Assessment of Autonomic Dysregulation in Sepsis and Sinoaortic Denervation by Analysis of Cardiovascular Variability and Complexity

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Abstract

Sinoaortic denervation (SAD) and sepsis both disrupt autonomic nervous system (ANS) control of cardiovascular (CV) function, but their shared and distinct effects remain unclear. This study compared ANS alterations induced by SAD and cecal ligation and puncture (CLP)–induced sepsis in Wistar rats. Invasive arterial pressure was recorded from a femoral catheter in three groups (n = 3 each): SAD, Sepsis, and Sham. From the pressure waveform, systolic (SAP), diastolic (DAP), and heart period (HP) series were derived. ANS modulation was assessed using (a) sample and approximate entropy for signal complexity, (b) spectral power in VLF, LF, and HF bands for variability indices, and (c) baroreflex sensitivity (BRS) via bivariate autoregressive models. Group differences were tested by two-way ANOVA with Tukey post-hoc.Entropy analysis showed higher DAP and HP unpredictability in Sham and Sepsis, whereas SAD exhibited greater SAP complexity ($p < 0.05$). SAD animals also differed in VLF and HF power of SAP and DAP, while the LF component of BRS showed high intragroup variability; sepsis suppressed BRS responses ($p < 0.05$). Integrating nonlinear and linear analyses may clarify mechanisms of ANS dysregulation in sepsis and acute illness.

1. Introduction

The autonomic nervous system (ANS) is a major player in the regulation of cardiovascular (CV) function [1,2], that can be disrupted by several physiological and pathological stimuli. For instance, both sinoaortic denervation (SAD) and sepsis significantly impair ANS control. However, it has not been determined whether these two conditions produce comparable effects on autonomic activity.

Autonomic control of circulation is typically investigated by cardiovascular variability (CVV) analysis, which is a convenient, non-invasive tool to probe the ANS under physiological conditions, stress and disease. While heart rate variability and blood pressure variability assessed in the frequency domain convey valuable pathophysiological information on the "sympathovagal balance", they are somehow limited to the detection of linear oscillations in CV signals. Thus, nonlinear metrics are often computed in addition to standard CVV spectral indices and baroreflex sensitivity (BRS) to provide a broader picture of autonomic balance and cardiovascular control [3-5].

The hypothesis underlying the comparison of SAD and sepsis is that the SAD-induced ANS impairment could be similar to the one observed in intact animals following sepsis and ensuing immune system activation and severe system wide inflammation. Hence, considering the still limited understanding of ANS dysregulation associated with sepsis, such a comparison could help attain greater insight into sepsis-induced circulatory abnormalities.

This study aims to 1) investigate whether the autonomic alterations observed in sepsis resemble those induced by SAD in a rat model of experimental SAD and sepsis, and 2) evaluate the potential clinical usefulness of combining linear and nonlinear variability metrics for prognosis, patient monitoring and therapeutic support in critical illness.

2. Methods

2.1. Experimental protocol

This study was approved by the ethics committee of the University of São Paulo Medical School (FMUSP, CEUA/004/14). Male Wistar rats (~300g) were housed in a temperature-controlled environment (22–24 °C) under a 12 h light/dark cycle, with ad libitum access to food and

water. On day 1, all animals underwent femoral artery and vein cannulation for arterial blood pressure (ABP) recordings by connection of the arterial line to a pressure transducer, and supplementation of fluids via the venous line. Animals were then divided into two groups: SHAM (n=6), subjected to cervical manipulation only, and animals that underwent sinoaortic denervation (SAD, n=3) by Krieger's method.

On day 4, baseline ABP measurements were obtained in all animals. Subsequently, half of the animals in the SHAM group were submitted to cecal ligation and puncture (CLP) to induce sepsis, forming three experimental subgroups (n=3 each): SHAM, Sepsis, and SAD. Additional recordings were performed at 2 hours post-CLP and at the final endpoint (defined by onset of septic shock, MAP < 65 mmHg), followed by euthanasia. Invasive ABP waveforms were collected throughout the experiment at a sampling rate of 2kHz [6].

