

DOES ENERGY DRINK INTAKE BEFORE EXERCISE AFFECT NONLINEAR DYNAMICS OF HEART RATE VARIABILITY RECOVERY? A RANDOMIZED, CROSSOVER, DOUBLE-BLIND AND PLACEBO-CONTROLLED TRIAL

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ABSTRACT

Introduction and Objectives: Energy drinks (ED) are recognized to influence the behavior of the sympathetic and parasympathetic components of the autonomic nervous system. We intended to study the influence of ED on non-linear heart rate variability (HRV) following exercise. **Material and Methods:** This randomized, crossover, double-blind, placebo-controlled clinical trial (Protocol number NCT02917889) was completed in a sample of 28 healthy males aged 24.11 ± 3.05 years (min-max 18-29). The first step involved the assessment of maximal oxygen consumption (VO_2 max). In the second protocol, the subjects received a placebo (250ml of water) or ED (250ml of energy drink) 15 minutes before the 30-minute exercise on a treadmill. In the third protocol, participants received the alternative protocol to the previous step. The nonlinear HRV were calculated at different times during the protocols. **Results:** Fractal analysis via Detrended Fluctuation Analysis (DFA) revealed that in the placebo protocol there was an increase in its values compared to recovery (Rec1) vs. Rest (Cohen's $d= 1.42$) and continued increasing in the last recording intervals: vs. Rec6 (Cohen's $d= 0.70$) and vs. Rec7 (Cohen's $d= 0.85$). In the ED protocol, the increase in DFA was only demonstrated when comparing Rec1 vs. Rest (Cohen's $d=1.78$). **Conclusion:** ED intake prior to modest aerobic exercise triggered a slight acceleration of recovery.

Key words: Energy drink. Exercise. Heart rate variability. Nonlinear dynamics.

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RESUMO

A ingestão de bebidas energéticas antes do exercício afeta a dinâmica não linear da recuperação da variabilidade da frequência cardíaca? Um ensaio randomizado, crossover, duplo-cego e controlado por placebo

Introdução e Objetivo: As bebidas energéticas (DE) são reconhecidas por influenciar o comportamento dos componentes simpáticos e parassimpáticos do sistema nervoso autônomo. Pretendemos estudar a influência da DE na variabilidade não linear da frequência cardíaca (VFC) após o exercício. **Material e Métodos:** Este ensaio clínico randomizado, cruzado, duplo-cego, controlado por placebo (número do protocolo NCT02917889) foi concluído em uma amostra de 28 homens saudáveis com idade de $24,11 \pm 3,05$ anos (min-max 18-29). A primeira etapa envolveu a avaliação do consumo máximo de oxigênio (VO_2 máx). No segundo protocolo, os sujeitos receberam placebo (250ml de água) ou ED (250ml de energético) 15 minutos antes do exercício de 30 minutos em esteira. No terceiro protocolo, os participantes receberam o protocolo alternativo à etapa anterior. A VFC não linear foi calculada em momentos diferentes durante os protocolos. **Resultados:** A análise fractal via Detrended Fluctuation Analysis (DFA) revelou que no protocolo placebo houve um aumento em seus valores em relação à recuperação (Rec1) vs. Repouso (Cohen's $d=1,42$) e continuou aumentando nos últimos intervalos de registro: vs. Rec6 (d de Cohen = $0,70$) e vs. Rec7 (d de Cohen= $0,85$). No protocolo ED, o aumento do DFA só foi demonstrado ao comparar Rec1 vs. Rest (Cohen's $d=1,78$). **Conclusão:** A ingestão de DE antes do exercício aeróbico moderado desencadeou uma ligeira aceleração da recuperação.

Palavras-chave: Bebida energética. Exercício. Variabilidade da frequência cardíaca. Dinâmica não linear.

INTRODUCTION

After physical exercise, changes in cardiovascular system aim to promote the body's physiological recovery to its baseline condition. Part of these changes are mediated by adjustments produced by the autonomic nervous system (ANS) (Guyton, Hall, 2011; Pastre and collaborators, 2009).

A rapid post-exercise heart rate (HR) deceleration is linked with good cardiovascular health and lower risk of developing adverse cardiovascular events and cardiovascular diseases (Romero, Minson, Hallwill, 2017; Cole and collaborators, 1999).

ANS activity can be prejudiced by a variety of factors, for instance the consumption of energy drinks (ED). ED are consumed globally and recognized for increasing the alert state and favoring the inclination to perform physical and mental activities.

