraguariensis*: A Folk Beverage with Antioxidant Power against Myocardial Ischemic Injury. *Acta Scientiarum. Health Sciences*, **44**, e55845. https://doi.org/10.4025/actascihealthsci.v44i1.55845

[10] Santiago, P.G., Gasparotto, F.M., Gebara, K.S., Bacha, F.B., Lívero, F.A.D.R., Strapazon, M.A., *et al*. (2017) Mechanisms Underlying Antiatherosclerotic Properties of an Enriched Fraction Obtained from *Ilex paraguariensis* A. St.-Hil. *Phytomedicine*, **34**, 162-170. https://doi.org/10.1016/j.phymed.2017.08.012

[11] da Veiga, D.T.A., Bringhenti, R., Copes, R., Tatsch, E., Moresco, R.N., Comim, F.V., *et al*. (2018) Protective Effect of Yerba Mate Intake on the Cardiovascular System: A Post Hoc Analysis Study in Postmenopausal Women. *Brazilian Journal of Medical and Biological Research*, **51**, e7253. https://doi.org/10.1590/1414-431x20187253

[12] Ruskovska, T., Morand, C., Bonetti, C.I., Gebara, K.S., Cardozo Junior, E.L. and Milenkovic, D. (2022) Multigenomic Modifications in Human Circulating Immune Cells in Response to Consumption of Polyphenol-Rich Extract of Yerba Mate (*Ilex paraguariensis* A. St.-Hil.) Are Suggestive of Cardiometabolic Protective Effects. *British Journal of Nutrition*, **129**, 185-205. https://doi.org/10.1017/s0007114522001027

[13] Westphalen, D.J., Angelo, A.C., Rossa, U.B., Helm, C.V., Radetski, C.M. and Gomes, E.N. (2022) Phytochemical Composition of Yerba Mate Leaves (*Ilex paraguariensis*) and Its Relation with Cultivation Conditions. *Revista Brasileira de Plantas Medicinais*, **22**, 99-107.

[14] Contreras-Esquivel, J.C., Cano-González, C.N., Ascacio-Valdes, J., Aguirre-Joya, J.A., Aguillón-Gutierrez, D., Breccia, J., *et al*. (2022) Polyphenolic-Rich Extracts from the Leaves of *Ilex paraguariensis* and Larrea Divaricata and Their Antioxidant and Anticovid-19 Potential. *Biotecnia*, **25**, 61-66. https://doi.org/10.18633/biotecnia.v25i1.1762

[15] Muccillo-Baisch, A.L., Rafael de Moura, F., Penteado, J.O., Fernandes, C.L.F., Costa Bueno, E., Menestrino Garcia, E., *et al*. (2023) Beneficial Effects of Mate-Herb, *Ilex paraguariensis* St. Hil. against Potassium Dichromate-Induced Oxidative Stress and Nephrotoxicity. *Journal of Toxicology and Environmental Health*, *Part A*, **86**, 446-457. https://doi.org/10.1080/15287394.2023.2216231

[16] Colpo, A.C., de Lima, M.E., Maya-López, M., Rosa, H., Márquez-Curiel, C., Galván-Arzate, S., *et al*. (2017) Compounds from *Ilex paraguariensis* Extracts Have Antioxidant Effects in the Brains of Rats Subjected to Chronic Immobilization Stress. *Applied Physiology*, *Nutrition*, *and Metabolism*, **42**, 1172-1178. https://doi.org/10.1139/apnm-2017-0267

[17] Hartley, T.R., Lovallo, W.R. and Whitsett, T.L. (2004) Cardiovascular Effects of Caffeine in Men and Women. *The American Journal of Cardiology*, **93**, 1022-1026. https://doi.org/10.1016/j.amjcard.2003.12.057

[18] Berger, A.J. and Alford, K. (2009) Cardiac Arrest in a Young Man Following Excess Consumption of Caffeinated "Energy Drinks". *Medical Journal of Australia*, **190**, 41-43. https://doi.org/10.5694/j.1326-5377.2009.tb02263.x