2.2. Datasets and preprocessing

Beat-by-beat series of systolic (SAP), diastolic (DAP), mean (MAP) arterial pressure, pulse pressure (PP), and heart period (HP) were derived by preprocessing of raw ABP signals and resampled in the time domain at 5 Hz, with post adaptive filtering to attenuate or remove artefacts and noisy components, and then normalised. For each rat, three 5-minute-long segments collected at different time frames were analyzed to observe changes on day 4 in comparison to baseline, at two hours after CLP and post-intervention, before euthanasia.

2.3. Spectral analysis and baroreflex sensitivity (BRS)

Spectral analysis was conducted using autoregressive modelling to estimate power within classical frequency bands, adapted to the rats, i.e., very low frequency (VLF, f < 0.25 Hz), low frequency (LF, $0.25 \leq f \leq 0.75$ Hz), and high frequency (HF, f > 0.75 Hz), which are known to be associated respectively with vascular tone, sympathetic, and parasympathetic modulation [5]. BRS was assessed using a closed-loop bivariate autoregressive model to capture the bidirectional dynamics between blood pressure and heart period fluctuations, offering a comprehensive measure of autonomic control [7].

2.4. Entropy indices

Sample entropy (SampEn) and approximate entropy (ApEn) were computed to assess the irregularity and complexity of each time series. These nonlinear metrics quantify signal unpredictability and have been increasingly applied in the analysis of physiological dynamics under both healthy and pathological conditions [3,8].

2.5. Statistical analysis

Group comparisons were performed using two-way ANOVA followed by Tukey's post-hoc test, considering experimental group and time as factors. A p-value < 0.05 was considered statistically significant.

3. Results

SAD animals presented consistently higher VLF and HF spectral bands of SAP and DAP compared to non-denervated groups.

The LF component of BRS in the SAD group exhibited high intragroup variability, while sepsis showed a suppression of the response, as shown below (figure 1).

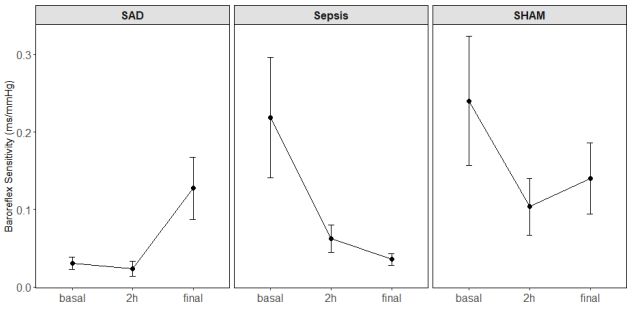

Figure 1. Baroreflex sensitivity (BRS, in ms/mmHg)) across experimental phases (Baseline, 2h, and Final) for each group.

The sepsis group exhibited a progressive decline in BRS, suggesting impaired baroreflex responsiveness. In contrast, the SHAM group showed a gradual increase in BRS over time. The SAD group displayed greater variability and less consistent trends, reflecting an irregular adaptation to experimental conditions.

Both sample and approximate entropies pointed to greater unpredictability of DAP and HP in SHAM and Sepsis, while SAD animals exhibited greater complexity in SAP, with opposite trends compared to the other groups). The results of frequency domain and entropy analysis of the ABP and HP time series are summarized in Table 1.

4. Discussion

Combining cardiovascular variability metrics such as spectral indices and baroreflex sensitivity (BRS) with nonlinear metrics like approximate and sample entropy provides a robust framework to assess autonomic nervous system (ANS) responses to physiological or pathological

Table 1. Mean and standard deviation of entropy indices (ApEn and SampEn) and spectral indices (VLF, LF, HF and LF/HF ratio) for each normalized physiological time series (systolic arterial pressure (SAP), diastolic arterial pressure (DAP), mean arterial pressure (MAP), and heart period (HP)). * $p<0.05$ after comparisons between SAD and SHAM groups.

