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Comparative analysis of the status of the nitric oxide system in the left ventricle of heart in rats with experimental hypertension of different origin

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Background: The hypertension development is accompanied by endothelial dysfunction. NO and its synthases (NOS) play a key role in this event. It is assumed, that neuronal isoform (nNOS) fulfills protective role providing vasodilatation, and inducible isoform (iNOS) aggravates endothelial dysfunction via nitrosative stress, whilst endothelial isoform (eNOS) may provide vasodilatation as well as produce an excessive amount of peroxynitrite. We believe that similar changes may occur in myocardium. The aim of study was to assess the status of the nitric oxide system in left ventricle myocardium in rats with experimental hypertension.

Methods: study was carried out in 3 groups of male rats in age of 7-8 month and weight of 220-290 grams. 1) 10 Wistar rats (mean blood pressure (mBP) 83.8±0.96 mm Hg); 2) 10 SHR (mBP 125.8±1.12 mm Hg); 3) 10 Wistar rats underwent the endocrine-saline modelling (ESM) of hypertension, as described earlier (mBP 137.8±1.23 mm Hg). Study followed the "Principles of laboratory animal care" and approved by local Commission on Bioethics.

The NOS isoforms expression was evaluated with immunofluorescent assay (IF) in paraffin-embedded myocardium slices. In left ventricle myocardium homogenates, we also assessed the mRNA expression to NOS isoforms using real-time polymerase chain reaction (RT-PCR) and the nitrites concentration (Ns) as the end reaction products of the NOS activity using biochemical assay. Moreover, we evaluated the plasma nitrotyrosine (NT) levels using ELISA with aim to assess its potential also as a biomarker.

Statistical analysis was performed using one-way ANOVA with post hoc Bonferroni correction or Kruskal-Wallis criterion with post hoc Dunn correction, when appropriate. Statistically significant differences were considered when p<0.05

Results are showed at the figure 1. Data presented as M±m (A,B), or as median, 1st and 3rd quartiles, min and max (C,D), or correlation dot plots (E,F).

The multifactorial linear regression model (aR²=0.559, p<0.001) indicated Ns (B=6.94±0.92, ß=0.72; ?<0.001) and mBP (B=0.417±0.057, ß=0.696; ?=0.001) as independent predictors of the NT plasma level in rats.

Conclusions: 1) The blood pressure elevation in rats is accompanied by increased expression both of nNOS and iNOS, whereas the eNOS level remained unchanged. 2) The changes stated above are accompanied by increased expression of mRNA of all NOS isoforms, including eNOS, which may be an evidence of the eNOS high demand in myocardium. 3) Considering the progressive decrease of the nitrates concentration in myocardium homogenates and the simultaneous plasma nitrotyrosine level increase, which are correspond to the mean blood pressure elevation, all the stated above may be considered as development of changes in myocardium, which are identical to endothelial dysfunction, including eNOS uncoupling with the shift of its activity to peroxynitrite production despite of NO.
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