From this, it was hitherto reported an increase in performance induced by ED and, so, there is a great use of ED in competitive sporting situations (Gutiérrez-Hellín, Varillas-Delgado, 2021).

Yet, some substances contained in ED are recognized to influence the behavior of the sympathetic and parasympathetic components of the ANS and cause antagonistic effects (Somers, Svatikova, 2020).

Caffeine, by blocking the action of adenosine, acts as an important stimulator of the central nervous system, besides increasing the sympathetic flow through the release of catecholamines from the adrenal medulla (Ribeiro, Sebastião, 2010) and is linked with delay in post-exercise ANS recovery (Somers, Svatikova, 2020; Benjamim and collaborators, 2021a).

In the study completed by Tank and collaborators (2007) it was revealed that the consumption of yohimbine promoted a decrease in the control of the heart rate baroreflex response in normotensive young people.

Additional components generally found in ED, such as ginseng, is similarly known for changing heart rhythm and causes QT interval prolongation (Torbey and collaborators, 2011).

One of the most reliable ways to assess ANS modulation is heart rate variability (HRV) analysis. In this procedure, the indices that translate the fluctuation of RR intervals (interval between consecutive heartbeats of the ECG) can be obtained via linear and non-linear

techniques. Linear indices have wide applications in studies that intended to stratify cardiovascular risk under the effect of certain interferences (Vanderlei and collaborators, 2009).

The behavior of nonlinear indices can better translate human systems owing to their complex dynamic nature (Vanderlei and collaborators, 2009; Godoy, Takakura, Correa, 2005).

Despite these accounts being widely described in the scientific literature, the study of the behavior of non-linear indices after physical exercise is rare. Hence, the study of the non-linear HRV after exercise may reveal other ways to evaluate the cardiovascular risk that the absorption of stimulant substances may impose.

Also, studying under these conditions can elucidate the knowledge concerning the behavior of the complexity of physiological systems.

Studies led by An, Park, Kim (2014), Porto and collaborators (2021) and Clark and collaborators (2020) evaluated the effects of ED intake on cardiac autonomic recovery after exercise in healthy young people and was unable to find significant effects on the HRV linear indices.

Still, the real impact of ED on nonlinear HRV recovery after effort is not totally understood. Together, the existing study aimed to investigate the influence of ED on cardiac nonlinear HR recovery after a single bout of exercise (Delliaux and collaborators, 2019).

MATERIALS AND METHODS

This is a randomized, crossover, double-blind, placebo-controlled clinical trial. The study was directed according to the CONSORT (Consolidated Standards of Reporting Trials) and is recorded in Clinical Trials.gov (Protocol number NCT02917889, <https://clinicaltrials.gov/ct2/show/NCT02917889>). In addition, this study was approved by the research ethics committee of the Faculty of Philosophy and Science of UNESP - campus of Marília, São Paulo state, under approval number 1.165.910.

Participants

We interviewed 55 males to participate in the experiments. Nevertheless, only 28 were physically active according to the International

Physical Activity Questionnaire (IPAQ) (Pardini and collaborators, 2001). We studied, in these participants, the existence of cardiorespiratory, neurological, musculoskeletal, renal, metabolic, endocrine and other medical conditions reported that would make it difficult to conform with the protocols (Figure 1).

