

Expression and Purification of Recombinant Protein of the *Toxocara canis*

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Introduction: *T. canis* and *T. cati* are gastrointestinal nematodes of the genus *Toxocara spp.* which complete their life cycle in the digestive tract of dogs and cats, and in paratenic hosts, including man, this cycle cannot be completed and the larvae remain in the host, passing between organs and systems. *Toxocara spp.*, infection triggers immune responses from the innate immune system, activating the adaptive system and consequently producing Th2 cytokines that cause eosinophilia and an increase in IgE. Currently, toxocariasis is diagnosed using the larval excretory-secretory antigens of this nematode (TES). Still, the cross-reactivity of antibodies (IgG or IgG4) with antigens from other helminths can lead to a false positive diagnosis. **Objective:** To evaluate the reactivity of the rFULL protein, a *T. canis* specific fusion protein (TES-26 and CTL-4), in the immunodiagnostic Toxocariasis. **Methods:** A synthetic plasmid containing the FULL sequence was transformed into different strains of *E. coli*, and expression of the heterologous protein was induced with IPTG for 4 hours. The bacterial extracts were solubilized in a pH 9 buffer solution and purified using affinity chromatography. Western Blot confirmed the expression and purification of the protein. Finally, the reactivity of the protein was evaluated with serum from patients using indirect ELISA. **Results and Conclusions:** The rFULL protein was constructed with all the probable B-cell epitopes found through the genetic sequences of two molecules (rTES-26 and rCTL4), resulting in a 38 kDa protein. The expression of the protein was observed and confirmed by western blotting after 4 hours of expression in the bacterial culture, and affinity purification was also carried out using the histag inserted at the C-terminal end of the protein. Through indirect ELISA, it was possible to confirm the immunogenic potential of the rFULL protein, with sensitivity of 71% and specificity of 87% to IgG, and respectively 62% and 91% para IgG4 when compared to TES.

Keywords: Toxocariasis, Immunodiagnosis, recombinant proteins, *Toxocara spp.*
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