

A COMBINED THERMODYNAMIC AND CRYSTALLOGRAPHIC STUDIE OF THE INTERACTION BETWEEN BOVINE BETA-TRYPSIN AND PENTAMIDINE

PERILO, C.S.,¹ NAGEM, R.A.P.,¹ PEREIRA, M.T.,¹ SANTORO, M.M.¹.

¹Laboratório de Enzimologia e Físico-Química de Proteínas – Instituto de Ciências Biológicas – Universidade Federal de Minas Gerais.

Serine proteinases play a central role in several physiologic processes. Trypsin is a model system for serine proteinases. Since proteolytic reactions control many biological processes, discovery of new and stronger inhibitors of proteolysis is an important field of modern chemistry. Kinetic studies have shown that pentamidine is a strong trypsin inhibitor but little is known about this interaction. The objective of the present study is to solve the structure of the complex between beta-trypsin and pentamidine using X-ray crystallography and determine the thermodynamic parameters of the interaction by using ITC. Bovine beta-trypsin was purified by chromatography. Crystals were grown at 18 °C by the hanging drop vapor diffusion method, using ammonium sulfate and PEG 3350 as precipitants. Diffraction data were collected at the LNLS in Campinas, São Paulo. Analyses of electron density map for the complex revealed the presence of the ligand in the catalytic cleft of the enzyme. Refinement of the structure is being conducted at the ICB at the UFMG. ITC results show that trypsin has potentially two different binding sites for pentamidine, each one with a different affinity.

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