[19] World Health Organization (WHO) (2023) Cardiovascular Diseases (CVDs). https://www.who.int/news-room/fact-sheets/detail/cardiovascular-diseases-(cvds)

[20] Karimi Galougahi, K., Ashley, E.A. and Ali, Z.A. (2015) Redox Regulation of Vascular Remodeling. *Cellular and Molecular Life Sciences*, **73**, 349-363. https://doi.org/10.1007/s00018-015-2068-y

[21] Cohn, J.N., Ferrari, R. and Sharpe, N. (2000) Cardiac Remodeling—Concepts and

Clinical Implications: A Consensus Paper from an International Forum on Cardiac Remodeling. *Journal of the American College of Cardiology*, **35**, 569-582. https://doi.org/10.1016/s0735-1097(99)00630-0

[22] Kilkenny, C., Browne, W.J., Cuthill, I.C., Emerson, M. and Altman, D.G. (2010) Improving Bioscience Research Reporting: The ARRIVE Guidelines for Reporting Animal Research. *PLOS Biology*, **8**, e1000412. https://doi.org/10.1371/journal.pbio.1000412

[23] de Resende, P.E., Kaiser, S., Pittol, V., Hoefel, A.L., D'Agostini Silva, R., Vieira Marques, C., *et al*. (2015) Influence of Crude Extract and Bioactive Fractions of *Ilex paraguariensis* A. St. Hil. (Yerba Mate) on the Wistar Rat Lipid Metabolism. *Journal of Functional Foods*, **15**, 440-451. https://doi.org/10.1016/j.jff.2015.03.040

[24] Ryu, J., Kwon, S., Ahn, J., Jo, Y.D., Kim, S.H., Jeong, S.W., *et al*. (2017) Phytochemicals and Antioxidant Activity in the Kenaf Plant (*Hibiscus cannabinus* L.). *Journal of Plant Biotechnology*, **44**, 191-202. https://doi.org/10.5010/jpb.2017.44.2.191

[25] Rząsa-Duran, E., Dobrowolska-Iwanek, E., Włodarczyk, J. and Zachwieja, D. (2023) Phenolic Profile, Antioxidant, Anti-Inflammatory, Anti-Angiogenic and Anti-Proliferative Properties of *Ilex paraguariensis* A. St.-Hil. Flowers Extract. *Molecules*, **28**, Article 2096.

[26] Pacagnelli, F.L., Sabela, A.K.D.D.A., Mariano, T.B., Ozaki, G.A.T., Castoldi, R.C., Carmo, E.M.D., *et al*. (2016) Fractal Dimension in Quantifying Experimental-Pulmonary-Hypertension-Induced Cardiac Dysfunction in Rats. *Arquivos Brasileiros de Cardiologia*, **107**, 33-39. https://doi.org/10.5935/abc.20160083

[27] Mariano, T.B., de Souza Castilho, A.C., de Almeida Sabela, A.K.D., de Oliveira, A.C., Cury, S.S., Aguiar, A.F., *et al*. (2021) Preventive Training Does Not Interfere with Mrna-Encoding Myosin and Collagen Expression during Pulmonary Arterial Hypertension. *PLOS ONE*, **16**, e0244768. https://doi.org/10.1371/journal.pone.0244768

[28] De Oliveira Mantovani, R., Pinheiro, D.G., De Oliveira, G.L.F., Perrud, S.N., Teixeira, G.R., Nai, G.A., *et al*. (2020) Effect of Different Doses of 2,4-Dichlorophenoxyacetic Acid (2, 4-D) on Cardiac Parameters in Male Wistar Rats. *Environmental Science and Pollution Research*, **28**, 3078-3087. https://doi.org/10.1007/s11356-020-10699-y

[29] Hamdani, H.E., Amane, M.E., Bouymajane, A. and Hamzaoui, N.E. (2023) Preparation, Spectral Characterization, Crystal Structure and Antibacterial Activity of Two New Supramolecular Complexes $[Ni(phen)_2Cl(H_2O)]_2(PF_6)_2$.2caf.$H_2O$(I), $[Ni(phen)_2(H_2O)_2]_2(PF_6)_4$. 3caf.4$H_2O$(II) Constructed via Hydrogen Bond Linking. *Journal of Molecular Structure*, **1274**, Article ID: 134342. https://doi.org/10.1016/j.molstruc.2022.134342

