Effect of β-Adrenoblockers on Heart Rate Variability in Awake and Narcotized Rats

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In narcotized rats, β-adrenoceptor blockers changed all parameters of heart rate variability indicating up-regulation of parasympathetic activity and down-regulation of the sympathetic one. In immobilized awake rats, the temporal and geometrical parameters varied similarly, while spectral analysis indicated possible activation of other neurohumoral mechanisms against the background of β-blocker treatment.

Key Words: *heart rate variability; autonomic nervous system; β-adrenoceptor blockers; short-term immobilization stress*

β-Adrenoceptor blockers belong to few pharmacological groups of therapeutic drugs that prolong lifetime of patients with such socially important diseases as arterial hypertension, chronic cardiac insufficiency, all the forms of CHD, and cardiac rhythm disorders [5]. Wide use of β-adrenoblockers is explained by their pronounced modulatiin effect on the sympathoadrenal system, whose hyperactivation is a pathogenetic link in the development of cardiovascular pathology. However, from the pharmacological point of view, β-adrenoblockers are a heterogeneous group of preparations. Depending on affinity to β_1- and β_2-adrenoreceptors, β-adrenoblockers are subdivided into cardioselective drugs predominantly blocking β_1-adrenoreceptors and non-cardioselective drugs that block both β_1- and β_2-adrenoreceptors. It is noteworthy that the heart is controlled by not only β_1-adrenoreceptors, but also by β_2-adrenoreceptors. This regularity is most evident in chronic cardiac insufficiency characterized by desensitization of β_1-adrenoreceptors and increased density of β_2-adrenoreceptors (this peculiar adaptive reaction rapidly loses its adaptive potency) [3,8]. Therefore it remains unclear which type of β-adrenoceptor blockade (complete or partial β_1) is preferable for the therapy of particular cardiac disease [6,7]. Moreover, it is not clear whether activity of β-adrenoblockers is similar in various functional states of the organism. Probably, these challenges could be met with the analysis of heart rate variability (HRV), which belongs to the most informative and sensitive methods for evaluating of cardiac activity regulation.

Our aim was to reveal peculiarities in cardiotropic effects of cardioselective β_1-blocker atenolol and nonselective β-blocker propranolol in narcotized and awake rats subjected to short-term immobilization stress.

MATERIALS AND METHODS

Two experimental series were carried out on mature random-bred albino male rats weighing 180-210 g. In series I (n=20), ECG was recorded in narcotized rats (nembutal, 40 mg/kg intraperitoneally) immobilized in the supine position. Than, group 1 rats (n=10) received 10 mg/kg propranolol (anaprilin, Medisorb) and group 2 rats (n=10) received 5 mg/kg atenolol (Pliva) in a volume of 1 ml through a gastric tube. To calculate the doses, the scale factor

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was taken for 5.9 [4]. After 1 hour, ECG was recorded again

In series II, the experiments were carried out on awake rats randomized into 3 groups. Group 1 rats received 1 ml physiological saline through a gastric tube (controls, n=20). Similarly, group 2 (n=10) and group 3 (n=10) rats received 10 mg/kg propranolol or 5 mg/kg atenolol in a volume of 1 ml, respectively. After 1 hour, the rats were immobilized in the supine position and ECG was recorded.

ECG was recorded using a dual-channel electrophysiological setup connected through an L-CARD E-440 digitizer to a Pentium II PC. ECG (standard lead II) was sampled at a rate of 4.0 kHz over 4 min using L-GRAPH software supplied with a digitizer. ECG preview and its primary processing were made with RRMatch software, thereafter final calculation of HRV parameters and their plotting were made within CRGraph software. The above software packages were developed in Department of Digital Signal Processing and Imaging (Yaroslavl State University) to analyze HRV in the laboratory animals. The power spectra of cardiac intervalograms were calculated with an algorithm based on fast Fourier transform and Hann window [13]. To improve statistical stability of the spectra, the calculations were performed by averaging power spectra of the overlapping epochs [13].

