Introduction and objectives: *Rhipicephalus microplus* is a cattle hematophagous ectoparasite related to economic losses in livestock. Elucidating tick anti-blood clotting agents contributes to understand host-parasite relationship, thereby opening new prospects to find targets for tick control as well as molecules with therapeutic potential for some blood disorders. This work aims to determine the amino acid sequence of a *R. microplus* midgut thrombin inhibitor (BmGTI); to clone its ORF sequence and to express the protein in a heterologous system.

Materials and Methods: To purify BmGTI, tick midgut was dissected and its homogenate was processed by ion exchange (HiTrap Q HP), size exclusion (Superose 12) and thrombin-affinity chromatographies, being analyzed by SDS-PAGE 14%. Thrombin inhibition was accessed using a fibrin-clotting assay. Purified BmGTI was digested by trypsin and analyzed by mass spectrometry. Available *R. microplus* database was used to identify the putative BmGTI sequence. As well, RNA was extracted from midgut, and cDNA was used to amplify BmGTI ORF to clone it in pGEM-T plasmid. Results and Conclusions: Purification procedures allowed obtaining a homogeneous preparation of BmGTI. Based on the mass of peptides from purified BmGTI, its correspondent cDNA sequence was identified in *R. microplus* INCT-EM database as a putative secreted protein. The comparison with protein database showed that the coding sequence has thioredoxin-like and ERp29c-like domains, and a putative signal peptide sequence. Also, there is 79% and 88% identity with a putative secreted protein from *Ixodes scapularis* and a BTSP from *Amblyomma variegatum*, respectively. The ORF was amplified by PCR and an amplicon with 732 pb was obtained. The identity of this amplicon cloned in pGEM-T was confirmed by DNA sequencing. Work is going on in order to express BmGTI in a heterologous system, characterize the inhibitory properties, and the immunoprotective potential of this protein.

Keywords: inhibitor, *R. microplus*, thrombin

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