T. cruzi Cell Death Caused by *Cratylia mollis* Seed Lectin is Mediated by Plasma Membrane Permeabilization Followed by Mitochondrial Ca^{2+} Overload and ROS Production

Fernandes, M. P.[1]; Inada, N. M.[1]; Araújo, F. F. B.[2]; Gadelha, F. R.[3]; Alves, M. J. M.[4]; Correia, M. T. S.[2]; Coelho, L. C. B. B.[2]; and Vercesi, A. E.[1]

[1]Dept. Patologia Clínica, FCM, UNICAMP, Campinas, SP, Brazil; [2]Dept. Bioquímica, CCB, UFPE, Recife-PE, Brazil; [3]Dept. Bioquímica, IB, UNICAMP, Campinas, SP, Brazil; [4]Dept. Bioquímica, IQ, USP, São Paulo, Brazil.

Lectins are proteins or glycoproteins that serve as tools in glycobiology research and can be employed for the detection of cell surface glycoconjugates. This work was aimed at evaluating whether *Cratylia mollis* seed lectin (Cramoll 1,4) have toxic effects on *Trypanosoma cruzi*. Cramoll 1,4 recognized glycoconjugates present on parasite cell surface leading to agglutination of the epimastigotes and trypomastigotes in a dose-dependent manner. Trypomastigotes plasma membrane permeabilization by Cramoll 1,4 was documented by fluorescence microscopy using propidium iodide. Lectin decreased the epimastigote proliferation, reaching 93% inhibition at 50 µg/ml. Incubation of epimastigotes (1.25×10^8 /ml) in the presence of Cramoll 1,4 (50 µg/ml) and 10 µM Ca^{2+}, during 1 h, induced plasma membrane permeabilization followed by mitochondrial Ca^{2+} overload. This increased the production of reactive oxygen species (ROS) by 5 times, significantly decreasing the mitochondrial membrane potential and impairing ADP phosphorylation. Interestingly, plasma membrane permeabilization by 20 µM digitonin in Ca^{2+} containing medium led to results similar to Cramoll 1,4. Basal and uncoupled respiration of *T. cruzi* epimastigotes were not affected by Cramoll 1,4 plus Ca^{2+} treatment, but oligomycin poisoned respiration was 60% higher than the control. In conclusion, Cramoll 1,4 toxicity to *T. cruzi* seems to result from a concerted action on parasite plasma membrane, mitochondrial Ca^{2+} overload and ROS production.

Keywords: Cell death, lectin, plasma membrane permeabilization, *Trypanosoma cruzi*.
Supported by: CAPES, CNPq and FAPESP.