

H⁺-UPTAKE AND H⁺-RELEASE IN ACIDOCALCISOMES OF *HERPETOMONAS SP.*

Soares Medeiros, L.C.A.; Tesch Ferreira, K.R.A., Barrabin, H.

Instituto de Bioquímica Médica, CCS
UFRJ, Rio de Janeiro, Brazil

Acidocalcissomes are acidic calcium storage organelles first described in trypanosomatids and then found in several microorganisms. They are electron-dense, possess a surrounding membrane, have variable size, and contain high amounts of Ca²⁺, Mg²⁺, Zn²⁺, Na⁺, and short and long chain polyphosphate. They have exchangers and a V-H⁺-PPase. This enzyme is similar to those found in plant tonoplast. Using *Herpetomonas* sp. lysed with glass beads, a pyrophosphate-driven H⁺-uptake was measured. This uptake was inhibited by sodium fluoride (NaF), imidodiphosphate (IDP) and dicyclohexylcarbodiimide (DCCD). H⁺ uptake was not promoted by ATP. The rate of H⁺ uptake was activated up to 100-250 μM pyrophosphate and inhibited at higher substrate concentrations. The addition of IDP, a non-hydrolysable PPI analogue, AMDP (aminomethylenediphosphonate), a PPI analogue and specific inhibitor of plant vacuolar pyrophosphatases, EDTA (Mg²⁺ chelator), DCCD or even high PPI concentrations promoted H⁺ release. On the other hand, addition of 100 μM Ca²⁺ or other divalent cations like Zn²⁺, but not choline or lysine (a basic amino acid present in acidocalcissomes matrix), induced a further release of H⁺. These results suggest that acidocalcissomes from *Herpetomonas* possess a Ca²⁺/H⁺ exchanger. The H⁺ release evoked by Zn²⁺ addition also suggests a putative Me²⁺/H⁺ exchanger. The unusual PPI dependence suggests the existence of a regulatory mechanism of H⁺ release dependent of PPI. SUPPORTED BY CNPq, CAPES, FAPERJ