Poincaré Plot in Ischemic Rabbit Hearts

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Abstract

Heart rate variability can be evaluated by Poincaré plot. Its parameter SD1 describes short-term variability which is mainly caused by respiratory sinus arrhythmia, whereas SD2 describes long-term variability. Ischemia is believed to affect non-autonomic mechanism of the heart and as a result of it also HRV. However not only this mechanism is responsible for HRV changes, because ischemia affects both parameters significantly even in isolated denervated hearts. Both parameters - SD1 and SD2 - show lower values in ischemic phases of *experiment than in reperfusion ones. Shape of Poincaré* plot*'s attractor has a form of line* both in ischemic and reperfusion periods; however in reperfusion phases of experiment the attractor is longer.

1. Introduction

1.1. Heart rate variability

Heart rate variability (HRV) is a non-invasive electrocardiographic diagnostic tool, based on analysis of instantaneous RR intervals. It expresses activity of autonomous nervous system which affects rhythm of the heart. The hearts automaticity is intrinsic property of cardiac tissue. Heart rhythm is controlled by sinoatrial node which serves as a pacemaker. This pacemaker is modulated by innervation from both sympathetic and parasympathetic branches of the autonomic nervous system [1]. Sympathetic system under physiological condition increases heart rate, whereas parasympathetic system inhibits the pacemaker and thus slows down the heart. Under physiological conditions the heart beats in sinus rhythm reflecting balanced sympathetic and vagal state. However in diseased heart this balance is disrupted and HRV shows a smaller variation between RR intervals. Moreover, several other mechanisms may influence HRV: (a) sympathetic system can modulate HRV indirectly through release of adrenomedullary catecholamines [1], (b) humoral factors including variation of the renin-angiotensin system [1] (c) stretch-induced mechanical effect [1], and (d) other intrinsic non-autonomic mechanisms [2].

Measures of heart rate variability may represent a promising diagnostic tool for identification and clarification of significant relationship between psychological and physiological processes [1]. At present, this measurement is commonly used as quantitative marker of autonomic activity which is associated with cardiovascular mortality, including sudden cardiac death [3, 4].

1.2. Isolated heart

Although numerous HRV studies have been published, only a few of them deal with isolated hearts. Mechanisms leading to presence of HRV despite cardiac denervation remain to be elucidated [4]. Study of HRV in isolated hearts may considerably contribute to understanding of its origin and may improve diagnostic methods based on HRV analysis.

The majority of data describing HRV modulated by non-autonomic mechanisms originates from cardiac transplanted patients. Cardiac transplant recipients can serve as a clinical model of the denervated heart. Immediately after transplantation patients have no HRV [1] or much reduced HRV [4], except small mechanically mediated RSA [1].

1.3. Poincaré plot

HRV can be assessed by statistical, spectral, or geometrical method. Although all of them are recommended for clinical use [3], both statistical and spectral method cannot reveal non-harmonic component of HRV. It has been proven that HRV includes non-linear and non-harmonic component. In biological terms, it is possible to hypothesise that non-linear dynamics of heart rate variability might contain some useful marker of neural control and of the mechanical characteristics of the cardiovascular system [5]. Non-linear component can be

assessed by Poincaré plot.

Poincaré plot is a graphical representation of dynamics of HRV. It has a form of diagram, where consecutive R-R interval of a tachogram (or HRV time series) is drawn as a function of the previous R-R interval, or in some instances as a function of n-th preceding R-R interval. The quantitative analysis of the Poincaré plot is based on the fact that each R-R interval is influenced by previous vagal and sympathetic modulations over the heart rate and therefore the pairs of successive R-R intervals form an attractor in the Poincaré plot [6].

This attractor allows visual classification of the HRV signal. However, the assessment and standardization of these qualitative classifications are difficult because they are highly subjective. A quantitative analysis of the HRV attractor displayed by the Poincare´ plot can be made by adjusting it to an ellipse. Using this technique, three indexes [6] can be obtained: SD1, SD2 and theirs ratio.

2. Method

All experiments followed the guidelines for animal treatment approved by local authorities and conformed to the EU law. Seven New Zealand rabbits were included in the study. Their isolated hearts were perfused according to Langendorff in the mode of constant perfusion pressure (85mmHg) [7].

In deep anaesthesia with xylasin and ketamin, the hearts were excised and fixed on perfusion apparatus filled with Krebs-Henseleit (K-H) solution (1.25mMCa2+, 37°C) and placed in a bath, where the hearts were stabilized for 30 minutes.

Each heart underwent three 15-minutes long episodes of coronary artery occlusion, interrupted with three reperfusion periods of the same duration (see scheme of experiment in Fig. 1). Global ischemia was achieved by complete restraint of perfusion (s.-c. flow ischemia). The ECG signal was measured by touch-less method [7, 8]. Briefly, three Ag-AgCl disc electrodes in three orthogonal directions x, y, and z are placed in the walls of the bath which is a part of the perfusion system. Each isolated rabbit heart used in this study was positioned in the same way in the bath.

