Influence of Ecto-Nucleoside Triphosphate Diphosphohydrolase Activity on *Trypanosoma cruzi* Infectivity and Virulence

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The protozoan *Trypanosoma cruzi* is the causative agent of Chagas disease. There are no vaccines or effective treatment, especially in the chronic phase when most patients are diagnosed. There is a clear necessity to develop new drugs and strategies for the control and treatment of Chagas disease. Recent papers have suggested the ecto-nucleotidases (from CD39 family) from pathogenic agents as important virulence factors. In this study we evaluated the influence of Ecto-Nucleoside-Triphosphate-Diphosphohydrolase (Ecto-NTPDase) activity on infectivity and virulence of *T. cruzi* using both in vivo and in vitro models. We followed Ecto-NTPDase activities of Y strain infective forms (trypomastigotes) obtained during sequential sub-cultivation in mammalian cells. ATPase/ADPase activity ratios of cell-derived trypomastigotes decreased 3- to 6-fold and infectivity was substantially reduced during sequential sub-cultivation. Surprisingly, at third to fourth passages most of the cell–derived trypomastigotes could not penetrate mammalian cells and have differentiated into amastigote-like parasites that exhibited 3-4 fold lower levels of Ecto-NTPDase activities. To evidence the participation of *T. cruzi* Ecto-NTPDase1 in the infective process, we evaluated the effect of known Ecto-ATPDase inhibitors (ARL 67156, Gadolinium and Suramin), or anti-NTPDase-1 polyclonal antiserum on ATPase and ADPase hydrolytic activities in recombinant *T. cruzi* NTPDase-1 and in live trypomastigotes. All tests showed a partial inhibition of Ecto-ATPDase activities and a marked inhibition of trypomastigotes infectivity. Mice infections with Ecto-NTPDase-inhibited trypomastigotes produced lower levels of parasitemia and higher host survival than with non-inhibited control parasites. Our results suggest that Ecto-ATPDases act as facilitators of infection and virulence in vitro and in vivo and emerge as target candidates in chemotherapy of Chagas disease.

Palavras Chaves: *Trypanosoma cruzi, ecto-nucleotidase, infectivity, virulence.*
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