THE *SCHISTOSOMA MANSONI* CATHEPSIN D-LIKE 3 GENE (SMCD3)

Vidal, L.E.L.[1], *Lanzarini, N.M.*[1], *Avelar, L.G.A.*[2], *Oliveira, G.C.*[2], *De Simone, S.G.*[1], *& Silva-Jr, F.P.*[1]

[1]*Laboratorio de Bioquímica de Proteínas e Peptídeos, IOC-FIOCRUZ;*
[2]*Laboratório de Parasitologia Molecular e Celular, CPqRR*-FIOCRUZ.

Earlier bioinformatics analysis of the *Schistosoma mansoni* genomic and cDNA sequences identified a family of aspartyl proteases (APs) in this parasite (here called SmAPs). One of these enzymes (SmCD1) was previously demonstrated to participate in the digestion of host hemoglobin, providing essential nutrients for the survival of *S. mansoni*. Thus, we believe that the study of SmAPs will, in the future, provide new pharmaceutical targets for schistosomiasis. The objective of this work was to clone a novel SmAP gene, SmCD3, to enable functional and structural studies. From RT-PCR of *S. mansoni* adult worms total RNA, it was possible to clone the SmCD3 gene. Sequencing of ten individual pGEM-T clones carrying the complete coding region of the SmCD3 gene disclosed several single nucleotide polymorphisms in comparison with a reference genomic sequence. One of the SmCD3 variants was chosen for subcloning in the pTBSG vector and expression in *Escherichia coli* to allow for the immunization of rabbits to obtain a polyclonal antiserum. An analysis of the expression of the SmCD3 gene as well as the other known SmAP genes, SmCD1 and SmCD2, was carried out by real-time PCR of cDNA samples from the schistosomula, cercariae, sporocysts and adult (male and female) *S. mansoni* life cycle stages. The miracidium stage was chosen as the calibrator and either SmTPI or SmActin genes were used as normalizers. The SmCD3 gene presented highest expression in the cercariae stage, presenting a 120-fold change in expression level when compared to the adult male stage. A preliminary phylogenetic analysis indicates that the SmCD3 gene is paralogous to the other known SmCD proteases.

Keywords: ASPARTYL PROTEASES, DRUG TARGET, PROTEIN EXPRESSION, *SCHISTOSOMA MANSONI*

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