ECG signals were recorded by data acquisition multifunction card PCI-6111E (National Instruments, USA) with sampling frequency fs=2000Hz. ECG signals were acquired by own application designed in LabView 7.1 software (Texas Instrument, 2008). The 12-bit analogue to digital conversion was used. The digital signal was stored on a hard disk for off-line processing.

Three ECG signals with duration approximately two hours were recorded. Afterwards, seven 5-minutes long parts were extracted in Matlab R2006a (MathWorks, 2006) from particular phases of experiment.

R peaks were detected automatically by own R-wave detector designed in Matlab R2006a (MathWorks, 2006). The results of automatic analysis were reviewed and any errors in detection were corrected manually by human revision.

HRV parameters were computed from RR series interpolated with cubic spline method and resampled at fs=30Hz. Slow trends were removed by detrending procedure based on smoothness priors regularization with regularization parameter λ=3000.

Signals were further analysed by Kubios HRV software [9] and parameters SD1 and SD2 were evaluated.

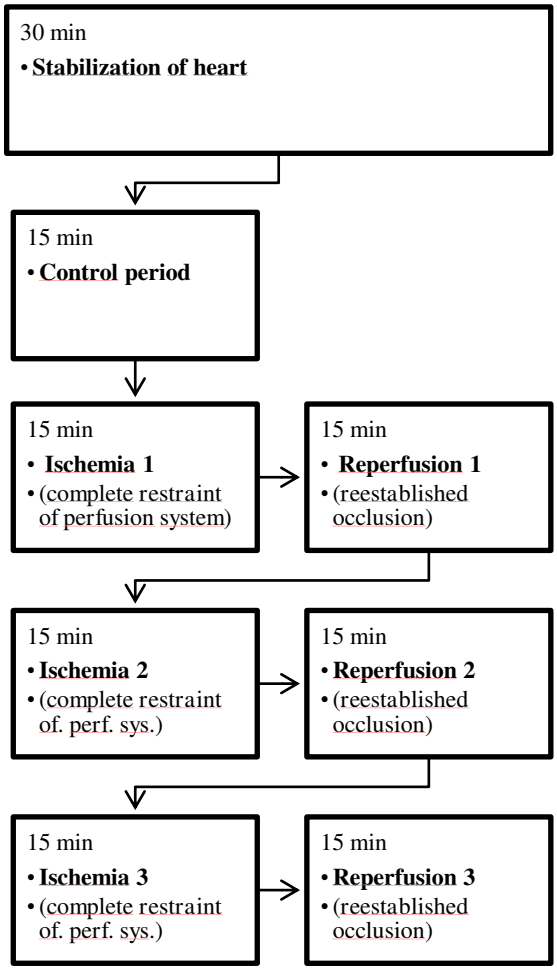

Fig 1. Experimental scheme

3. Results

Poincaré plots have been constructed for each phase of experiment (control, 3x ischemia, and 3x reperfusion). The data of seven isolated rabbit hearts have been used for statistical analysis. Two standardized [3] parameters SD1 and SD2 have been evaluated which represent short-time (SD1) and long-time (SD2) HRV variability. In ischemic phases SD1 was 2.21 - 5.21times higher than in reperfusion phases (Fig. 2). Similarly, SD2 in ischemic phases was 1.48 – 2.34times higher than in reperfusion phases (Fig. 3). Attractor of the Poincaré plot has the same shape for ischemic and reperfusion periods (Fig. 4), however for reperfusion periods has a longer SD1 and SD2. Values of SD1 and SD2 for each phase of experiment are shown in Table 1 and 2.

Table 1: SD1 in isolated rabbit hearts (n=7)

	phase of experiment						
	Control (ms)	Isch. 1 (ms)	Rep. 1 (ms)	Isch. 2 (ms)	Rep. 2 (ms)	Isch. 3 (ms)	Rep. 3 (ms)
1.	2.0	17.6	263.4	15.5	105.4	0.9	198.1
2.	2.4	6.2	36.2	4.5	11.6	0.9	10.0
3.	1.5	4.2	4.8	3.8	13.2	15.2	14.2
4.	15.1	8.0	30.9	7.9	2.7	10.9	63.7
5.	3.9	2.7	6.4	3.4	5.1	4.6	6.8
6.	0.4	13.2	24.8	5.2	18.9	6.0	23.9
7.	16.5	33.0	24.9	25.9	1.7	1.3	36.3

Table 2: SD2 in isolated rabbit hearts (n=7)

	phase of experiment						
	Control (ms)	Isch. 1 (ms)	Rep. 1 (ms)	Isch. 2 (ms)	Rep. 2 (ms)	Isch. 3 (ms)	Rep. 3 (ms)
1.	23.7	136.6	685.6	141.9	432.6	59.1	855.2
2.	9	34.9	89	80.6	314.1	59.2	273.8
3.	2.3	6.3	14.7	12.1	27.9	35.1	22.4
4.	10.7	106	121.8	54.7	60.6	34.6	103
5.	2.8	11.3	48.4	12	23.9	8.7	22.6
6.	8.2	82.2	165.1	45.6	143.9	44.8	102.6
7.	15.3	91.7	256.8	213.4	146.8	78.6	74.6

