

# Changes in the level of phosphates, production of nitric oxide and pH levels of rats blood after spinal cord injury

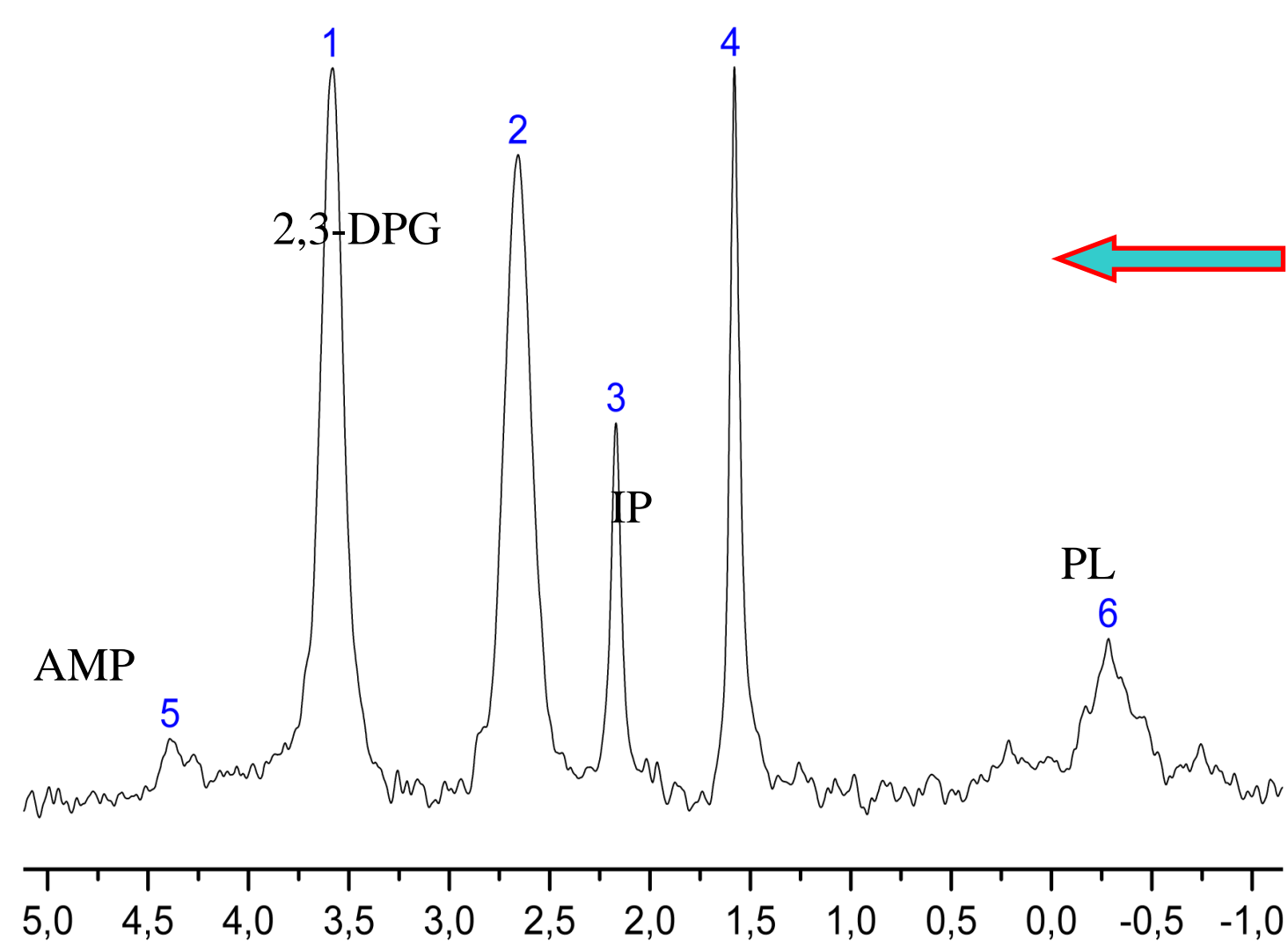
V. Andrianov<sup>1,2</sup>, G. Yafarova<sup>1,2</sup>, S. Yurtaeva<sup>2</sup>, M. Volkov<sup>2</sup>, K. Gainutdinov<sup>1,2</sup>

<sup>1</sup>Kazan Federal University,

<sup>2</sup>Zavoisky Physical-Technical Institute of the Russian Academy of Sciences, Kazan, Russia

