

Recent researches have widened the functions of nitric oxide from potentially toxic free radical to universal regulatory factor of gene expression and physiological systems. It was shown that NO synthesized by commensal intestinal bacteria can diffuse into *Caenorhabditis elegans* tissues and enhance lifespan and stress resistance of the worm. The discovered lifelong value of commensal bacteria to their host expanded to human microbiota may improve the credibility of the probiotic concept. Here we aim to characterize NO production by lactobacilli from probiotic preparations and dairy products. Nitric oxide was detected using electronic paramagnetic resonance (EPR) and staining with NO-sensitive dyes (DAF-FM DA, 4-amino-5-methylamino-2',7'-difluorofluorescein diacetate; DAA, 1,2-diaminoanthraquinone sulfate) followed by fluorescent microscopy. We revealed NO production in *Lactobacillus plantarum* 8P-A3 from probiotic preparation "Lactobacterin Dry" (Biomed, Perm, Russia). To elucidate the exact mechanism of NO synthesis by *L. plantarum* 8P-A3, nitric oxide production was examined using L-arginine or nitrate as substrates. We showed that lactobacilli produced NO via L-arginine pathway and not by the generally accepted denitrification. This notion was supported by genomic analysis of bacterial NO-synthases (bNOSs) in *L. plantarum* WCFS1 by bioinformatics methods. We also identified conditions of NO biosynthesis induction in *L. plantarum* 8P-A3 cells. Like in mammalian cells, NO production in *L. plantarum* 8P-A3 was activated by exogenous L-arginine while traditional inhibitors of three mammalian NOS isoforms (L-NAME, L-NIL, nNOS Inhibitor I) had no effect on bacterial NO synthesis. Notably, stress conditions such as heating also resulted in increased levels of NO production.

Biofilms are a widespread form of occurrence of the microorganisms in nature, including intestine, therefore further we studied the role of nitric oxide in biofilm formation by *L. plantarum* 8P-A3. The micromolar concentrations of exogenous NO were shown to have a negative effect on this process due to its toxic effect on the cells. However, the decrease in the level of endogenous NO in bacteria in the presence of a nitric oxide scavenger cPTIO impaired characteristics of the forming biofilms, as was evident from the decrease of their size.

Due to frequent use of lactobacilli as health-promoting probiotics further work is needed to elucidate the exact role between human organism and NO derived from consumed bacteria.

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DYNAMICS OF NITRIC OXIDE IN THE HEART OF THE RAT DURING HYPOKINESIA.

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Study of the effect of a hypokinesia (the restriction of the physical activity) on the body is an actual problem of physiology. Limitation of the physical activity leads to various

changes in the basic life-support systems, and affects the cellular genetic apparatus. Most significantly during hypokinesia (HK) the muscular-skeletal system is affected.

There are many data in the literature on the effect of HA on the metabolism, digestion, respiration, circulation and the function of the endocrine system. Prolonged HK causes various changes in a water-electrolyte metabolism and its regulation mechanisms: changes of a renal concentrating ability, develops a negative balance of potassium and calcium, which depends on the duration of stay in the HK. Disorder of a calcium metabolism in HK is systemic and can affect the contractility of skeletal muscle and myocardium.

Nitric oxide (NO) - gas transmitter, which is a free radical, is now seen as a new signaling molecule that plays the role of a universal regulator of many physiological processes in the body. The study of the content of NO in the tissues of different age rats under a hypokinesia is an urgent task, but there is a question about using the exact method for the quantitative determination of a nitrogen oxide. Aim of this study was to investigate the role of NO in the effects of the hypokinesia, and to identify changes in the content of NO during an ontogenesis.

Studies have been performed on white laboratory rats which were divided into 2 groups: I - control group, which was contained in a standard vivarium conditions with unrestricted motor activity (C); II - the experimental group, which was contained in the conditions of limited motor activity (HK). Investigations were carried out at 4 age groups: 28, 56-, 81-, 110-day-old, these age groups were divided also into 4 experimental groups according to the duration of the HK: 7, 30, 60, 90 daily HK.

NO content in the rat heart was determined by the spin trapping, which has been developed at the Institute of Chemical Physics, Russian Academy of Sciences by Professor A.F. Vanin and his staff. As previously the complex Fe²⁺ + c diethyldithiocarbamate (DETC) was applied. As anesthesia a 25% solution of urethane was used from a calculation of 1200 mg / kg body weight of the animal, which was injected intraperitoneally. The water-insoluble complex of DETC-Fe²⁺ + can react with NO to form a stable radical (DETC) 2-Fe²⁺ + -NO, which can be detected using an electron paramagnetic resonance (EPR).

The smallest amount of NO in the tissues of the heart is formed in 56-days-old rats and significantly increased to 110-day-old is 26.4%. Hypokinesia leads to increased levels of NO in the tissues of rat heart: 30 daily HK by 141.4% 60 41.8% daily, daily 90 to 63.0% compared with control rats. When 7-day HK (age 28 days) is not observed significant changes in the content of NO in the examined tissues.

The largest production of NO in the tissues of the atria and ventricles was found after 30-day hypokinesia. This is most likely due to the nature of this age - the beginning of puberty, it is possible to have a stress - limiting reaction.

As our model consists of two components: direct hypokinesia and stress of the applied procedures, it means that there are NO-dependent mechanisms of the body's reaction to hypokinesia and immobilization stress. The results may help to explain the role of nitric oxide in the mechanisms of the formation and the occurrence of various diseases and will assess the impact of stress conditions on the generation of nitric oxide in the growing organism.

NITRIC OXIDE IS

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Hypokinesia (HK) social problems for all age regulatory mechanisms in endogenous nitric oxide, an EPR spectroscopy it was shown 21-day-old, in the heart, live production was observed.

Increased intensity relationship the NO levels consideration of the literature system, the internal organs, that some of these changes oxide, a key activity for the

The use of a non-stress in a reduction of NO to a results show that in the in contribution has an enzyme proportion of NO, formed by to hypokinesia rats caused which suggests that the increase during hypokinesia.

Significant NO role as lack of an information hypokinesia of the growing quantification of NO will oxide in the growing organism control the adaptive reaction where the determination of This research deserves pharmacology and pediatric

The obtained data NO-synthase in the inter postnatal ontogenesis. Due excluded and shifts of the humans, and this may be o