For the analysis of HRV we used time-domain, frequency-domain, and geometrical (Poincare) scatterplot indices: HR, standard deviation of normal-to-normal R-R intervals (SDNN) [1,2]), variation coefficient, root mean square of successive differences (RMSSD) of R-R intervals; variational range, mode, mode amplitude, strain index, scatterplot area, scatterplot width-to-length ratio, low frequency (LF) wave power, high frequency wave (HF) power, total spectrum power, relative LF wave power, relative HF power, and the autonomic balance index. In calculations of geometric scatterplot indices, the histogram step was 2 msec. There is no consensus on the frequency ranges of slow and fast waves revealed by spectral analysis [11,12,14,15]. To avoid typical errors [9], we chose these ranges with due account of all obtained spectrum data. Thus, in narcotized rats, the LF and HF ranges were 0.02-0.15 Hz and 0.15-2.00, respectively. Similarly, in awake rats the corresponding values were 0.02-0.75 and 0.75-3.00 Hz.

The data were processed with Statistica 6.0 software, which calculated the mean value, square deviation, standard error of the mean, and Student's t-test at p<0.05.

RESULTS

In narcotized rats, injection of β-adrenoblockers significantly changed most HRV indices (Table 1). Propranolol and atenolol decreased HR by 1.5 and 1.2 times, respectively. Both drugs significantly increased standard deviations of normal R-R intervals (by 70.5 and 42.1% in propranolol and atenolol groups, respectively), the square root of the sum of squared differences of the successive pairs of R-R intervals (by 2.3 and 1.4 times, respectively), variational range (by 66.4 and 39.1%), mode (by 49.9 and 26.0%), and the scatteroplot area (by 3.3 and 1.9 times). By contrast, these drugs significantly decreased the mode amplitude (by 41.5 and 32.5% in propranolol and atenolol groups, respectively) and the strain index (by 4.1 and 2.8 times). In narcotized rats, β-adrenoblockers did not modify LF power, but increased HF power by 4.1 times (propranolol, p<0.01) and by 1.4 times (atenolol). In these rats, the total spectrum power increased by 2.3 and 1.2 times, respectively. In addition, propranolol and atenolol decreased the share of LF power in total spectrum by 2.9 and 1.7 times, respectively. In contrast, the share of HF power increased by 1.5 and 1.3 times, respectively. Propranolol and atenolol decreased LF/HF ratio by 4.7 and 2.5 times, respectively (p<0.05).

Thus, propranolol and atenolol increased HRV and parasympathetic tone. In addition, they moderated the sympathetic influences on the hearts of narcotized rats. These phenomena agree with published data on the effect of β-adrenoblockers on HRV in behaviorally unrestrained rats [10-12,15]. It is noteworthy, that some indices (square root of the sum of squared differences of the successive pairs of R-R intervals, LF power, and total spectrum power) revealed greater activity of propranolol in comparison with atenolol. Taking into consideration the value of statistical error in the analysis of other indices, one can conclude that in narcotized rats propranolol produced a greater effect on HRV than atenolol.

In awake animals subjected to short-term immobilization stress, propranolol and atenolol decreased HR by 1.2 and 1.3 times, respectively, in comparison with controls receiving physiological solution (Table 2). Other indices obtained with time-domain and geometrical analyses were changed by both drugs similarly as in the group of narcotized rats. The drugs significantly increased standard deviations of normal R-R intervals (by 88.7 and 93.2% in propranolol and atenolol groups, respectively), variation coefficient (by 59.1 and 53.7%, respectively), the square root of the sum of squared dif-

TABLE 1. Effect of β-Adrenoblockers on HRV Indices in Narcotized Rats (*M*±*m*)

Index	Initial state	One hour postinjection	
		propranolol	atenolol
HR, min^{-1}	409.0±13.2	276.0±17.5***	330.0±20.2**
SDNN, msec	1.900±0.246	3.240±0.393**	2.700±0.214*
VC, %	1.260±0.135	1.470±0.161	1.470±0.141
RMSSD, msec	1.670±0.157	3.79±0.44***	2.320±0.273*+
MxDMn, msec	11.00±1.29	18.30±2.65*	15.30±1.07*
Mo, msec	148.60±5.01	222.70±16.42***	187.30±11.28**
AMo, %	44.40±3.03	26.00±2.76**	30.00±2.76**
SI, rel. units	16 334±2339	3942±1025**	5801±938**
EllSq, $msec^2$	85.10±19.46	279.20±53.86***	162.90±24.86*
EllAs, %	57.50±5.63	72.00±7.89	49.80±7.33
LF power, $msec^2$	0.23±0.09	0.230±0.063	0.170±0.047
HF power, $msec^2$	0.380±0.132	1.550±0.356**	0.530±0.093++
TP, $msec^2$	0.610±0.191	1.780±0.392**	0.700±0.126+
Relative LF power, %	41.60±4.92	14.60±4.52**	24.50±4.02*
Relative HF power, %	58.40±4.92	85.40±4.52**	75.50±4.02*
Autonomic balance index LF/HF	0.890±0.174	0.190±0.073*	0.350±0.072*

