# Heart Rate Variability in Chinchilla Rabbits

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Temporal and spectral parameters of heart rate variability were studied as criteria for classification of mature Chinchilla rabbit population according to their basal neurovegetative status. The absolute values of total spectral power density and individual frequency bands differed significantly in vagotonics and sympathotonics. However, standardized spectral power distributions in high-, low- and very low-frequency ranges were similar in both groups. Our results suggest that the intensity of regulatory influences on the heart at all levels of regulation, which can be evaluated by spectral analysis, is similarly distributed in both groups. Hence, the observed differences in the heart rate variability can not be explained by peculiarities in sympathovagal balance, but are determined by more universal mechanisms.

**Key Words:** *heart rate variability*; *rabbit*; *sympathovagal balance*

Rabbits are often used as experimental animals, but their basal neurovegetative status is usually ignored. At the same time, emotional and motivation strain affects responsiveness and adaptation reserves in both humans and animals. About twenty articles published in the last decade analyze various aspects of cardiac vagal and sympathetic control in rabbits, in particular under conditions of passive or active wakefulness, using spectral analysis of heart rate variability (HRV) [5,6,8-10]. Although temporal structure of HRV changes was thoroughly studied [1], no evidence on individual dispersion of HRV in rabbits were reported.

We earlier showed that population of Chinchilla rabbits is very heterogeneous. It consists of two subpopulations, which significantly differ in HRV parameters, responsiveness to orthostatic stress, and sensitivity to radiostimulation [2-4,7]. To study this problem in detail, a large body of experimental data accumulated in our Research Center for the last 5 years was analyzed repeatedly with modern powerful software for data processing and multivariate statistics. Our aim was to study HRV time-domain (TP) and spectral parameters (SP) as criteria for rabbit classification by their neurovegetative status.

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## MATERIALS AND METHODS

Experiments were carried out on 118 Chinchilla rabbits (2.5-3.0 kg) in autumn and winter. The rabbits were bred and kept in a vivarium. After rabbit pups became 4 month old, they were placed into individual cages, in which they were kept under standard conditions with free access to food and water throughout the observation period.

In order to get stable ECG records in awake rabbits, they were habituated to the recording chamber (plexiglas box with large vent holes). ECG signals were recorded using a Siemens-Elema Mingograph recorder. Precordial wire electrodes were attached to the skin with miniature clips. These electrodes did not restrict free moving and posturing.

Signal preprocessing and automatic calculation of *R—R* intervals (with 0.1 msec accuracy) was performed using original GURI-95/GURI-97 hardware/software analyzer. The number of *R—R* intervals, record time, number of duplicates in the series, and other

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**TABLE 1.** Distribution of Spectral Parameters of Heart Rate Variability (msec$^2$) in Vagotonics and Sympathotonics ($M\pm m$)

| Parameter | Vagotonics ($n$=64) | | Sympathotonics ($n$=47) | |
|---|---|---|---|---|
| | abs. | % | abs. | % |
| TSP | 104.8±15.6 | 100 | 33.1±5.4* | 100 |
| VLF | 42.6±8.7 | 40.6±4.1 | 10.1±2.2* | 36.5±2.9 |
| LF | 17.9±2.2 | 17.1±1.8 | 6.2±1.2* | 18.0±1.9 |
| HF | 10.9±1.2 | 10.6±1.1 | 4.3±1.0* | 12.1±1.4 |
| LF/HF | 1.64±0.29 | — | 1.48±0.32 | — |

**Note.** *$p$<0.001 compared to vagotonics (Student's $t$ test).

parameters were set in the program and remained unchanged throughout experiment. The data and experimental requisites were saved as standard files. Detailed mathematical analysis of primary data files was performed using standard and original software products designed in MatLab and Statistica operational environments. SP of HRV were evaluated by conventional (Fourier transform and autoregression method) and alternative techniques such as wavelet analysis.

