Abstract

Ticks are globally important parasites involved in transmission of a wide variety of infectious agents. The most common tick species found in Europe is the hard tick *Ixodes ricinus*, which transmits bacterium *Borrelia burgdorferi* (a causative agent of Lyme disease) or tick-borne encephalitis virus. Cathepsin proteases are important in the process of digestion of blood proteins in the tick gut. This work is focused on cathepsin L, an important digestive cysteine protease of ticks.

Recombinant *I. ricinus* cathepsin L was expressed in *Pichia pastoris* and separated from the culture medium by chromatographic purification. N-terminal protein sequencing and labeling by activity-based probe Green-DCG-04 were used for characterization of purified cathepsin L. Substrate and inhibitor specificity were analyzed using peptide substrates and inhibitors. This analysis showed that Z-FR-AMC is a suitable substrate with pH optimum 3.5, and that Z-FF-DMK is an efficient inhibitor. It was demonstrated that cathepsin L cleaves protein substrates in strongly acidic environment (pH 3.5-4.5).

Cathepsin L-like proteolytic activity was demonstrated in salivary gland extract and in saliva of the *I. ricinus* tick. The presence of a cathepsin protease in tick saliva is reported here for the first time. This finding suggests that cathepsin L is secreted into the saliva and can be involved in tick-host interactions.

Key words: cysteine proteases, cathepsin L, tick, *I. ricinus*, substrate and inhibitor specifity, proteomic activity-based labeling

(In Czech)