

## Phenol Enhances Acto-S1 Interaction

Machado, L.E.S.F<sup>1</sup>, Salerno, V.P.<sup>1,2</sup>, Sorenson, M.M<sup>1</sup>.

<sup>1</sup>Instituto de Bioquímica Médica-CCS/UFRJ, <sup>2</sup>Departamento de Biociências da Atividade Física-EEFD/URFJ - Rio de Janeiro, Brazil.

**Introduction and Objective:** Phenol (1-7%) is an organic solvent used to alleviate muscle spasticity, leading to neurolysis and reducing hyperexcitability of the muscle. However, phenol (0.1-0.5%) reduces isometric tension, and also increases  $Mg^{2+}$ -ATPase activity of myosin subfragment-1 (S1) from skeletal muscle. Previously, we showed a high actin-activated and S1- $Mg^{2+}$ -ATPase activity, and more molecules of S1 bound to actin in the presence of phenol. The aim of this work was to analyze the mechanism of action of phenol on acto-S1 interaction using fluorescence, light-scattering and fast-kinetics techniques on proteins purified from skeletal muscle. **Results and Conclusion:** In low KCl buffer, where acto-S1 binds strongly (20 mM KCl, 50 mM HEPES pH 7, S1, actin, 21°C), phenol (0.5 %) enhances light scattering of acto-S1 and leads to greater fluorescence quenching of pyrene-acto-S1 than in the control. This result demonstrates that phenol promotes a very strong interaction between S1 and actin. Kinetically, we observed an increase in pyrene-acto-S1 association rate ( $k_{-5}$ ) and a decrease in pyrene-acto-S1 dissociation rate ( $k_{+6}$ ,  $k_{+5}$ ), regardless of whether ATP (40  $\mu$ M) or unlabeled actin (10  $\mu$ M) was used to induce pyrene-actin dissociation. Thus phenol affects acto-S1 association and dissociation, to enhance acto-S1 affinity, altering the kinetic steps involved.

Keywords: pyrene-acto-subfragment-1, phenol.

Support: PIBIC, CNPQ, PRONEX, FAPERJ