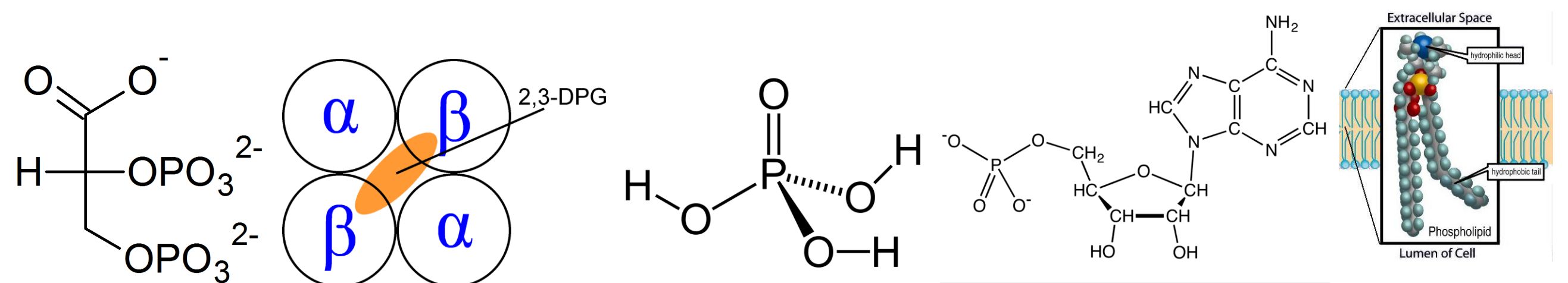
## Redox Signalling

The search for new approaches to treatment of spinal cord injury (SCI) should take into account the multifactorial nature of metabolic disorders in SCI using new methods to obtain information about the molecular mechanisms. Therefore we investigated metabolic changes in the blood to assess possible shifts in redox system on a model of dosed SCI in rats. It was measured the level of organic and inorganic phosphorus compounds, pH level of blood by <sup>31</sup>P NMR-spectroscopy and nitric oxide (NO) production by EPR-spectroscopy.

