

Immunogenicity studies of recombinant multi-cystatin-like domain protein from *Trichinella britovi* produced in yeast

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Trichinellosis is a zoonotic parasitic disease caused by nematode worms of the genus *Trichinella*, which shows a world-wide distribution. The infection is acquired by the consumption of raw or undercooked meat and meat products harbouring nematode larvae. *T. spiralis* is considered as an etiological agent of most human infections and deaths caused by trichinellosis globally. However, every year the outbreaks of human trichinellosis caused by other encapsulating and nonencapsulating species have been reported. From several species of *Trichinella*, that occur in European area, *T. britovi* is one of the most common species, that may affect human health, but relatively poorly investigated. Identification of *Trichinella* proteins is very important in elucidating host-parasite interactions, and is a key factor which will help in the development of immunological detection methods and vaccines. Therefore the broad study of *T. britovi* antigens and its immunological properties are needed. In our previous study we used immunoproteomic approach and identified multi-cystatin-like domain protein (CLP) as an immunologically active protein from *T. britovi* muscle larvae, recognized by sera from infected pigs. This protein was selected for cloning and expression in *Pichia pastoris* yeast system, which is able to carry out post-translational modifications and efficiently secrete protein of choice into the culture medium. Then, obtained 47 kDa recombinant protein was used for immunization of mice. Immunological potential of rCLP was analyzed by detection of IgG, IgG1, IgG2a antibodies in serum and selected cytokines (IFN γ , IL-2, IL-4, IL-10), secreted by stimulated splenocytes isolated from immunized and/or infected animals. Furthermore, muscle larvae burden was measured at the end of experiment (48 days post infection), as an indicator of potential protective role of rCLP.

Results indicate that *T. britovi* rCLP elicited high titer antibodies level, visibly affected the profile of cytokines secreted by stimulated splenocytes, in comparison to naïve animals, and reduced the number muscle larvae. It makes rCLP promising candidate for subsequent studies and future usage in immunodetection and vaccination against *T. britovi* parasites.

Financial support for this study was provided by the National Science Centre Poland (grant UMO-2015/18/E/NZ6/00502).