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Partial Characterization of Proteases from Culture Filtrate of *Mycobacterium tuberculosis*

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Two proteases were partially characterized from culture filtrate of *Mycobacterium tuberculosis* KIT110. Their molecular weights were approximately 200 and 180 kDa, respectively and they exhibited similar enzymatic characteristics. These enzymes were inhibited significantly by EDTA and to some extent by EGTA. Their activity were enhanced by Ca^{2+} and Mg^{2+} to some degree. However, Cu^{2+} and Ag^{2+} completely inhibited the enzyme activity at the concentration of 2.5 and 5 mM, respectively. The optimal pH was 7.0 and optimal temperature was around 40℃. These enzymes were rapidly inactivated at 80℃. Therefore, they were heat-labile, neutral metalloproteases. These enzymes exhibited antigenicity reacting with serum from the patients with pulmonary tuberculosis. These enzymes were able to degrade serum proteins including hemoglobin, bovine serum albumin, lysozyme and immunoglobulin G and structural matrix protein such as collagen type I. Therefore, these enzymes may contribute to tissue necrosis and pathogenesis during infection.

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Cytopathogenicity of Proteinases from Excretory and Secretory Products of *Acanthamoeba culbertsoni* to HEp-2 Cell

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Virulent protozoan's proteinases are important in tissue invasion, migration and host pathology. We have characterized the five proteinases(P1, P2, P3, P4, and P5) from excretory and secretory products(ESP) of *Acanthamoeba culbertsoni* previously. P1, P2, P3, and P4 were neutral serine proteinases. P5 was acidic aspartic proteinase. These proteinases degraded collagen(type I), BSA and rabbit corneal extracts. In this study, we showed that excretory and secretory products of *A. culbertsoni* exhibited cytopathogenicity to HEp-2 cell and represented more directly the fact that proteinases in excretory and secretory products of *A. culbertsoni* were associated with its pathogenesis during infection. When the excretory and secretory products which was treated with PMSF added to HEp-2 cell, the cytophatogenicity was not appeared. Therefore, these suggested that the proteinases of *A. culbertsoni* play an important role in its pathogenesis.