[30] Martin, D., Marques, J., Amado, A.M., Barroca, M.J., Moreira da Silva, A., Batista de Carvalho, L.A.E., *et al*. (2019) Shedding Light into the Health-Beneficial Properties of *Corema album*—A Vibrational Spectroscopy Study. *Journal of Raman Spectroscopy*, **51**, 313-322. https://doi.org/10.1002/jrs.5775

[31] Martin, D., Lopes, T., Correia, S., Canhoto, J., Marques, M.P.M. and Batista de Carvalho, L.A.E. (2021) Nutraceutical Properties of Tamarillo Fruits: A Vibrational Study. *Spectrochimica Acta Part A*: *Molecular and Biomolecular Spectroscopy*, **252**, Article ID: 119501. https://doi.org/10.1016/j.saa.2021.119501

[32] Butt, S., Hasan, S.M.F., Hassan, M.M., Alkharfy, K.M. and Neau, S.H. (2019) Directly Compressed Rosuvastatin Calcium Tablets That Offer Hydrotropic and Micellar Solubilization for Improved Dissolution Rate and Extent of Drug Release.

*Saudi Pharmaceutical Journal*, **27**, 619-628.
https://doi.org/10.1016/j.jsps.2019.03.002

[33] Marcelo, M.C.A., Pozebon, D. and Ferrão, M.F. (2015) Authentication of Yerba Mate According to the Country of Origin by Using Fourier Transform Infrared (FTIR) Associated with Chemometrics. *Food Additives & Contaminants: Part A*, **32**, 1215-1222. https://doi.org/10.1080/19440049.2015.1050702

[34] Amigo-Benavent, M., Wang, S., Mateos, R., Sarriá, B. and Bravo, L. (2017) Antiproliferative and Cytotoxic Effects of Green Coffee and Yerba Mate Extracts, Their Main Hydroxycinnamic Acids, Methylxanthine and Metabolites in Different Human Cell Lines. *Food and Chemical Toxicology*, **106**, 125-138.
https://doi.org/10.1016/j.fct.2017.05.019

[35] Bracesco, N., Sanchez, A.G., Contreras, V., Menini, T. and Gugliucci, A. (2011) Recent Advances on *Ilex paraguariensis* Research: Minireview. *Journal of Ethnopharmacology*, **136**, 378-384. https://doi.org/10.1016/j.jep.2010.06.032

[36] Boaventura, B.C.B., Di Pietro, P.F., Stefanuto, A., Klein, G.A., de Morais, E.C., de Andrade, F., *et al.* (2012) Association of Mate Tea (*Ilex paraguariensis*) Intake and Dietary Intervention and Effects on Oxidative Stress Biomarkers of Dyslipidemic Subjects. *Nutrition*, **28**, 657-664. https://doi.org/10.1016/j.nut.2011.10.017

[37] Riachi, L.G. and De Maria, C.A.B. (2017) Yerba Mate: An Overview of Physiological Effects in Humans. *Journal of Functional Foods*, **38**, 308-320.
https://doi.org/10.1016/j.jff.2017.09.020

[38] Kuropka, P., Zwyrzykowska-Wodzińska, A., Kupczyński, R., Włodarczyk, M., Szumny, A. and Nowaczyk, R.M. (2021) The Effect of *Ilex* × *meserveae* S. Y. Hu Extract and Its Fractions on Renal Morphology in Rats Fed with Normal and High-Cholesterol Diet. *Foods*, **10**, Article 818.
https://doi.org/10.3390/foods10040818

[39] Fayad, E., El-Sawalhi, S., Azizi, L., Beyrouthy, M. and Abdel-Massih, R.M. (2020) Yerba Mate (*Ilex paraguariensis*) a Potential Food Antibacterial Agent and Combination Assays with Different Classes of Antibiotics. *LWT*, **125**, Article ID: 109267.
https://doi.org/10.1016/j.lwt.2020.109267