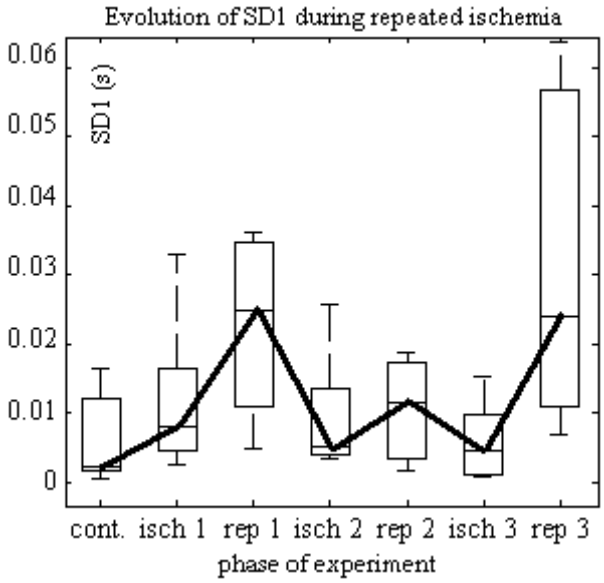

Figure 2: Evolution of SD1 during three repeated ischemic periods (15 min each), interrupted with three reperfusion periods (15 min each). Abbreviations in x-axis label: cont. – control period, isch. – ischemia, rep. – reperfusion.

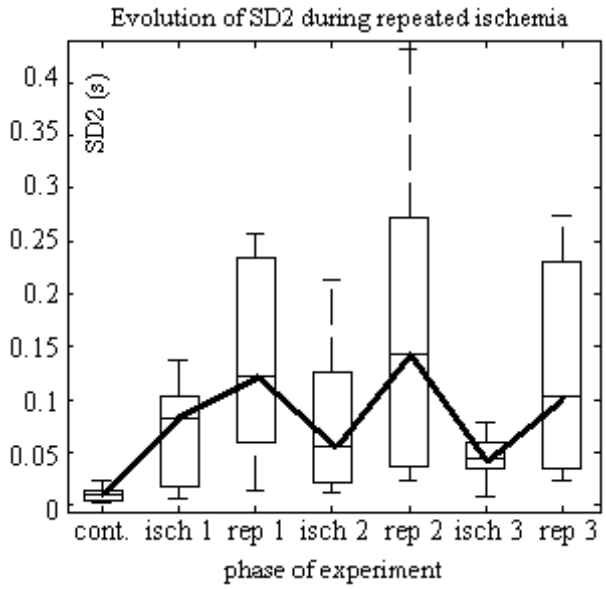

Figure 3: Evolution of SD2 during three repeated ischemic periods (15 min each), interrupted with three reperfusion periods (15 min each). Abbreviations are the same as in Figure 2.

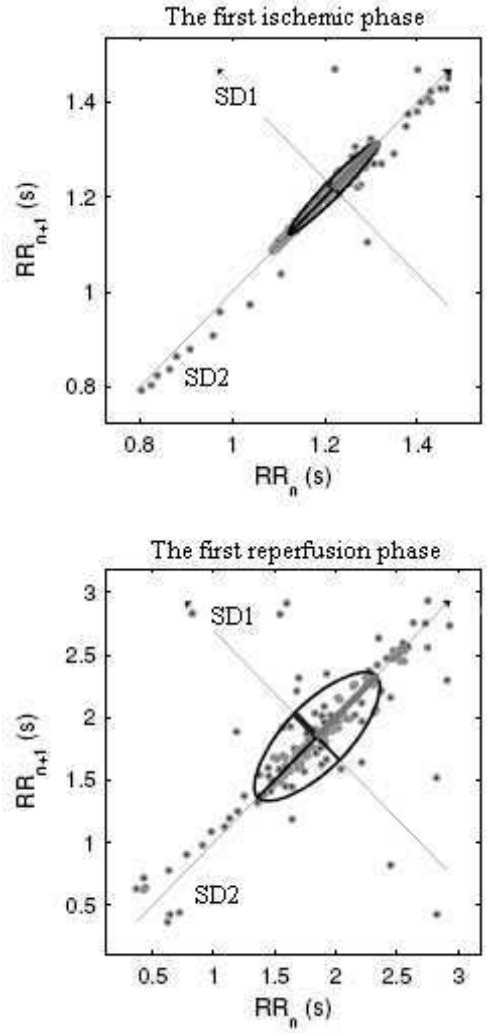

Fig. 4. Shape of attractor in ischemic and reperfusion phase of experiment. (Performed with usage of Kubios HRV software [9]).

4. Conclusions

Poincaré plots of ischemic phases are more compact and smaller than the reperfusion ones. These findings are in accordance with majority of studies dealing with working, non-isolated hears. Global ischemia has probably the same effect in both isolated and normal working hearts; however confirmation of this hypothesis is limited by difficulties associated with performing of ischemia in living animals. Further studies are needed for clarification of ischemia effect on the heart.

Acknowledgements

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