CONSORT

TRANSPARENT REPORTING of TRIALS

CONSORT 2010 Flow Diagram

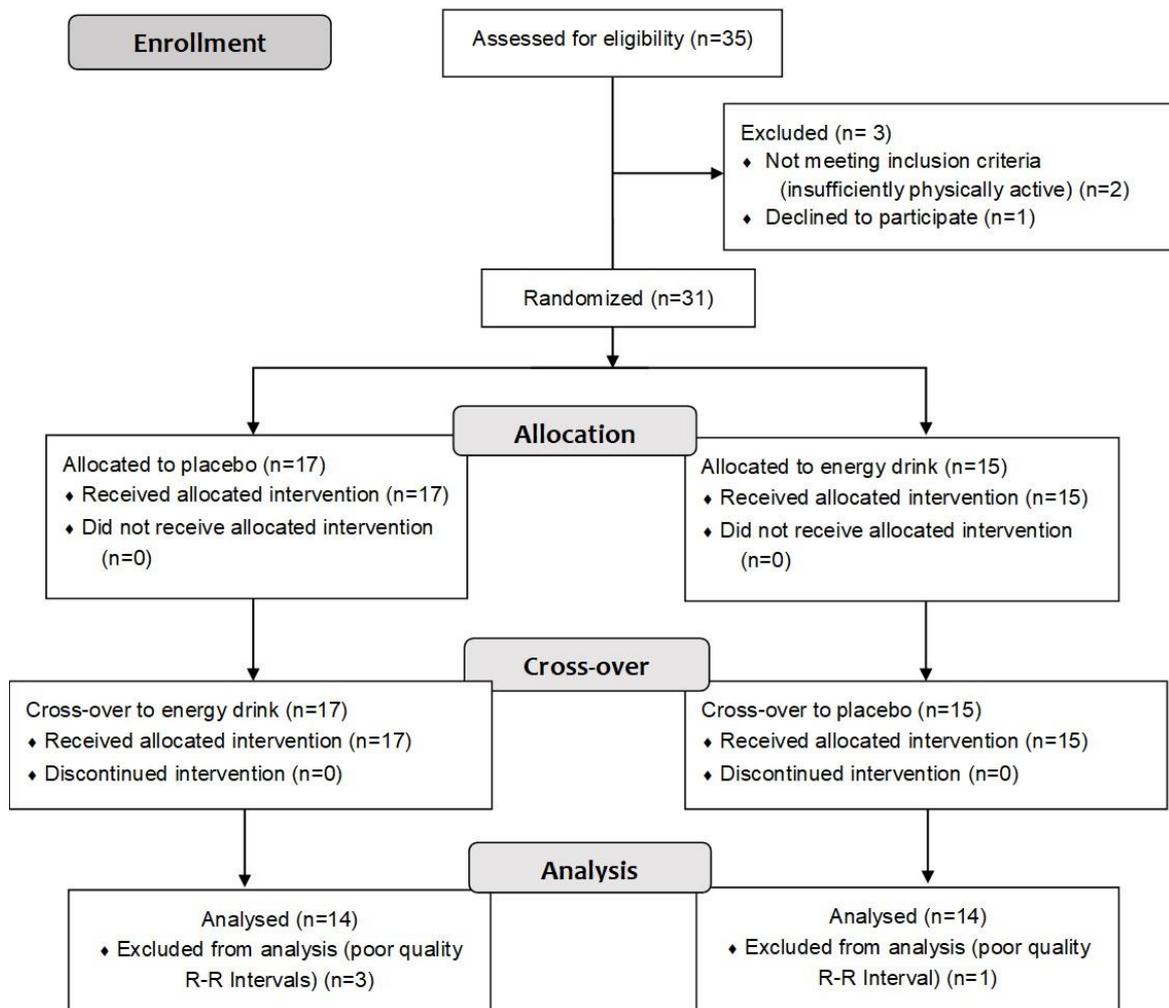


Figure 1 - Consort Flow Diagram

Initial Assessments

Data such as age (years), mass (kg), height (cm) and body mass index (kg/m²) were logged at an initial assessment. Anthropometric dimensions were obtained according to recommendations (Lohman and collaborators, 1992).

Interventions

The experimental protocols comprised of three stages and each stage was finalized with a minimum interval of 48 hours, so as to allow the subjects to recover properly. The study was performed between 5:30 pm and 9:30 pm, to standardize the circadian rhythm effects, in a silent room with humidity between 60% and 70% and temperature between 23°C and 24°C (Black and collaborators, 2019).

The subjects were told to refrain from drinking alcoholic beverages or performing exhaustive exercises 24 hours prior to each protocol and to avoid ingesting beverages or foods containing caffeine 24 hours before the experimental procedures. Individuals were told to wear suitable and comfortable clothes to permit the necessary physical effort and to have a light meal just two hours before the procedures.

Following the recommendations of the American College of Sports Medicine (ACSM), a hydration protocol was included in the study to avoid dehydration of participants throughout physical exercise and hence prevent dehydration from interfering in the collected data (Moreno and collaborators, 2013). Participants were instructed to drink 500 ml of water two hours prior to the experimental protocols.

The first step involved measuring VO₂ max, which was achieved on a treadmill (TPEE; Inbrasport ATL 2000) using the Bruce protocol (Novitsky and collaborators, 1995).

