PARTIAL CHARACTERIZATION OF ATP, ADP AND AMP HYDROLYSIS IN RAT CARDIAC SOLUBLE FRACTION

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The cardiovascular effects of adenine nucleotides and adenosine have been described in many different physio-pathological processes. Several enzymes are capable of hydrolyze extracellular nucleotides, modulating the purine signaling. These include E-NTPDases and 5'-nucleotidase. The objective of this study is to characterize soluble enzymes involved in ATP, ADP e AMP hydrolysis in heart. Soluble fraction was obtained from centrifugation of ventricles homogenate. The preparation was incubated with ATP, ADP or AMP (2.0 mM) at 37°C. The reaction was stopped by the addition of TCA and the amount of Pi liberated was measured by the method of Chan et al., 1986. The protein concentration and the incubation time were chosen in order to ensure the linearity of the reaction. Then, for ATP, ADP and AMP assays were chosen 90 µg/tube of protein and 6, 25 and 40 minutes of incubation, respectively. The inhibitor olygomycin was used in all enzymatic ATP and ADP assays and the inhibitor NEM was used in all experiments with ADP. NTPDase and 5'-nucleotidase were cation-dependent and 5'-nucleotidase showed a preference for ion Mg²⁺. Our results demonstrate ATP, ADP and AMP hydrolysis by soluble fraction obtained from rat heart. The study of the enzymes that modulate the ratio nucleotides/nucleoside in the heart could be important for the understanding of the processes related to signaling of these molecules.