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# BRAIN CORTEX TEMPERATURE AND HEART RATE IN NEWBORN RATS AFTER INFLUENCING ENDOGENOUS NITRIC OXIDE SYNTHESIS

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The significant role of nitric oxide (NO) in various functions of organism was confirmed during last two decades. Among others its action as a retrograde messenger and/or neuromodulator in synaptic transmission has been shown (5, 11) and its circulatory effect has been identified as that of vascular endothelial relaxing factor, EDRF (7). NO vasodilatory effects explain the well known beneficial role of various nitro-compounds on local blood flow in many important regions of the human body. The classical studies about positive effects of NO on the cerebral circulation (4, 6) as well as improved hemodynamics in lungs after NO administration, including newborns (2) showed possible new therapeutic strategies.

The effect of NO, however, are not uniformly positive, as e.g. during focal cerebral schemia it impairs its outcome (1). The site of NO producing enzyme – NO synthase (NOS), that is colocalized with NADPH-diaphorase, in cerebral arteries and cranial parasympathetic ganglia points to its important role in cerebral blood flow regulation (9). The informations about a general hemodynamic effects were obtained from experiments with intracerebroventricular or intravenous administration of drugs influencing endogenous NO (3).

We have interested generally in previous research area in mechanisms of learning and memory during early ontogeny and have shown a significant participation of NO in these phenomena (8, 10). To obtain information on consequences of NO manipulations on some circulatory parameters we injected intraperitoneally (i.p.) a NOS blocker N-nitro-L-arginine on the one hand, and NOS substrate, L-arginine, on the other hand, in acute experiments. Changes of brain and body temperature and heart rate were monitored and finally evaluated.

## MATERIAL AND METHOD

Twenty-four rat pups, Long-Evans strain, 4–11 days of age, were injected by 10 mMol nitro-L-arginine or 20 mMol L-arginine; both i.p. administration in physiological saline solution. Physiological saline alone was used as control. All experiments were done under general anaesthesia (Equithesine 0.2–0.3 ml /100 g of body weight); in several experiments

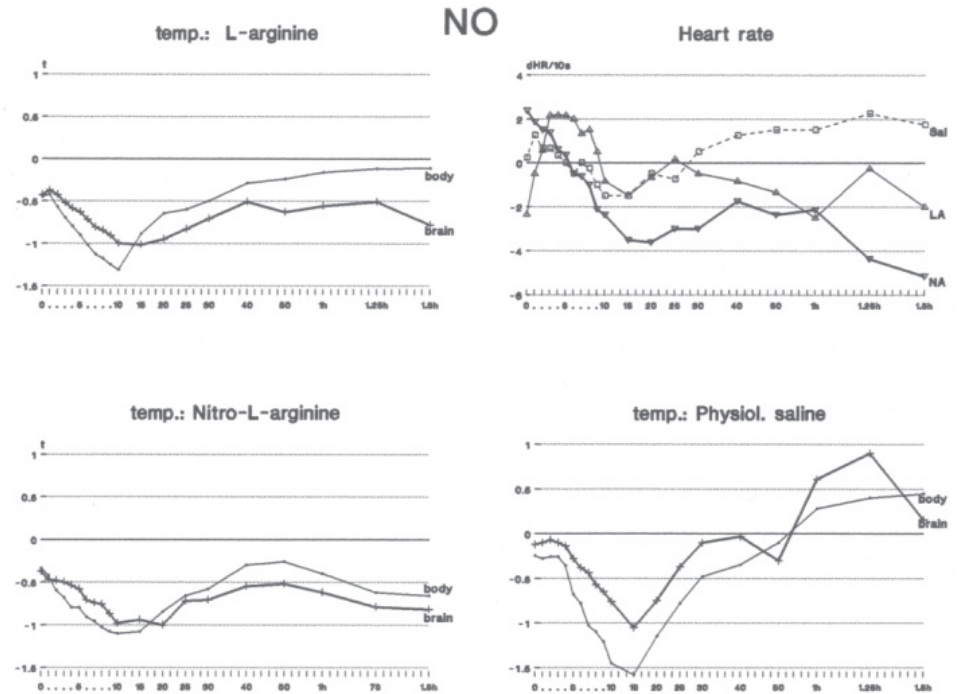


**Fig. 1** Temperature and heart rate changes after intraperitoneal administration of L-arginine, N-nitro-L-arginine and saline in 4–11 days old rat pups

the drugs were applied consecutively with a 2-hour interval. The animal lying on and covered with cotton wool was warmed from sides by a bent metallic tubing (10 mm in diameter) flushed by water, 37 °C warm. Body temperature was measured in a skin fold, brain temperature was recorded in the parietal cortex by means of a thermally insulated, glass-covered thermistor tip (external diameter 1 mm) and heart rate was registered on Bioscript BST 1 with stainless steel electrodes from distal limbs. Recording continued for 1–3 hours after injection and the data were collected every minute up to 10 min, then every 5 min up to 30 min, every 10 min up to 1 hour and thereafter every 15 minutes. The differences related to the values obtained before drug administration were statistically evaluated by means of analysis of variance (ANOVA).