The subjects remained at rest on the treadmill in an orthostatic position to stabilize their initial values, then the exercise test was underway, whose protocol promoted a progressive increase in the workload by elevating the inclination and speed of the treadmill every three minutes. Verbal encouragement was enforced in an attempt to achieve maximum physical effort. The test was interrupted because of the subject's exhaustion or the appearance of clinical and/or electrocardiographic fluctuations that prohibited

the continuation of evaluation (Billat and collaborators, 2000).

During test implementation, heart rate and perceived exertion (RPE) were monitored at the end of each stage by the Borg Scale for perceived pain and exertion (Billat and collaborators, 2000).

For the test to be documented as a maximum, individuals needed to attain 90% of the earlier estimated maximum heart rate (220 - age) (Barroso and collaborators, 2021).

The analysis of expired gases was accomplished via the Quark PFT commercial system (Comend, Rome, Italy), which measured VO₂ peak as the highest VO₂ reached throughout the test. In the second phase of the intervention protocols, subjects took a placebo (placebo protocol-PP) (250ml water) or ED (ED protocol-EP) (250ml energy drink) 15 minutes before exercise. In the third phase, the participants received the inverse protocol to the previous step.

An independent researcher who did not partake in the study's data collection administered the drinks. The random sequence of the intervention was blinded to both researchers and subjects.

The ED (250ml) had 45 kcal and was comprised of 11.2 g carbohydrate, 80 mg sodium, 32 mg caffeine, 400 mg taurine, 4.6 mg niacin, 2 mg pantothenic acid, vitamin B6 0.5 mg, vitamin B12 0.4 mg, glucuronolactone 240 mg, Inositol 20 mg (An, Park, Kim, 2014).

Physical exercise intensity was agreed and founded on the VO₂ max. The treadmill test was performed for 30 minutes. The subjects completed aerobic exercise on a treadmill at a speed of 5km/h in the first session for 5 minutes of warm-up.

After this step, the speed was elevated to 60% of the VO₂ peak for 25 minutes. After the final exercise régime, the subjects were in the dorsal decubitus position for 60 minutes all through recovery.

HRV analysis

RR intervals were recorded beat-to-beat throughout the procedures using a HR monitor (Polar Model RS800CX, Finland) (Benjamim and collaborators, 2021b).

The recordings were undertaken for 5 minutes for each interval and performed during eight phases: Before Exercise (Rest): (10-15min); After Exercise (Rec): Rec1 (0-5min); Rec2 (5-10min); Rec3 (15-20min); Rec4 (25-

30min); Rec5 (35-40min); Rec6 (45-50min); and Rec7 (55-60min). These time series were combined with computations that represent non-linear HRV indices (Richman, Moorman, 2000) so as to understand the behavior of non-linear HRV recovery following physical exercise.

The non-linear HRV indices were computed using the PyBios® software (Biomedical Signal Analysis in Python version 1.2.0, 2020) developed at the School of Medicine of Ribeirão Preto, University of São Paulo, São Paulo, Brazil (Silva, Fazan, Marin-Neto, 2020).

The stationarity of the series was maintained by manual and visual filtering of artifacts. Data filtering was achieved respecting the correction of up to 2.5% of the time series. The default value adopted for the baseline window length was 10 and the tolerance value considered was 0.1 (Rincon Soler and collaborators, 2018).

HRV non-linear index

Symbolic analysis of HRV

Symbolic analysis was accomplished by distributing the number of RR intervals into six levels (0 to 5), which transforms them into a sequence of symbols, from which there is a spatial method (sequence of three symbols). All possible patterns were grouped into four clusters, individually, consistent with the number and type of variation between symbols: (1) 0V corresponds to no variation [three identical symbols, e.g. (2.2.2) or (4.4.4)]; (2) 1V corresponds to a variation [two consecutive symbols are equal and the remaining symbol is different, e.g. (4.2.2) or (4.4.3)]; (3) 2LV represents two similar variations [the three symbols ramp up or down, e.g. (5.4.2) or (1.3.4)]; (4) 2ULV represents two opposite variations [three symbols form a peak or trough, e.g. (3.5.3) or (4.1.2)].

Occurrence rates of these clusters (0V%, 1V%, 2LV% and 2ULV%) were examined (Porta and collaborators, 2007).

Earlier studies with pharmacological blockade identified that the 0V% index represents cardiac sympathetic modulation. The 1V% indices signify the global modulation of HRV, namely, the concurrent presence of sympathetic and vagal modulation. Lastly, the

2V% and 2UV% indices have the potential to infer cardiac vagal modulation.