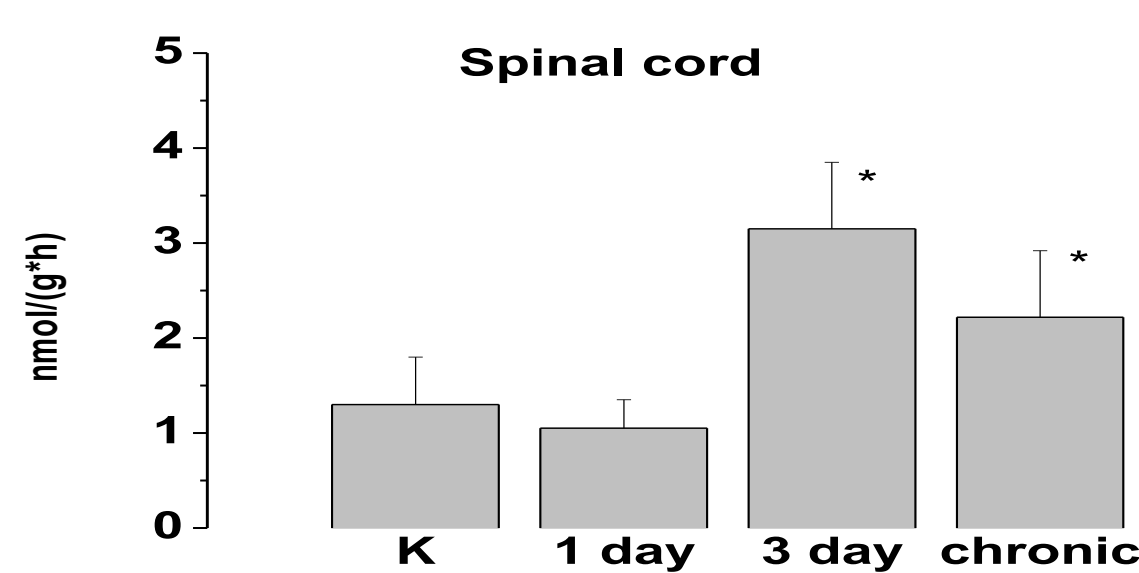
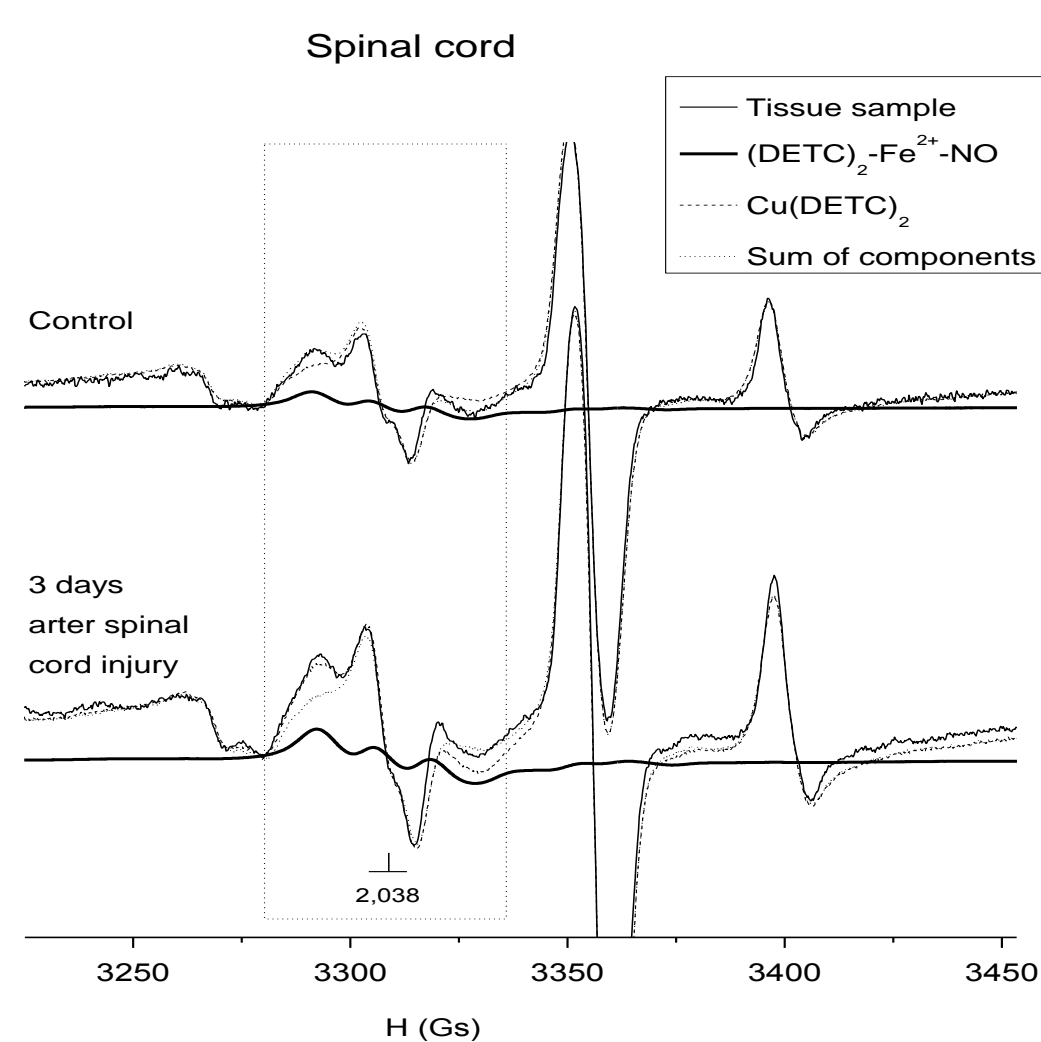


