Production of a recombinant *Cryptosporidium parvum* 12kDa protein in *Escherichia coli* and development of specific antibodies

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Abstract

**Introduction:** Parasite recombinant proteins are gaining wide-spread attention due to its diagnostic application. This work aims at the evaluation of a novel fusion system for the production of a 12 kDa surface adherence protein from *Cryptosporidium parvum*, which has been reported as a potential target for cryptosporidiosis prevention and therapy [Yao et al, 2006]. It is also intended to obtain specific antibodies and to study its ability to recognize the surface of sporozoites and oocysts.

**Methods:** Specific primers were designed to amplify and sub-clone part of the gene that encodes for *Cryptosporidium parvum* 12kDa protein lacking its transmembrane domain. This truncated protein was expressed in *E. coli* using a novel fusion system that significantly increases protein production yields. After its purification by affinity chromatography on Ni-NTA columns, the protein was dialysed against phosphate buffer pH 8.0 and later injected in mice. The immune response obtained in mice was then evaluated by ELISA and immunofluorescence assays.

**Results:** The expression levels of fused protein in *E. coli* increased about three folds when compared to the respective non-fused protein levels. Results from ELISA and immunofluorescence assays confirmed the potential diagnostic interest of this recombinant antigen.

**Conclusions:** Results from this work may provide an important advance on diagnostic and therapeutic field on cryptosporidiosis.