Detrended Fluctuation Analysis (DFA)

To evaluate the fractal properties of the heart during the placebo and energy drink protocols, trendless fluctuation analysis (DFA) was enforced onto a time series of RR intervals attained from the experiments. The technique for calculating the DFA was as follows. Firstly, the RR time series obtained experimentally was integrated using the expression:

$$y(k) = \sum_{i=1}^k (RR(i) - RRave)$$

where $y(k)$ is the k^{th} term of the series integrated ($k=1,2, 3... N$); $RR(i)$ is the n^{th} value of the RR intervals; and $RRave$ is the mean of the RR intervals of the original time series, with length N .

The integrated time series was then split into intervals with a length of n , ($n=1,2, 3... N$). At each of these intervals, the local trend of the series was computed by a straight line of least squares fitted to the data. The y coordinate of this straight line is called $Yn(k)$. Each trend line is then subtracted from the signal $Y(k)$, generating an accumulated stretched series $Yn(k)$. To finish, the root mean square of the accumulated series is computed:

$$F(n) = \sqrt{\frac{1}{N} \sum_{k=1}^N [Y(k) - Yn(k)]^2}$$

This method was repeated for all intervals of size n , so obtaining a relationship between the mean of the fluctuations $F(n)$ and the size of the intervals (n). A linear association on a log-log graph indicates a scale exponent law, founded on the following mathematical formula:

$$F(n) \propto n^{\alpha}$$

Here, α is the scale exponent, which can be calculated by linear regression on a log-log graph. The spectrum of the short (α_1) and

long-term (α_2) fractal exponents of the DFA are computed. When $\alpha = 0.05$, there is no correlation and the signal comprises of white noise; if $\alpha = 1.5$, the signal resembles the random walk (Brownian motion); then if $0.5 < \alpha < 1.5$, there are positive correlations. If α is close to 1.0 it indicates a more complex (nonlinear) system, and if it attains values greater than 1.0 the system tends to be less complex and more linear (Makikallio and collaborators, 1999).

Sample Entropy (SampEn)

SampEn was presented to measure the complexity of the RR interval time series under different circumstances, such as the placebo and energy drink. SampEn is defined as the negative natural logarithm of the conditional probability that two similar sequences for 1 point remain similar at the next point within a tolerance r , where the auto-matches are excluded from the probability calculation. An irregular sequence will lead to higher SampEn values, whilst a regular and predictable signal is associated with lower SampEn.

The mathematical expression for Sample Entropy is:

$SampEn(r, l) = -\ln(A/B)$, where A and B are the total numbers of direct matches of length $l + 1$ and l respectively.

Theoretically, SampEn is independent of the length of the time series. While l and r

critically affect the result of SampEn, there are no procedures for their values optimal selection. Here we apply the two values $l = 2$ and $r = 0.15 \times SD$ (RR), where SD is the standard deviation of the 5-minute time series RR (Makikallio and collaborators, 1999; Goldberger and collaborators, 2000; Richman, Moorman, 2000).

Statistical analysis

Initially, the Gaussian distribution of the data was completed using the Shapiro-Wilk normality test (z value >1.0). For parametric distributions, one-way ANOVA (treatment versus recovery) was enforced for a repeated measures test and followed by Bonferroni post-test. For non-parametric distributions, the Friedman test was enforced followed by the Dunn's post-test (Laborde, Mosley, Thayer, 2017).

The effect size was computed by Cohen's d to measure the magnitude of differences for significant results (Quintana, 2017). We considered >1 a large effect size, between <1 to > 0.8 a medium effect size and between <0.8 to 0.5 a small effect size.

RESULTS

Sample profile

The anthropometric features and VO_2 peak values of the study participants are stated in Table 1.

Table 1 - Anthropometric characteristics and VO_2 peak value of the study volunteers.

	Média \pm DP	Min - Máx
Age (years)	24,11 \pm 3,05	[18 - 29]
Height (meters)	1,79 \pm 0,08	[1,65 - 1,94]
Weight (kg)	83,51 \pm 13,18	[60 - 107]
BMI (kg/m ²)	25,79 \pm 2,90	[20,05 - 29,41]
VO_2 peak (ml/kg/min)	50,94 \pm 12,53	[33,83 - 77,77]

SD = standard deviation; Min = minimum value; Max = maximum value; BMI = body mass index; kg = kilogram; m = meter; VO_2 peak = peak oxygen consumption; ml = milliliter; min = minutes.

