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ePoster Viewing

Vaccine development

**IMMUNIZATION WITH ENOLASE OF TRYPANOSOMA CRUZI CONFERS PROTECTIVE IMMUNITY AGAINST ACUTE PHASE OF CHAGAS DISEASE IN MICE.**

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**Objective:** Determine the humoral and cellular immune response generated by the immunization with the *T. cruzi* enolase in a murine model during the experimental acute phase of Chagas disease.

**Methods:** The *T. cruzi* enolase gen was obtained from cDNA by RT-PCR, the PCR product was cloned into pCR™2.1-TOPO® Vector and afterwards was sub-cloned into pRSETB plasmid to obtain pRSETB::TcENO. The recombinant protein (rTcENO) was obtained by nickel affinity chromatography. BALB/c female mice were immunized with rTcENO with Freund adjuvant (complete and incomplete), each mouse received four doses intraperitoneally every seven days. A second control group received only PBS. Preimmune and immune sera were obtained. The isotype of antibodies was done by ELISA. After the immunization, mice were infected with  $8 \times 10^4$  blood trypomastigotes; then were bled every three days and the parasitemia was determinate by direct observation in an optical microscope. The survival was monitored daily. Cytokines were analyzed by flow cytometry during the peak of parasitemia. Finally, hearts were isolated aseptically, rinsed with steril PBS, and fixed for 24 h in 4% paraformaldehyde in PBS (pH 7.4). Fixed hearts were embedded in paraffin, sectioned (5µm), stained with hematoxylin & eosin, and examined by light microscopy.

**Results:** Mice vaccinated with rTcENO were able to generate specific antibodies (IgG1, IgG2a and IgG2b) typical for Th1/Th2 immune response. Furthermore, the group vaccinated with rTcENO showed 75 percent of survival with respect to control group. The parasitemia burden was reduced in 69.8% in vaccinated mice with rTcENO. At the end of parasitemia an increase of antibodies IgG1, IgG2a and IgG2b but not IgG3 was observed with respect to control. Meanwhile, the cytokines generated by immunization with rTcENO and after parasite challenge were IFN- gamma and IL-2, showed that a type Th1 immune response was polarized. Furthermore, hearts from vaccinated mice showed minimal inflammatory response compared with control mice (non- immunized and infected) that demonstrated abundant inflammatory cells and amastigote nests

**Conclusions:** On the basis of the above results, we found that the immune response generated by immunization with rTcENO after the parasite challenge, favoring a reduction in parasitemia and increase the survival of immunized and infected mice, demonstrating the feasibility of using the enolase as a potential vaccine in Chagas disease.