

ANALYSIS OF PROTEIN EXPRESSION OF *TRYPANOSOMA CRUZI* ZYMODEME 3

Kikuchi,S¹, Sodr ,C¹, Chapeaurouge D³, Perales,J³, Mendonça,L² and Fernandes,O¹.

¹Departamento de Medicina Tropical-IOC, Fiocruz;²Departamento de Bioqu mica e Biologia Molecular, ³Departamento de Fisiologia e Farmacodin mica – IOC, Fiocruz.

Studies based on enzyme electrophoresis profile clustered *T.cruzi* isolates into 3 zymodemes:Z1, Z2 and Z3.Ecological and molecular data suggest that Z3 are more related to *T. cruzi* I than *T. cruzi* II, but their exact phylogeny is an unresolved issue. A phenetic dendrogram based on RFLP-ITS-rDNA divided Z3 into two subgroups:Z3A and Z3B.Proteomic analysis of Z3 strains could contribute to elucidate this question. This work describes standardized protocols to obtain reproducible extraction and separation of soluble proteins from epimastigotes using 2D electrophoresis from 3663-Z3A and 4167-Z3B strains. Epimastigotes were lysed by freezing/thawing and soluble proteins were precipitated by TCA. The pellet was resuspended in Urea, CHAPS, DTT and ampholytes. Experiments carried out in pH 3-10,revealed a great diversity of spots between 3663 and 4167.Separation using pH range 4-7 showed an increase in number of focused spots. 3663 and 4167 strains presented 300 and 325 spots respectively, and 181 were common to both. A considerable diversity in protein expression evidenced by difference in the intensity of several spots between both strains was encountered. Proteins identified by mass spectrometry were grouped in metabolism, cellular organization and protein destination functions. These results can be the starting point to the understanding of the parasite biology and to the elucidation of the correct Z3 phylogenetic position.

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