Non-linear HRV (Rest versus Recovery)

In the PP, the 0V% index increased throughout the first recovery interval (Rec1) vs. rest (Cohen's $d = 2.52$); the similar effect was observed in the EP (Cohen's $d = 3.54$). In the

1V% index, there was a decline through Rec1 vs. rest (Cohen's $d = 1.55$) in the EP (Figure 2).

The 2V% index decreased between Rec1 vs. rest (Cohen's $d = 1.52$) in the PP. In the EP protocol this result was also observed (Cohen's $d = 2.0$). In the PP, the 2UV% index

reduced when comparing Rec1 vs. rest (Cohen's $d= 1.65$) and remained low during Rec5 (Cohen's $d= 0.50$) and Rec7 (Cohen's $d=$

0.78). In the EP, this index decreased in Rec1 vs. rest (Cohen's $d= 2.63$) (Figure 2).

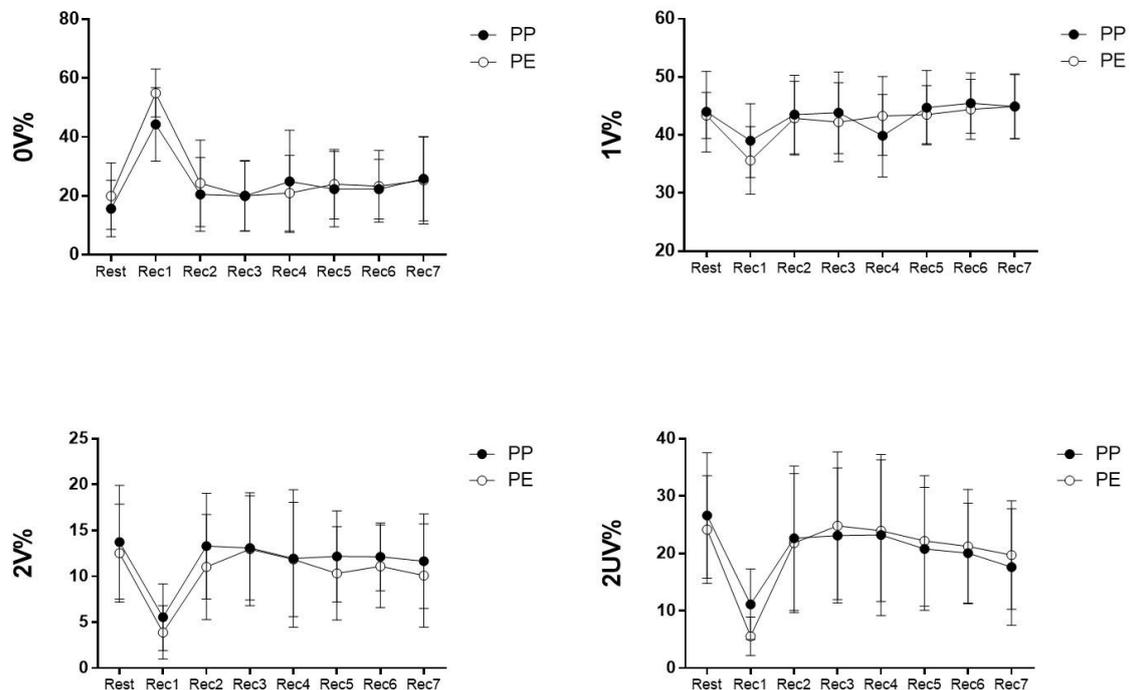


Figure 2 - Mean values and their respective standard deviations of Symbolic analysis indices during rest and recovery (Rec).

SampEn's analysis in the PP decreased in Rec1 vs. rest (Cohen's $d= 2.48$) and remained decreased throughout the final recovery interval (Rec7) (Cohen's $d= 0.81$). In the EP, SampEn declined in Rec1 vs. Rest (Cohen's $d= 3.13$) (Figure 3).

For the fractal analysis (via DFA) in the PP increased when comparing Rec1 vs. Rest (Cohen's $d= 1.42$) and kept increasing in the last recording intervals: Rec6 (Cohen's $d= 0.70$) and Rec7 (Cohen's $d= 0.85$). In the EP, the increase in DFA was only realized when comparing Rec1 vs. Rest (Cohen's $d= 1.78$) (Figure 3).

