The active site thiol group is oxidised, and inhibitors is feasible. The active site thiol group is oxidised, and inhibitors insertion anymore. Although aspartic proteinases are widely dispersed in the proteolysis can be activated by high, non-physiological concentrations of calcium. In the absence of cysteine proteases.

While cathepsin L is biosynthesised as a pre-pro form of 333 residues and loses a signal sequence to give a 316 residue pro-form, which is cleaved at low pH to yield a 219 residue form, the tradi- m·e aspart in the structure of factor XIII bound to the stmcture of factor XIII bound to the stmcture of factor XIII bound to cardosine A, isolated from the cardo flover (1), which is traditionally used in Portugal for cheese making, was crystalized and its structure solved by the molecular replacement method, using the structure of human cathepsin D (2) as a model. Cardosine A is composed of two polypeptide chains and is expressed with the ca. 100 residues insertion, also found in other vegetable aspartic proteinases. However, its mature native form does not show the insertion anymore.

Cysts were obtained in space group C2, a=119.0, b=88.1, c=82.3 Å, β=104.4, with two molecules in the asymmetric unit. Synchrotron diffraction data to 2.9 E resolution were collected at the EMBL outstation, Hamburg. Refinement is in progress, with an actural of R=29.4% and Rfree=35.0%.


PS04.01.36 THE 2.5Å CRYSTAL STRUCTURE OF PENICILLIN G ACYLASE FROM A MUTANT FORM OF P. RETGERI, Michael A. McDonough1, Herbert E. Klei2, Gayle K. Schulte3, and Judith A. Kelly1, Departments of Molecular and Cell Biology, and Institute of Materials Science, University of Connecticut, Storrs, CT 06269-3125, Bristol-Myers Squibb, Princeton, NJ 08543-4000, Pfizer Central Research, Groton, CT 06340

Penicillin G acylase (EC 3.5.1.11) catalyzes the hydrolysis of the inexpensive antibiotic penicillin G into phenylacetic acid (PAA) and 6-aminopenicillanic acid (6-APA). The 6-APA is used commercially in the synthesis of new penicillins with broader inhibitory profiles. A mutant form of the acylase from P. retgeri with an altered substrate profile was crystalized in space group P6_12 (a=b=1.405 Å, c=202.8 Å) from 50mM potassium phosphate, pH 7.5, 5 percent ammonium sulfate and 10 to 15 percent v/v glycerol (Klei, H. E. et al., 1995, Protein Science 4, 433-441). Native and heavy atom data have been collected to 2.5Å. The native data are 96 percent complete, and the Rsym is 11 percent on I. Data were collected at 100K. The structure has been solved by molecular replacement using AMoRe with the E. coli penicillin acylase as a model(Duggleby, H. J. et al., 1995, Nature 373, 264-268). The P. retgeri enzyme is a heterodimer with a 23.7kDa alpha subunit, and a 62.2 kDa beta subunit with the reactive serine residue being the first residue of the beta chain. All but ten resi-dues at the carboxyl terminus of the alpha chain have been modelled, and the native structure is being refined using X-PLOR. Currently, the R factor is 0.24, and the rms deviations in bond distances and angles are 0.011Å and 1.6° respectively. Addition of solvent molecules to the model is in progress.

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