Series containing 500 *R—R* intervals were used for spectral analysis. The area under the power spectral density graph was integrated for the high-frequency power (HF) between 1.7-0.4 Hz, the low-frequency power (LF) between 0.4-0.15 Hz, and the very low-frequency power (VLF) between 0.15-0.04 Hz. The total spectrum power (TSP) was calculated for the frequency range including frequencies below 0.04 Hz and above 1.7 Hz. The power was expressed both in absolute and normalized units (in %), obtained by dividing the power of each component by TSP and multiplying it by 100.

In addition to SP, TP were evaluated including the mean heart rate, standard deviation of the mean *R—R* interval ($SD_{R—R}$); mode of *R—R* interval distribution (Mo) and its amplitude (AMo), the width of *RR* interval dispersion (DX) and strain index by R. M. Baevskii (SI), which was normalized and adapted to rabbits.

Preliminary assessment of heterogeneity of a given group of rabbits and their classification by test parameters was performed using cluster analysis (agglomerative-hierarchical clustering method). The correlation between test parameters and specific group of rabbits was analyzed by multivariate statistical methods (MANOVA). The significance of intergroup variations was evaluated by Student's $t$ test, Fisher's $F$ test, and Wilk's λ test.

## RESULTS

Cluster analysis of the four spectral components revealed 3 groups of animals with high, moderate, and low TSP, respectively. Sixty-four rabbits (~54%) were vagotonics, 47 rabbits (~40%) were sympathotonics, and only 7 rabbits (~8%) were normotonics. Because of very little number of normotonic animals, they were excluded from the analysis.

Functional strain of autonomic mechanisms of heart rate regulation and, therefore, neurovegetative status differed significantly in vagotonics and sympathotonics (Table 1). Multivariate analysis (Wilk's λ test) showed that these differences were highly significant (λ=0.325; $p_\lambda$<<0.001). At the same time, LF/HF ratios were similar in both groups. Moreover, the normalized values of individual spectral components differed insignificantly in all frequency bands (Table 1). These findings suggest that although functional strain of heart rate regulation mechanisms differed significantly, the structure of interaction between individual regulatory mechanisms was similar in both groups.

Similarly, temporal parameters also differed in these two groups (Table 2). Thus, SI in sympathotonic rabbits 3-fold surpassed the corresponding parameter in vagotonics, which attested to predominance of adrenergic mechanisms in heart rate regulation in sympathotonics.

Thus, Chinchilla rabbits considerably differ by their neurovegetative parameters (judging from analysis of HRV) and cannot be pooled in the same samples or in the same control or experimental groups). Accor-

**TABLE 2.** Distribution of Temporal Parameters of Heart Rate Variability in Vagotonics and Sympathotonics ($M\pm m$)

| Parameter | Vagotonics ($n$=64) | Sympathotonics ($n$=47) |
|---|---|---|
| HR, bpm | 239.2±8.1 | 268.3±10.3* |
| $SD_{R—R}$, msec | 8.4±1.5 | 4.3±0.7* |
| AMo, % | 13.9±2.1 | 23.6±3.8*** |
| DX, msec | 39.3±7.4 | 19.6±3.2** |
| SI | 2.9±1.0 | 8.9±2.3** |

**Note.** *$p$<0.01, **$p$<0.02, ***$p$<0.05 compared to vagotonics (Student's $t$ test).

ding to our data, seasonal or environmental changes (ambient temperature, diet, *etc.*) have practically no effect on neurovegetative organization. Thus, our results indicate that all experimental studies on rabbits (and, probably, other species) are incorrect without preliminary analysis of the neurovegetative status, which reflects emotional and motivation strain and the adaptive-compensatory reserves of each animal. On the other hand, the fact that standardized values of power spectrum distribution between HF, LF, and VLF are similar in both groups suggests that the intensities of all chronotropic autonomic influences are distributed in similar proportion, and that the observed differences can not be explained only in terms of sympathovagal balance, but ask for more universal principles of autonomic control. Otherwise, one can conclude that estimation of sympathovagal balance by the linear spectral methods of HRV analysis can not be quite adequate.

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