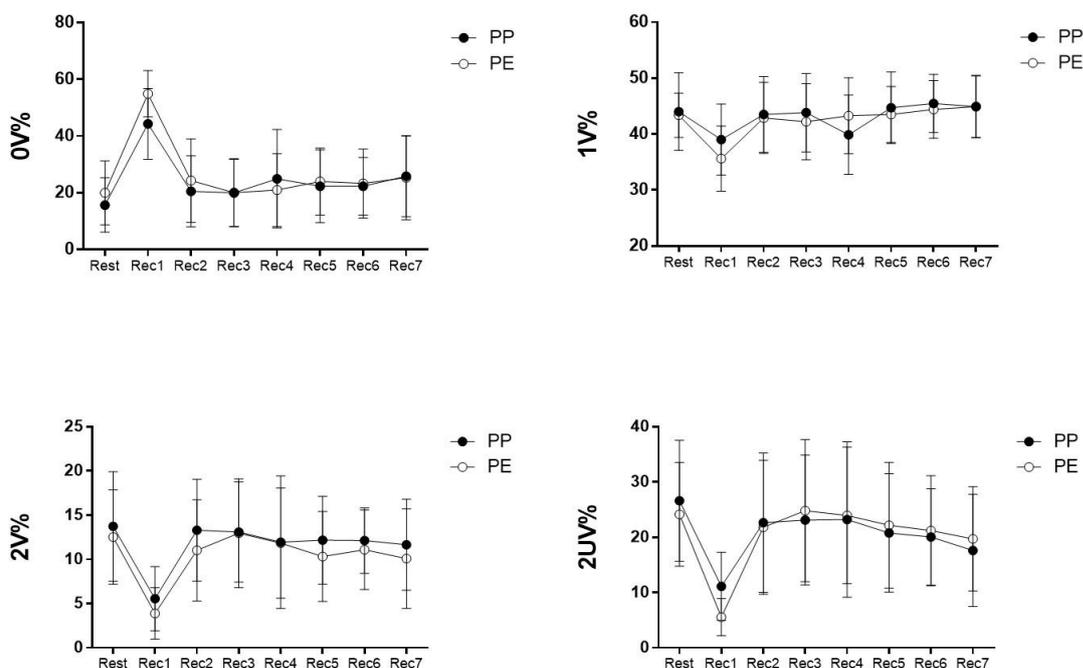


Figure 3 - Mean values and their respective standard deviations of Dfa and SampEn during rest and recovery (Rec)

DISCUSSION

Our results established that the acute intake of ED prior to physical exercise:

- a) intensified the recovery via symbolic analysis via 2UV%;
- b) supported in the recovery of fractal analysis (via DFA); and,
- c) intensified the recovery of the Sample Entropy metric (SampEn).

ED has caffeine as its key ingredient - via the antagonism of the angiotensin receptor, it is a stimulant that can impact the activity of neuronal control pathways in the central and peripheral nervous system (Jones, 2008).

Thus, caffeine is known for stimulating the central nervous system, it is able to promote the activation of the medullary adrenal sympathetic system and raise blood pressure via physiological stress situations, for instance physical exercise (Pincomb and collaborators, 1985; Lovallo and collaborators, 1989).

One more key ingredient in ED is taurine. Previous studies have detected that it suppresses the stimulation of sympathetic activity and acts by modulating the cyclic nucleotide content in cardiac cells (Mal'chikova, Elizarova, 1981).

Furthermore, taurine has been demonstrated to lessen the impact of experimental cardiac ischemia or reperfusion through inhibition of the systemic renin-angiotensin system and cardiac hyperactivity (Kulthinee, Wyss, Roysommuti, 2019).

The scientific research literature presents studies including ED consumption, exercise and HRV recovery. And collaborators (2014) assessed 15 young males. Participants undertook the Bruce test to monitor VO_2 max. After measuring the VO_2 max, the individuals consumed the ED or not, then were submitted to treadmill running at 80% of VO_2 max up to 90% of the maximum heart rate or surpassing the respiratory exchange ratio of 1.15.

Subjects were coached to walk for 60 seconds at 1.2 mph after discontinuing running and sitting on a chair to monitor the variables. After data analysis, they did not observe significant values of interaction between groups in HRV indices after physical exercise. These substantiate the findings presented in our earlier study (Porto and collaborators, 2021).

