

DEVELOPING OF METHOD TO MEASURING OF THE  $\text{Na}^+\text{K}^+\text{ATPase}$  ACTIVITY  
IN GUINEA PIG LIVER UNDER *IN SITU* PERFUSION

Santos, M.C.B.<sup>1</sup>; Ferreira, A.C.R.<sup>1</sup>; Gonçalves, C.F.<sup>1</sup>; Castro-Faria Neto, H.C.<sup>2</sup>;  
Burth, P.<sup>3</sup>; Castro-Faria, M.V.<sup>1</sup>

<sup>1</sup>DBCG, IBRAG, UERJ, RJ, Brasil; <sup>2</sup>DFF, Fiocruz, RJ, Brasil; <sup>3</sup>DBCM, UFF, Niterói,  
Brasil.

We had shown that leptospiral glicolipoproteic endotoxin (GLP) is a potent and specific  $\text{Na}^+\text{K}^+\text{ATPase}$  inhibitor *in vitro*. We had proposed that, after organ colonization (mainly liver), lysis of leptospira consequent to the host immune response would release GLP, leading to the inhibition of  $\text{Na}^+\text{K}^+\text{ATPase}$ , thus, the characteristic liver malfunctions seen in this disease. In this communication, we propose a technique for the measurement of the activity of  $\text{Na}^+\text{K}^+\text{ATPase}$  in liver through hepatocyte incorporation of  $\text{Rb}^+$  during *in situ* liver perfusion. Guinea pigs under anesthesia were submitted to laparotomy. The portal vein were cannulated and livers were perfused with a modified Hank's solution, by addition of  $\text{RbCl}$ , in presence or not of ouabain. Finishing the perfusion, Hank's solution was substituted by cold saline containing or not  $\text{Ba}^{2+}$ . Liver were then removed, weighted and mineralized with nitric acid.  $\text{Rb}^+$  was determined by atomic absorption spectrometry. Results show the kinetics of  $\text{Rb}^+$  incorporation, the  $\text{Ba}^{2+}$  inhibits the  $\text{Na}^+\text{K}^+\text{ATPase}$  activity *in vitro* and can't be used in Hank's solution. Cold saline containing  $\text{Ba}^{2+}$  decrease  $\text{Rb}^+$  incorporation by liver cells. When Hank's solution had ouabain, the  $\text{Na}^+\text{K}^+\text{ATPase}$  activity decrease 77.3% as occur *in vitro*. This is appropriated technique for the study of the activity of  $\text{Na}^+\text{K}^+\text{ATPase}$  *in situ*.

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