Typical <sup>31</sup>P NMR blood spectrum of injured rat

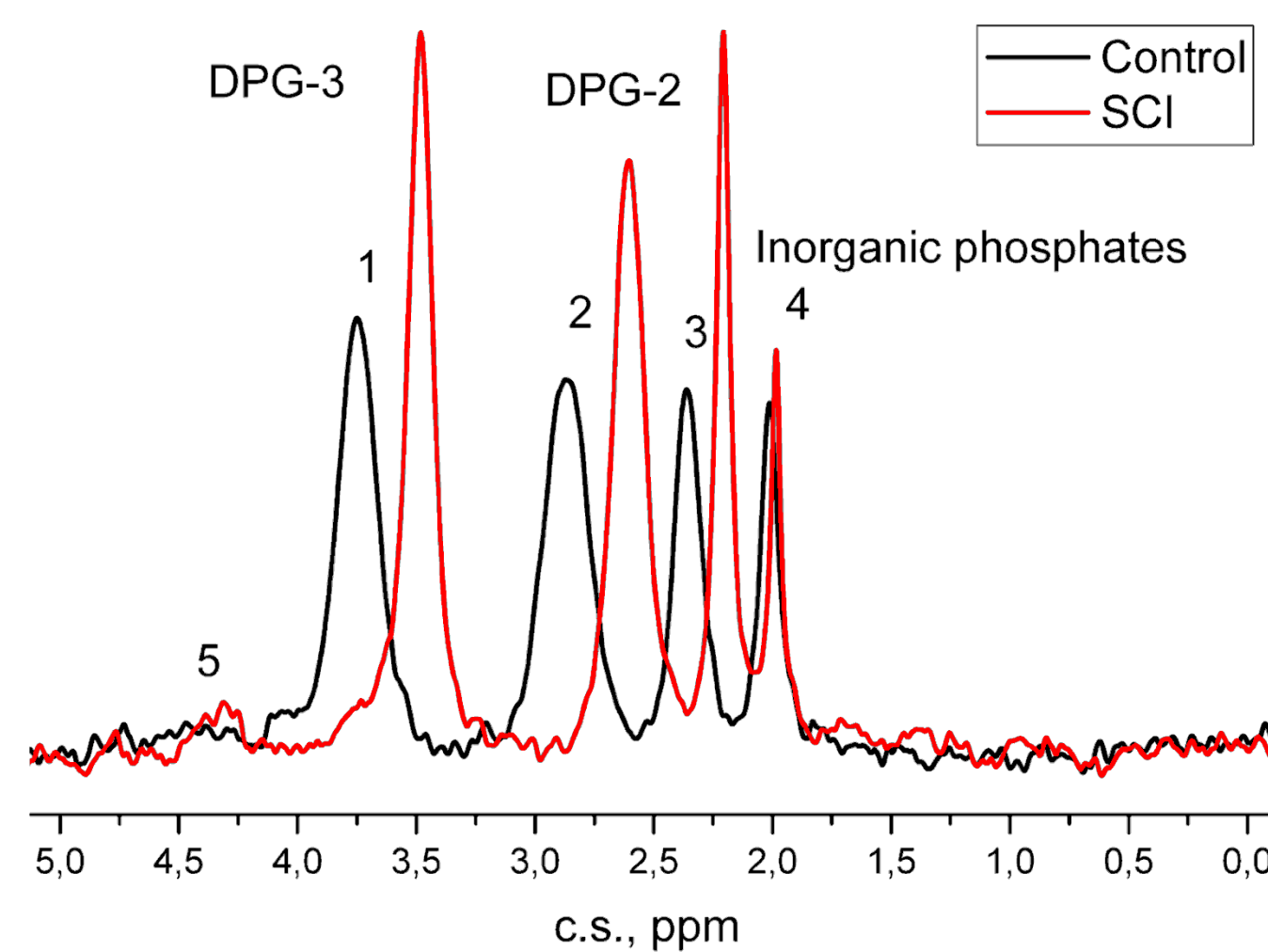
The blood samples of two groups of experimental animals (intact rats and rats after experimental model of spinal cord contusion) were investigated. In <sup>31</sup>P NMR spectra of blood we detected the following intensive signals (Fig.1): the signals of 2,3-diphosphoglycerates (2,3-DPG) – signals 1, 2, the signals of inorganic phosphates – signals 3, 4, in some cases weak signals of adenosine monophosphate (AMP) – signal 5, and phospholipids (PL) – signal 6.