In a previous study directed in our laboratory, we submitted 29 healthy males aged between 18 and 30 years to three protocols:

- (i) VO_2 max measurement using an adapted Bruce test,

(ii) Placebo protocol - water intake 15 minutes prior to exercise, rest in decubitus position for 15 minutes after 5 minutes of running on a treadmill with an incline of 1%, initial speed of 5 km/h for 5 minutes, followed by 25 minutes with 60% speed compatible with VO_2 max and 60 minutes of recovery at rest in the supine position; and (iii) experimental protocol - similar to the placebo protocol mentioned above, but the individuals consumed ED.

Time domain, frequency and geometric indices were studied before and following effort. There was a significant effect ($p < 0.05$, or $< 5\%$) on the HRV index (HR-nu and ms2, LF-nu and ms2, LF/HF, SD1, SDNN and RMSSD), indicating a reduction in HRV in the first 5 minutes following exercise in both protocols, which can be attributed to exercise effects. Consequently, ED was unable to influence the post-exercise HRV recovery according to those data and metrics (Porto and collaborators, 2021).

With an exercise protocol different from those presented until now, Nelson and collaborators (2014) performed a randomized, placebo-controlled, crossover clinical trial in 15 individuals, wherein they investigated males ($n=8$) and females ($n=7$) with a mean age of 24 and 27 years old, who were active recreationally. Individuals consumed ED or placebo and were acquiesced to a 30-minute submaximal load on cycle ergometers. - Individuals completed 10 minutes at 80% of the ventilatory threshold and cycled until voluntary fatigue. Even though resting heart rate is statistically higher after ED than placebo (ED: 65 ± 10 bpm vs. placebo: 58 ± 8 bpm, $p = 0.02$), HRV metrics computed as RMSSD, SDNN, pNN50, HF, LF and LF/HF ratio were not significantly changed.

In the second clinical trial that similarly involves ED intake by males and females prior to exercise Clark and collaborators (2020) completed a double-blind, placebo-controlled and balanced study. Seventeen young participants (seven males and ten females) received an ED formula comprised of 140 mg caffeine and a placebo in random sequence before finishing a 10-minute steady-state warm-up (WUP) and a graded exercise test to exhaustion (GXT) on a stationary bicycle with an electromagnetic brake and subsequently a short 15-minute rest period (STR). Data were split into WUP, GXT and STR phases.

Significant increases in HF and RMSSD were established during WUP after ED

consumption. The study discovered that ED consumption influenced cardiac autonomic responses during low-intensity exercise, and there might be gender-based changes in response to gradual exercise to exhaustion. Still, no difference was observed between groups in the post-exercise rest period.

In the study directed by Porto and collaborators (2021), we recognize that ED did not stimulate the release of catecholamines sufficiently to delay the HRV recovery. The studies presented above explained that ED was unable to influence the post-exercise HRV recovery by evaluating the linear indexes.

Nonetheless, Delliaux and collaborators (2019) defined the cardiovascular system as a complex mechanism. So, it cannot be completely explained by linear methods alone. Then, suggested that to overcome this capability, non-linear methods are needed. Non-linear indices are described in the literature as more suitable for characterizing the dynamics and complexity of the cardiovascular system.

Caliskan and Bilgin (2020), assessed the effects of an ED with caffeine and cola on non-linear HRV indices via electrocardiograms in 48 participants (both genders). They detected that the group that ingested ED demonstrated an increase in the complexity of the cardiovascular system in adults. These results support our findings, even if it is vital to emphasize that our study, according to our knowledge, is the first in the research literature to clarify ED intake in the nonlinear HRV recovery.

This study presents some ideas worth highlighting. Our sample was entirely comprised of healthy males, so as to circumvent the influence of sexual hormones. Nonetheless, as a consequence, we cannot extrapolate the interpretation of our results to another population or group.

For further studies related to ED and physical exercise, we recommend considering some vital issues not covered in the current research literature: assessment of plasma catecholamine concentration, sympathetic nerve activities, association with oxidative stress, different sample profiles and different types of physical exercises.

Lastly, while we did not administer the quantities of caffeine proportional to the participant's mass and did not separate participants by body mass index, the study design permitted us to apply strict procedures

so as to reduce the vulnerability to selection, performance and other biases.

CONCLUSION

Our study stated that ED intake before exercise slightly enhanced nonlinear HRV recovery.

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The authors declare absence of financial and non-financial interests.

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