

Structural and Biochemical Characterization of a New Inorganic Pyrophosphatase from the Cattle Tick *Rhipicephalus (Boophilus) microplus*

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Soluble inorganic pyrophosphatase (sPPase, EC 3.6.1.1), is an essential metal-dependent enzyme that converts pyrophosphate into orthophosphate. This enzyme is represented by two families: PPase Family I ( $Mg^{2+}$ -dependent) and Family II ( $Mn^{2+}$ -dependent) distributed among living organisms which typically produce either one or another. In the present work, we report the cloning of a 1020 bp cDNA encoding the sPPase from *R. microplus* (RmPPase) and the characterization of the protein. The RmPPase cDNA encodes a putative protein of 341 aa with theoretical molecular mass and pI values of 38.7 kDa and 5.56, respectively. Multiple alignment of RmPPase with other PPase sequences revealed the conservation of three characteristic Asp residues of the family I PPase signature domain (PDOC00325), and 10 other residues surrounding the active-site. Moreover, potential phosphorylation sites on RmPPase were predicted by SCAN PROSITE including casein kinase II phosphorylation and N-myristoylation sites. Relative expression of RmPPase mRNA was determined in fat body, midgut and ovary, from two developmental stages, partially and fully engorged females. However, higher transcription levels were found in ovaries. Structurally, a remarkable characteristic of RmPPase was the presence of Cys residues in 138 and 339 positions, which differed from other family I PPases with solved structure. Taken together these data describe the structural and biochemical characteristics of a new member of family I PPase.

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