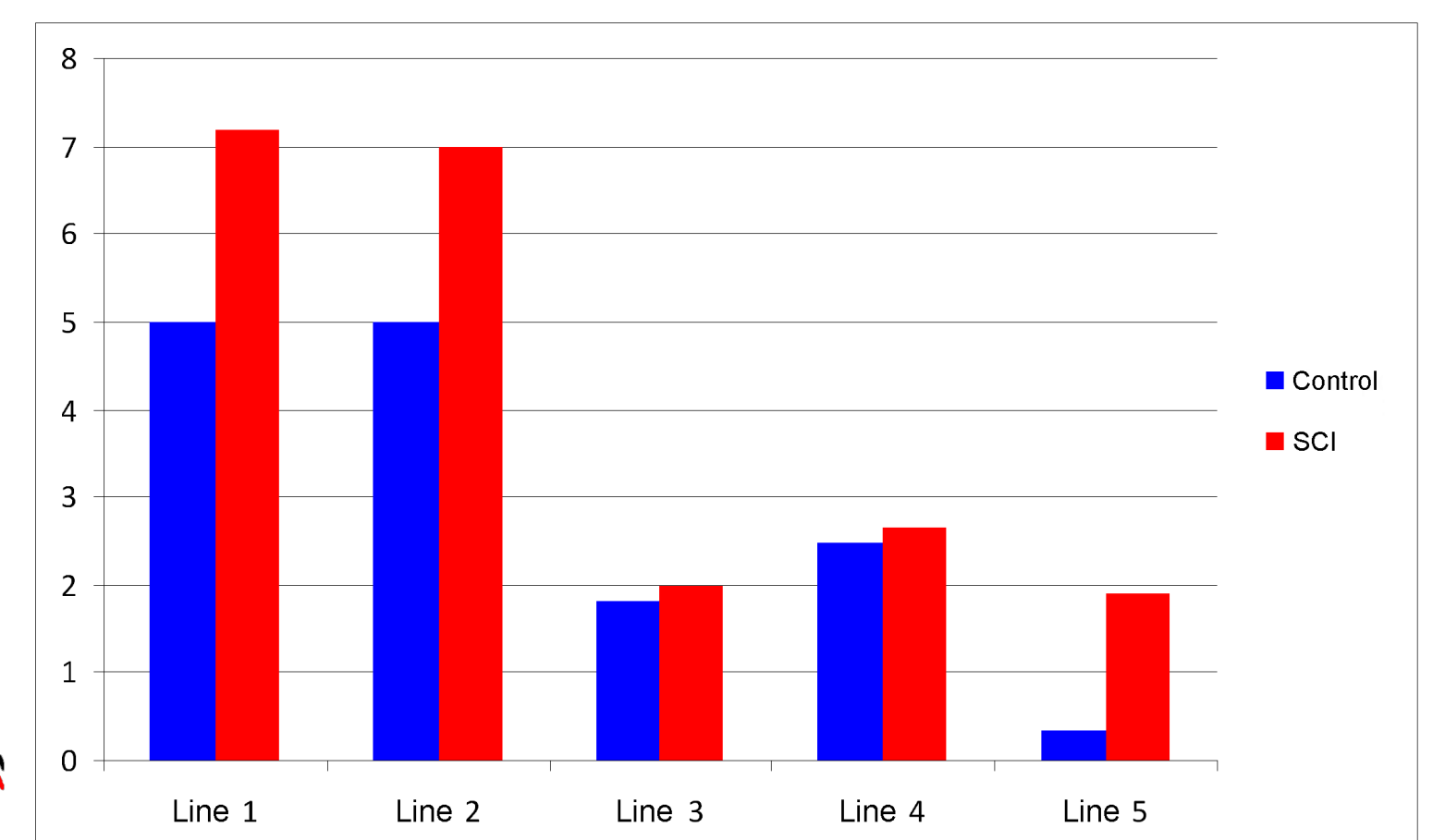
Phosphoric compounds in blood: 2,3-DPG, inorganic phosphates, AMP, phospholipids



Change in NO production after spinal cord injury.



The comparison of <sup>31</sup>P NMR signals of blood of injured and intact rats. In the blood of injured rats the increase of 2,3-DPG and inorganic phosphate signals intensities were observed, that means that the hypoxia in condition of SCI is growing. In the blood spectra of injured rats we can see displacement of the most intensive lines 1, 2, 3, 4 to the lower chemical shifts that indicates the decrease of the environment pH.



The changes in the integrated intensity of NMR lines after SCI

All the experiments were performed on an "Avance 400" Bruker NMR spectrometer with using 5 mm glass ampoules at the temperature 20°C.

It was found increase the content of inorganic phosphates, phospholipids and monophosphates, as well as a decrease in rat's blood pH 3 days after SCI. There were also an increase in the amount of 2,3-diphosphoglycerate in red blood cells, which is one of the ways to increase oxygen supply to tissues under hypoxia, affecting the affinity of hemoglobin to oxygen. It was also found the increased NO production in damaged spinal cord tissue, blood, liver and heart. It is known that hypoxia induced synthesis of vasoactive metabolites, such as NO, increased the flow of blood oxygen to the tissues due to vasodilatation. Thus, the information obtained by NMR and EPR-spectroscopy makes it possible to determine the metabolic parameters that characterize the state of microcirculation and vasomotor reactivity, as well as the degree of tissue hypoxia and changes of acid-base balance in the blood with SCI. Activation of the NO production system and the detected increase in 2,3-diphosphoglycerate after SCI indicate compensatory-adaptive changes.

Supported RFBR (grant 17-04-01746).