Cloning and Expression of an Ovary Protein from *Riphicephalus (Boophilus) microplus*

Oldiges, D. P.[1,4], Parizi L. F.[1], Seixas, A.[1], Lorenzini, D. M.[1], Da Silva Vaz Jr, I.[1,3], Termignoni, C.[1,2]

[1]Centro de Biotecnologia; [2]Departamento de Bioquímica; [3]Faculdade Veterinária - UFRGS; [4]UFCSPA

The tick *Rhipicephalus microplus* causes several losses in cattle breeding. The use of anti-tick vaccines has proved to be a promising alternative strategy to substitute acaricides. The success of this strategy depends on the characterization of molecules with critical physiological roles in the tick. Vitellin Degrading Cysteine Endopeptidase (VTDCE) is an enzyme involved in vitellin degradation during embryogenesis and larval stages. When the native protein was inoculated into bovines, it induces an humoral response partially protective against *R microplus*. The protein was sequenced by mass spectrometry using LCMS/MS. Based on the identified peptides we found a corresponding expressed sequence tag (EST) in a cDNA bank of *R. microplus*. The objective of the present study was to clone and express a recombinant VTDCE. By RT-PCR, the coding region of VTDCE was amplified from *R. microplus* ovary RNA. The amplicon was ligated into the plasmid vector. *Escherichia coli* was transformed with the plasmid and correct cloning was confirmed by PCR and DNA sequencing. The 285-bp fragment was subcloned in pET32a expression vector, and *E. coli* strain BL21 Star (DE3) was transformed with the plasmid. The expression was obtained after 2 hours of incubation at 37 °C with 1mM of IPTG. By western blot, the sera of bovines and rabbit immunized with native VTDEC recognized the recombinant VTDCE in *E. coli* extracts, confirming the identity of the recombinant protein. The immunogenic and immunoprotector potentials of the recombinant protein are under study.

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