ABSTRACT

This study is focused on cercarial cysteine peptidases of the trematode *Diplostomum pseudospathaceum*. It follows previous research which confirmed the presence of a 24kDa cysteine peptidase in cercariae biochemically and by mass spectrometry. It was postulated, that the function of this peptidase is histolytic, when cercariae penetrate the tissues.

During an attempt to purify this peptidase and characterize its peptidolytic activity, it was found out that the cercarial homogenate containsmore different peptidases varying in their pI. Tests of peptidolytic activity and inhibition have shown that these peptidases are cathepsin L-like. They are active over a broad spectrum of pH with optima of activities in weakly acidicor neutral pH.

Using degenerate primers based on conserved motifs of cysteine pepridases, partial sequences of three genes for cathepsin L of *D. pseudospataceum* (DpCL1, 2 a 3) were obtained. Then the complete sequences of DpCL2 and 3 genes and partial sequence (without 5´end) of DpCL1 were obtained by RACE PCR.

To confirm function of these peptidases we tried to immunolocalize them. We assumed that they are localized in penetration glands. Preliminary results suggested that some of the cathepsins could be also localized in the gut of cercariae.

For more detailed biochemical characterization and immunolocalization the recombinant cathepsin DpCL3, which is probably the most expressed one by cercariae, has been produced.

Key words:

trematode, cercaria, *Diplostomum*, protease, peptidase, penetration, cathepsin L