[40] Santos, A.F., Schiefer, E.M., Surek, M., Martins, C.A.F., Worfel, P.R., Sassaki, G.L., *et al.* (2022) Chemical, Biological, and Pharmacological Evaluation of the Aqueous Extract of *Ilex paraguariensis*, St. Hill. (Aquifoliaceae). *Research*, *Society and Development*, **11**, e3011225335. https://doi.org/10.33448/rsd-v11i2.25335

[41] Biluca, F.C., da Silva, B., Caon, T., Mohr, E.T.B., Vieira, G.N., Gonzaga, L.V., *et al.* (2020) Investigation of Phenolic Compounds, Antioxidant and Anti-Inflammatory Activities in Stingless Bee Honey (*Meliponinae*). *Food Research International*, **129**, Article ID: 108756. https://doi.org/10.1016/j.foodres.2019.108756

[42] Olszowy, M. (2019) What Is Responsible for Antioxidant Properties of Polyphenolic Compounds from Plants? *Plant Physiology and Biochemistry*, **144**, 135-143.
https://doi.org/10.1016/j.plaphy.2019.09.039

[43] Torres-Fuentes, C., Suárez, M., Aragonès, G., Mulero, M., Ávila-Román, J., Arola-Arnal, A., *et al.* (2022) Cardioprotective Properties of Phenolic Compounds: A Role for Biological Rhythms. *Molecular Nutrition & Food Research*, **66**, Article ID: 2100990. https://doi.org/10.1002/mnfr.202100990

[44] Santos, D., Frota, E.G., Vargas, B.K., Tonieto Gris, C.C., Santos, L.F.D. and Bertolin, T.E. (2022) What Is the Role of Phenolic Compounds of Yerba Mate (*Ilex paraguariensis*) in Gut Microbiota? *Phytochemistry*, **203**, Article ID: 113341.
https://doi.org/10.1016/j.phytochem.2022.113341

[45] Choi, M., Park, H.J., Kim, S.R., Kim, D.Y. and Jung, U.J. (2017) Long-Term Dietary Supplementation with Yerba Mate Ameliorates Diet-Induced Obesity and Metabolic Disorders in Mice by Regulating Energy Expenditure and Lipid Metabolism. *Journal of Medicinal Food*, **20**, 1168-1175. https://doi.org/10.1089/jmf.2017.3995

[46] Lima, N.S., Franco, J.G., Peixoto-Silva, N., Maia, L.A., Kaezer, A., Felzenszwalb, I., de Oliveira, E., de Moura, E.G. and Lisboa, P.C. (2014) *Ilex Paraguariensis* (Yerba Mate) Improves Endocrine and Metabolic Disorders in Obese Rats Primed by Early Weaning. *European Journal of Nutrition*, **53**, 73-82. https://doi.org/10.1007/s00394-013-0500-3

[47] Gambero, A. and Ribeiro, M. (2015) The Positive Effects of Yerba Maté (*Ilex paraguariensis*) in Obesity. *Nutrients*, **7**, 730-750. https://doi.org/10.3390/nu7020730

[48] Pachura, N., Kupczyński, R., Lewandowska, K., Włodarczyk, M., Klemens, M., Kuropka, P., *et al*. (2022) Biochemical and Molecular Investigation of the Effect of Saponins and Terpenoids Derived from Leaves of Ilex Aquifolium on Lipid Metabolism of Obese Zucker Rats. *Molecules*, **27**, Article 3376. https://doi.org/10.3390/molecules27113376

[49] Amin, N.N., Grant, S.B., Yamrozik, J.A., Williams, R.B., Thompson, D.V., Doyle, M., *et al*. (2015) The Concordance Rates between LV Hypertrophy and RV Hypertrophy in Patients with Hypertrophic Cardiomyopathy as Diagnosed by Cardiovascular MRI with Fibrosis Imaging. *World Journal of Cardiovascular Diseases*, **5**, 171-180. https://doi.org/10.4236/wjcd.2015.57020