Entropy indices						
	ApEn			SampEn		
	SAD	SHAM	Sepsis	SAD	SHAM	Sepsis
SAP	0.59 ± 0.43*	0.10 ± 0.10	0.71 ± 0.52	1.46 ± 0.14	0.96 ± 0.50	1.20 ± 0.38
DAP	0.19 ± 0.10	0.20 ± 0.12	0.25 ± 0.18	0.98 ± 0.23*	1.31 ± 0.33	1.35 ± 0.23
MAP	0.14 ± 0.07	0.21 ± 0.14	0.20 ± 0.16	0.86 ± 0.18*	1.26 ± 0.33	1.26 ± 0.29
HP	0.07 ± 0.04*	0.16 ± 0.11	0.15 ± 0.13	0.86 ± 0.18*	1.26 ± 0.33	1.26 ± 0.29
Spectral indices						
	VLF			LF		
	SAD	SHAM	Sepsis	SAD	SHAM	Sepsis
SAP	8.41 ± 5.79*	3.20 ± 1.06	3.07 ± 0.84	0.13 ± 0.08	0.24 ± 0.11	0.28 ± 0.29
DAP	8.87 ± 3.39*	2.84 ± 1.41	3.31 ± 1.50	0.15 ± 0.07	0.27 ± 0.11	0.39 ± 0.42
MAP	10.09 ± 4.31*	4.11 ± 2.46	4.06 ± 1.23	0.14 ± 0.08	0.22 ± 0.12	0.35 ± 0.44
HP	1.74 ± 0.80	1.80 ± 0.80	1.36 ± 0.76	0.32 ± 0.07	0.42 ± 0.19	0.37 ± 0.09
	HF			LF/HF ratio		
	SAD	SHAM	Sepsis	SAD	SHAM	Sepsis
SAP	63.06 ± 59.48*	13.90 ± 4.28	13.59 ± 4.25	0.004 ± 0.005	0.02 ± 0.02	0.02 ± 0.02
DAP	78.27 ± 51.93*	12.64 ± 5.99	15.01 ± 6.84	0.003 ± 0.002	0.03 ± 0.03	0.03 ± 0.03
MAP	84.88 ± 57.70*	18.08 ± 10.29	18.05 ± 6.21	0.002 ± 0.002	0.02 ± 0.03	0.02 ± 0.02
HP	10.69 ± 5.99	9.67 ± 4.39	7.43 ± 4.42	0.04 ± 0.03	0.05 ± 0.03	0.08 ± 0.08

stress. Here, we jointly analyzed linear and nonlinear indices in rats subjected to sinoaortic denervation (SAD) and cecal ligation and puncture–induced sepsis to examine autonomic imbalance during acute illness.

BRS was lower at baseline in both SAD and Sepsis groups, confirming impaired baroreflex buffering either by loss of baroreceptor input (SAD) or sepsis-related inflammation. While BRS progressively increased in SAD animals, sepsis (with or without denervation) caused a sustained suppression, suggesting a sepsis-induced inhibition of baroreflex function, possibly through sympathetic overactivity.

Spectral analysis supported these findings. SHAM rats exhibited higher high-frequency (HF) power and lower LF/HF ratios, reflecting predominant parasympathetic modulation. SAD animals displayed sympathetic dominance (higher LF/HF) and reduced HF power, whereas sepsis further decreased HF and increased LF/HF ratios. These patterns align with Tracey's concept of neuro-immune interaction, where sympathetic activation parallels inflammation and vagal activity exerts anti-inflammatory effects.

Entropy-based measures confirmed distinct autonomic profiles. SHAM animals showed high heart-period (HP) and diastolic-pressure entropy but low systolic-pressure entropy, indicating balanced adaptability. SAD and Sepsis groups exhibited lower SampEn of HP and DAP and higher ApEn of SAP, suggesting reduced adaptability and unbuffered systolic instability from baroreflex failure. The overall entropy increase in sepsis may reflect global disruption of autonomic coordination.

The irregular BRS pattern in denervated rats is consistent with afferent signal loss and chaotic baroreflex output, whereas sepsis led to a progressive BRS decline, indicating functional suppression even with intact reflex circuits.

In summary, BRS, spectral power, and entropy captured complementary facets of ANS regulation. SAD increased CV signal complexity, while sepsis inhibited baroreflex responses and disorganized autonomic control. These results support the integration of linear and nonlinear analyses to elucidate ANS dysregulation in sepsis and highlight entropy metrics as potential markers of dysfunction severity. The main study limitation is its small sample size, warranting further validation and exploration of clinical translation.

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