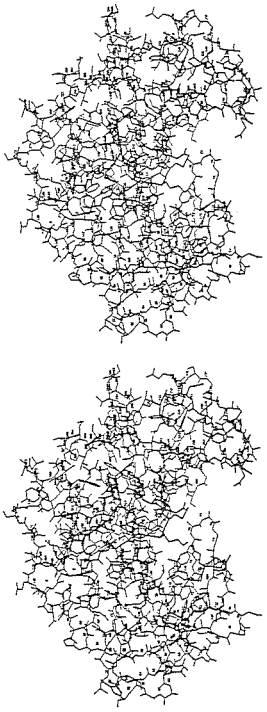

The structure of endothia pepsin

02.1-27 CRYSTAL STRUCTURE OF PORCINE PEPSINOGEN. By S.N. Rao, S.N. Koszelak and J.A. Hartsuck, Oklahoma Medical Research Foundation, Oklahoma, OK 73104

Porcine pepsinogen, molecular weight 39,630, crystallizes from lithium sulfate solutions of pH 6.0 in space group C2 with a=104.8, b=43.1, c=88.4 Å and β=91.4°. Diffraction data have been collected by diffractometry to 5.4 Å resolution for the native crystal and for a K_2PtCl_4 derivative. Molecular replacement techniques have placed a model in the pepsinogen crystal space. The model was derived from the penicillopepsin atomic coordinates and corresponds to 64% of the pepsinogen molecule. Translation parameters were determined by a Tl translation function and were confirmed by packing analysis. For the platinum derivative, five sites were identified by direct methods on normalized structure factor differences. A difference Fourier map phased by molecular replacement yielded the same five platinum locations. Phasing of the x-ray data was achieved by a combination of the isomorphous replacement and molecular replacement information.

Pepsinogen differs from pepsin and the other active acid proteases in that it has 44 more residues at the amino terminal end of the protease chain. This activation peptide is spontaneously removed when pepsinogen is exposed to low pH. Peaks in the pepsinogen map which are not due to the model can be attributed to the activation peptide. Our crystal structure shows that the peptide wraps around the pepsin molecule and contains little or no secondary structure. It blocks access to the pepsin active site even though it is not located in the active site cleft. A conformation change to place the activation peptide in contact with the pepsin active site must occur as activation takes place.

Photographic data at 2.5 Å resolution are being collected and results of those experiments will be discussed.

02.1-28 A STRUCTURAL STUDY OF THE ELASTASE-ELASTATINAL COMPLEX AT 2.5Å RESOLUTION. L. Presta, G. Cole, and E. Meyer, Biographics Laboratory, Department of Biochem. and Biophys., Texas A&M University, College Station, Texas 77843 U.S.A.

Elastase is a serine protease possessing an extended substrate binding site. An understanding of the energetics of recognition and binding may be sought only after resolution of the binding interaction into discrete components (H-bonds, hydrophobic associations, etc.). A structural study of elastase + substrate would help illuminate the several structural aspects of these interactions.

Elastatinal is a naturally occurring, highly specific inhibitor of porcine and human elastase, resembling a tetrapeptide. The hemiacetal complex formed with Ser 195 at the active site gives a geometry which should closely resemble the tetrahedral intermediate involved in normal substrate hydrolysis. Thus, the structure of the elastase + elastatinal complex may indicate the conformations and interations involved in extended substrate binding, which could be useful to those who seek to design inhibitors of disease-related elastases.

A set of 11000 diffractometer data was collected from a crystal soaked first in elastatinal (pH5), then in His buffer(pH7). Initial phases were derived from our 1.7A elastase + methanol structure. Refinement using EREF (Jack-Levitt-Deisenhofer) is in progress. Results will be presented in pictorial form.

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02.1-29 CRYSTAL STRUCTURE OF STREPTOMYCES ERYTHRAEUS LYSOZYME. By R. Sarma, S. Harada, T. Ikenaka and S. Hara, Department of Biochemistry, State University of New York, Stony Brook, N.Y. 11794, U.S.A. and Department of Chemistry, Osaka University, Toyonaka, Osaka 560, Japan.

Streptomyces erythraeus lysozyme found in its culture broth differs from other mammalian lysozymes in its molecular weight, amino acid composition and specificity. The enzyme has about 185 residues in its primary structure and has a molecular weight of about 20,000 daltons.

The three-dimensional structure of the bacterial lysozyme has been determined using x-ray crystallographic data to 2.9A resolution and refined using 2.5A resolution. The tertiary structure is different from that of hen egg white lysozyme or the T4-phage lysozyme.

The primary structure of the bacterial lysozyme is only partially determined, it resembles the primary structure of lysozyme from the fungus chalaropsis. This similarity has been used to speculate on the structure of the active site of the enzyme.