[50] Yin, X., Yin, X., Pan, X., Zhang, J., Fan, X., Li, J., *et al*. (2023) Post-Myocardial Infarction Fibrosis: Pathophysiology, Examination, and Intervention. *Frontiers in Pharmacology*, **14**, Article 1070973. https://doi.org/10.3389/fphar.2023.1070973

[51] Frangogiannis, N.G. (2019) The Extracellular Matrix in Ischemic and Nonischemic Heart Failure. *Circulation Research*, **125**, 117-146. https://doi.org/10.1161/circresaha.119.311148

[52] Cowling, R.T., Kupsky, D., Kahn, A.M., Daniels, L.B. and Greenberg, B.H. (2019) Mechanisms of Cardiac Collagen Deposition in Experimental Models and Human Disease. *Translational Research*, **209**, 138-155. https://doi.org/10.1016/j.trsl.2019.03.004

[53] de Souza, R.R. (2002) Aging of Myocardial Collagen. *Biogerontology*, **3**, 325-335. https://doi.org/10.1023/a:1021312027486

[54] Bashey, R.I., Martinez-Hernandez, A. and Jimenez, S.A. (1992) Isolation, Characterization, and Localization of Cardiac Collagen Type VI. Associations with Other Extracellular Matrix Components. *Circulation Research*, **70**, 1006-1017. https://doi.org/10.1161/01.res.70.5.1006

[55] Moreira, R.D., Moriel, A.R., Murta Junior, L.O., Neves, L.A. and Godoy, M.F.d. (2011) Dimensão fractal na quantificação do grau de rejeição celular miocárdica pós-transplante cardíaco. *Revista Brasileira de Cirurgia Cardiovascular*, **26**, 155-163. https://doi.org/10.1590/s0102-76382011000200004

[56] Vilades, D., Garcia-Moll, X., Gomez-Llorente, M., Pujadas, S., Ferrero-Gregori, A., Doñate, T., *et al*. (2021) Differentiation of Athlete's Heart and Hypertrophic Cardiomyopathy by the Fractal Dimension of Left Ventricular Trabeculae. *International Journal of Cardiology*, **330**, 232-237. https://doi.org/10.1016/j.ijcard.2021.02.042

[57] Guo, Y., Yin, H.J., Shi, D.Z. and Chen, K.J. (2005) Effects of Tribuli Saponins on Left Ventricular Remodeling After Acute Myocardial Infarction in Rats with Hyperlipidemia. *Chinese Journal of Integrative Medicine*, **11**, 142-146. https://doi.org/10.1007/bf02836472

[58] Kanno, Y., Watanabe, R., Zempo, H., Ogawa, M., Suzuki, J. and Isobe, M. (2013) Chlorogenic Acid Attenuates Ventricular Remodeling after Myocardial Infarction in Mice. *International Heart Journal*, **54**, 176-180. https://doi.org/10.1536/ihj.54.176

[59] González Arbeláez, L.F., Ciocci Pardo, A., Fantinelli, J.C., Schinella, G.R., Mosca, S.M. and Ríos, J. (2018) Cardioprotection and Natural Polyphenols: An Update of Clinical and Experimental Studies. *Food & Function*, **9**, 6129-6145. https://doi.org/10.1039/c8fo01307a

[60] Horn, M.A. and Trafford, A.W. (2016) Aging and the Cardiac Collagen Matrix: Novel Mediators of Fibrotic Remodelling. *Journal of Molecular and Cellular Cardiology*, **93**, 175-185. https://doi.org/10.1016/j.yjmcc.2015.11.005

[61] Shi, L., Xu, H., Wei, J., Ma, X. and Zhang, J. (2014) Caffeine Induces Cardiomyocyte Hypertrophy via P300 and CaMKII Pathways. *Chemico-Biological Interactions*, **221**, 35-41. https://doi.org/10.1016/j.cbi.2014.07.011

[62] de Lima, G.G., Ruiz, H.Z., Matos, M., Helm, C.V., de Liz, M.V. and Magalhães, W.L.E. (2019) Prediction of Yerba Mate Caffeine Content Using near Infrared Spectroscopy. *Spectroscopy Letters*, **52**, 282-287. https://doi.org/10.1080/00387010.2019.1622567