Kinetic Properties of a Membrane-Bound Acid Phosphatase Isolated from *Burkholderia*.

Rombola, T.H.[1], Pedrinho, E.A.N.[1], Lemos,E.G.M.[1] , Sato, V.S.[1], Pizauro, J.M.[1]
[1]Depto. Tecnologia, FCAVJ-UNESP.

The membrane-bound acid phosphatase (MBAP) was purified from *Burkholderia* isolated from an agricultural soil in Ponta Grossa-Paraná State-Brasil and identified by the 16S rDNA gene analysis. The enzyme main characteristic enzyme is its similar reactivity toward p-nitrophenylphosphate, inorganic pyrophosphate, ATP and tyrosine phosphatase. The enzyme showed a broad substrate specificity hydrolyzing different substrates at pH 6.0, such as PNPP (103.8 U/mg), ATP (98.9 U/mg), and pyrophosphate (117.2 U/mg). The hydrolysis of PNPP, ATP and pyrophosphatae shown cooperativy kinetics (n ranging from 1.3 –1.4) with $K_{0.5}$ values of 0.06 mM, 0.07 mM and 0.12 mM, respectively. The kinetic data confirmed that the same enzyme hydrolyzed these substrates. Additionally, the phosphotyrosine phosphatase and acid phosphatase enzymatic properties were identical. The PNPPase activity was inhibited by arsenate, phosphate, but not by calcium, levamisole, EDTA, zinc, magnesium, cobalt, ouabain, oligamycin and pantoprazol. It is generally believed that *Burkholderia* species are versatile organisms that solubilize insoluble minerals through the acids production. Our results suggest that the production of a membrane-bound acid phosphatase might be one mechanism of mineral phosphate solubilization performed by this microorganism, which in turn increase the availability of nutrients to plants.

Financial support: FAPESP, CNPq and CAPES.

Key Words: acid phosphatase, pyrophosphatase, p-nitrophenyl phosphate