

An Ecto-Pyrophosphatase Activity From *T. rangeli* Epimastigotes
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Phosphate is a nutrient needed to almost all cellular functions. The starvation of this nutrient causes injuries to cells, especially to the microorganisms that live in several hostile environments. The life cycle of *Trypanosoma rangeli* is better characterized in invertebrate hosts and it starts with ingestion of blood trypomastigotes during the blood meal by the insect. Once in midgut, the parasites differentiate into epimastigote forms that multiply and across the intestinal barrier, achieving the hemolymph. From hemolymph, the parasites drive to salivary glands to perform metacyclogenesis, producing metacyclic trypomastigotes, infective forms to vertebrate hosts. During its life cycle, the protozoa pass through different compartments of insect, that present different phosphate content. So, it is of great importance the study of enzymes that release inorganic phosphate from several phosphorylated compounds. In this context, we have characterized a pyrophosphatase activity on the external cell surface of *T. rangeli*. This activity is stimulated by $MgCl_2$ but not by $MnCl_2$, $CaCl_2$, $SrCl_2$ and $ZnCl_2$. This activation occurs in a dose-dependent manner, with a $S_{0.5}$ value of 1.0mM $MgCl_2$. Sodium orthovanadate and ammonium molybdate, two acid phosphatase inhibitors, and levamisole, alkaline phosphatase inhibitor, did not inhibit the ecto-pyrophosphatase activity. However, sodium fluoride inhibited the ecto-pyrophosphatase activity in a dose-dependent manner. The pyrophosphatase activity of *T. rangeli* was stimulated in pH 7.5-8.5, presenting optimum pH at alkaline range. The addition of $CaCl_2$ inhibits at 50% the Mg^{2+} -dependent ecto-pyrophosphatase activity. We are studying now the response of *T. rangeli* ecto-pyrophosphatase activity to inorganic phosphate content in culture medium and the effects of specific inhibitors of pyrophosphatases.

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