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ACCEPTED MANUSCRIPT

Leptospira interrogans thermolysin refolded at high pressure and alkaline pH displays proteolytic activity against Complement C3

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Highlights

- HHP at alkaline pH induces non-denaturing solubilization of thermolysin IB
- Thermolysin is refolded at high yield by dialysis to pH 8.0
- SEC-purified thermolysin suffers autoproteolysis and became enzymatically active
- Recombinant thermolysin cleaves C3 in a dose- and time-dependent fashion.

Abstract

Enzymes from the thermolysin family are crucial factors in the pathogenesis of several diseases caused by bacteria and are potential targets for therapeutic interventions. Thermolysin encoded by the gene LIC13322 of the causative agent of leptospirosis, *Leptospira interrogans*, was shown to cleave proteins from the Complement System. However, the production of this recombinant protein using traditional refolding processes with high levels of denaturing reagents for

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