PROTON TRANSPORT IN ACIDOCALCISOMES IN HERPETOMONAS AND LEPTOMONAS

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Acidocalcisomes are acidic calcium storage organelles found in several microorganisms. They are characterized by their acidic nature, high electron density, high content of polyphosphates and heavy metals like Zn and Fe. Electron microscopy contrast tuned images of *Herpetomonas* sp and *Leptomonas wallacey* shows the presence of several electron dense organelles ranging from 100 to 300 nm in size. X-ray element mapping associated with energy-filtering transmission electron microscopy shows that most of Na, Mg, P, K, Fe and Zn were located in their matrix. Using acridine orange as an indicator dye, a pyrophosphate-driven H^+ uptake is measured in permeabilized by digitonin as well as in mechanically disrupted cells. H^+ uptake is not promoted by ATP in any parasite, even in mechanically disrupted cells. The pyrophosphate-dependent H^+ uptakes are dependent of ion K^+ and inhibited by Na^+. This uptake is also inhibited by sodium fluoride (NaF) and imidodiphosphate (IDP), aminomethylenediphosphonate AMDP (three H^+-pyrophosphatase inhibitors) and by dicyclohexylcarbodi-imide (DCCD). Addition of 50 µM free Ca^{2+} induces the release of H^+, suggesting the presence of a Ca^{2+}/H^+ countertransport. Unexpectedly, high levels of PPi or IDP induces fast release of H^+ from acidocalcisomes. This release was independent of Mg^{2+}. Since a PPi/H+ antiport is unlikely, it is suggested that the action of PPi must act by stimulating some intrinsic (unknown) mechanism of H^+ release. Together, these results suggest that the electron dense organelles found in *Herpetomonas* sp and *Leptomonas* are homologous to the acidocalcisomes described in other trypanosomatids, however, they have not ATP promoted H^+ transport and presents a mechanism of H^+ release stimulated by PPi.

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