## RESULTS

After L-arginine i.p. (Fig. 1), the changes of the brain temperature, even if small, displayed a typical trend, characterized by a decrease during the first ten minutes after injection, and a slow increase with oscillations thereafter. In the first 10 minutes, the

decreasing trend was significant; ANOVA: $F(10,108) = 2.625$; $p < 0.01$. Also was the difference between the brain and body temperature; ANOVA: $F(1,108) = 5.203$; $p < 0.01$. The difference between the heart rate values to 8 min. after injection vs. those up to 1 hour met the significance level, too; ANOVA: $F(1,106) = 58.107$; $p < 0.001$.

Intraperitoneal administration of nitro-L-arginine did not substantially change the brain temperature, and its small fluctuations did not show any significant trend or difference relative to body temperature oscillations. No significant difference appeared between the brain and body temperature immediately to 10 min. after injection. The immediate heart rate rise with its gradually decrease during the first eight minutes differed significantly from the following intervals; ANOVA (0–8 vs. 9–60 min): $F(1,142) = 22.977$; $p < 0.001$. The decrease of the heart rate during the first hour after nitro-L-arginine was significant, too; ANOVA: $F(17,126) = 1.703$; $p < 0.05$.

The differences in immediate changes of brain temperature between animals applied L-arginine and nitro-L-arginine were not significant; the heart rate difference within the first two minutes after administration was, however, significant; ANOVA: $F(1,43) = 5.881$; $p < 0.025$.

After i.p. injections of saline, there were observed oscillations of the brain and body temperature that were not significant; the differences between their values were, however, significant; ANOVA: $F(1,152) = 5.326$; $p < 0.025$.

## CONCLUSIONS

Our results show that after administration of substrate for NOS, L-arginine, only minute changes of brain relative to body temperature and, immediately after injection, only small (nevertheless significant) differences of heart rate between animals given L-arginine and those applied NOS blocker, nitro-L-arginine, appear. This is probably due to a reflex response in which the carotid bodies might play a decisive role, as shown by some findings (12), rather to a direct effect on brain tissue.

## SUMMARY

Possible role of gaseous messenger and/or neuromodulator, nitric oxide (NO), in regulation of brain cortical and body temperature of laboratory rat pups was evaluated. Measurement of temperature (and heart rate) after intervention in NO metabolism confirmed its effects which revealed a complex consequences in various tissues (especially in the vascular endothelium and neurons).

## *Teplota kůry mozku a srdeční frekvence u novorozených laboratorních potkanů po ovlivnění syntézy oxidu dusnatého*

SOUHRN

Funkce plynného mediátoru a neuromodulátoru – oxidu dusnatého (NO) v mechanismu regulace mozkové a tělní teploty byla zkoumána u mláďat laboratorního potkana. Analýza změn teplot (a srdeční frekvence) po ovlivnění metabolizmu NO potvrdila jeho předpokládanou roli, která zahrnuje komplexní reakce v různých typech tkáně (zejména v cévním endotelu a v neuronech).

*In memory of prof. J. Mysliveček, MD, DSc, a great Czech neurophysiologist, which inicialized the nitric oxide research in Department of Pathophysiology, Charles University, Faculty of Medicine in Pilsen during his "second period" 1990–1997.*

REFERENCES

1. Buisson A., Plotkine M., Boulu R. G.: The neuroprotective effect of a nitric oxide inhibitor in a rat model of focal cerebral ischemia. Brit. J. Pharmacol. 106, 1992: 766–7. – 2. Day R. W., Klitzner M. J., Ignarro L. J.: Endothelium dependent relaxation and the acute pulmonary vascular response to alveolar hyperoxia in neonatal pig. Biol. Neonate 63, 1993: 389–96. – 3. El Karib A. D., Sheng J. J., Betz A. L. et al.: The central effects of nitric oxide inhibitor N-omega-nitro-L-arginine on blood pressure and plasma renin. Clin. Exp. Hypertension 15, 1993: 819–32. – 4. Faraci F.: Regulation of the cerebral circulation by endothelium. Pharmacol. Ther. 56, 1992: 1–22. – 5. Hölscher C.: Nitric oxide, the enigmatic neuronal messenger: its role in synaptic plasticity. Trends Neurosci. 20, 1997: 298–303. – 6. Iadecola C.: Regulation of the cerebral microcirculation during neuronal activity – is nitric oxide the missing link? Trends neurosci. 16, 1993: 206–16. – 7. Moncada C., Palmer R. M. J., Higgs E. A.: Nitric oxide: Physiology, pathophysiology and pharmacology. Pharmacol. Rev. 43, 1991: 109–142. – 8. Mysliveček J.: Inhibitory learning and memory in newborn rats. Prog. Neurobiol. 53, 1997: 399–430. – 9. Pelligrino D. A., Koenig H. M., Albrecht L. F.: Nitric oxide synthesis and regional blood flow response to hypercapnia and hypoxia in the rat. J. Cereb. Blood Flow Metab. 13, 1993: 80–7. – 10. Petrovický P., Barcal J., Mysliveček J.: Nitric oxide synthase (NADPH-diaphorase) content in brain neurons of neonatal rats after inhibitory learning and intervention into nitric oxide metabolism. Neurosci. 89, 1999: 1151–7. – 11. Snyder S. H.: Nitric oxide and neurons. Current Opin. Neurobiol. 2, 1992: 323–7. – 12. Wang Z. Z., Bredt D. S., Fidone S. J. et al.: Neurons synthesizing nitric oxide innervate mammalian carotid body. J. Comp. Neurol. 336, 1993: 419–32.

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