Note. Here and in Tables 2: SDNN, standard deviation of normal *R-R* intervals; CV, variation coefficient; RMSSD, root mean square of successive differences of R-R intervals; MxDMn, variational range; Mo, mode; AMo, mode amplitude; SI, strain index; EllSq, scatterplot area; EllAs, scatterplot width-to-length ratio; TP, total spectrum power. *p<0.05, **p<0.01, ***p<0.001 relative to the initial state; +p<0.05, ++p<0.01 between propranolol and atenolol.

TABLE 2. Effect of β-Blockers on HRV Indices in Awake Immobilized Rats (*M*±*m*)

Index	Control	Propranolol	Atenolol
HR, min^{-1}	509.0±4.1	429.0±10.1***	404.0±9.6***
SDNN, msec	1.770±0.115	3.340±0.581***	3.420±0.694***
VC, %	1.490±0.093	2.370±0.402**	2.290±0.456*
RMSSD, msec	1.540±0.083	2.210±0.256**	2.090±0.229**
MxDMn, msec	10.20±0.63	19.60±3.64***	17.80±2.67***
Mo, msec	118.30±0.94	139.80±3.42***	149.20±3.55***
AMo, %	44.90±1.82	29.20±2.86***	29.40±3.79***
SI, rel. units	22 025±2088	7611±1784***	7415±1659***
EllSq, $msec^2$	71.50±8.05	233.00±74.59***	228.20±78.21**
EllAs, %	52.90±3.45	38.20±2.11*	38.1±3.9*
LF power, $msec^2$	0.33±0.06	1.420±0.425***	1.330±0.472**
HF power, $msec^2$	0.290±0.034	0.53±0.08**	0.500±0.084**
TP, $msec^2$	0.620±0.078	1.950±0.501***	1.83±0.53***
Relative LF power, %	46.1±3.7	65.20±4.48**	64.70±4.42*
Relative HF power, %	53.9±3.7	34.80±4.48**	35.30±4.42*
Autonomic balance index LF/HF	1.180±0.204	2.270±0.415*	2.370±0.585*

Note. *p<0.05, **p<0.01, ***p<0.001 compared to the control.

ferences of the successive pairs of R-R intervals (by 43.5 and 35.7%, respectively), variational range (by 1.9 and 1.7 times), mode (by 18.2 and 26.1%), and the scatteroplot area (by 3.3 and 3.2 times). By contrast, these drugs significantly decreased the mode amplitude (by 53.8 and 52.7% in propranolol

and atenolol groups, respectively), the strain index (by 2.9 and 3.0 times), and the scatteroplot width-to-length ratio (by 38.5 and 38.8%, respectively).

Propranolol and atenolol produced virtually equal effects on spectral parameters in stressed awake rats: they increased LF, HF, and total spectral powers by 4.0-4.3, 1.8-1.7, and 3.0-3.1 times, respectively. Adrenoceptor blockers increased the contribution of LF power by 1.4 times and decreased the contribution of HF power by 1.5 times. As a result, LF/HF index increased 2-fold compared to the control.

Thus, in narcotized and awake rats, propranolol and atenolol produced similar changes: increase the sympathetic tone and decrease the parasympathetic tone against the background of elevated total HRV. Spectral analysis revealed certain peculiarities in the action of β-adrenoblockers: in narcotized rats, changes in LF and HF powers corroborated the data of time-domain and geometrical analysis, although in awake rats the spectrum, time-domain, and geometrical data seem to be contradictory. Indeed, in addition to the increase of HF power, adrenoblockers pronouncedly increased LF component in awake rats. Evidently, blockade of β-adrenoceptors in stressed rats activates some other mechanisms (probably, serotoninergic, histaminergic, and cholinergic), which elevate LF component in HRV power spectrum [1]. The decrease in strain index and increase of the total power of HRV spectrum by β-adrenoblockers attest to elevation of the adaptive potencies in both narcotized and in stressed awake rats [1].

The present data showed that propranolol produced more pronounced effect on HRV than atenolol, especially in narcotized rats. Probably, this peculiarity relates to the ability of propranolol to block both β_1- and β_2-adrenoceptors, which attests to the important role of the latter in heart rhythm regulation.

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