Characterization of a Recombinant Family I Inorganic Pyrophosphatase and a mutant (C339A) from the embryos of Cattle Tick *Rhipicephalus (Boophilus) microplus*

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The cattle tick, *Rhipicephalus (Boophilus) microplus*, hinders livestock production in tropical and subtropical parts of the world where it is endemic, and thus major efforts have been directed toward developing immunoprophylactic tick control tools. The family I inorganic pyrophosphatase (PPase, EC 3.6.1.1) is an essential metal-dependent enzyme that catalyzes the hydrolysis of inorganic pyrophosphate (PP\(_i\)) into orthophosphate, which is formed mainly as a by product of the many biosynthetic reactions that utilize ATP, playing an important role in cell anabolism. In order to understand how critical its role is during tick embryogenesis, we are studying many biochemical aspects of this enzyme. In this current work we report the characterization of recombinant protein (rBmPPase) wild type and a mutant obtained by site-directed mutagenesis, called Mut1 (C339A). The results demonstrate that rBmPPase wt is more sensitive to fluoride and calcium than Mut1. The rBmPPase wt activity was inhibited about 75% by 50 mM fluoride and totally abolished by 500 mM. Calcium treatment inhibited about 30% by 10 nM and remained around 50% from 100 nM up to 500 nM. In addition, we could observe that 1 mM H\(_2\)O\(_2\) decreased rBmPPase wt activity in about 30%. On the other hand, Mut1 activity was inhibited about 30% by 50 mM fluoride and 90% by 1 mM. Remarkably, Mut1 was not inhibited up to 500 nM calcium. Finally, we are also investigating whether other oxidizing and reducing agents can modulate rBmPPase wild type and Mut1 activities.

**Keywords:** tick, redox control, embryo, pyrophosphatase

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