PS-03.07.05 STRUCTURE COMPARISON BETWEEN TRICHO- 
SAMTHIN AND MOMORDICIN. By Gao Ben*, Wang Yongping, 
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The similarities and differences between the two RIPs (ribosome inactivating proteins) molecular structures were determined and analyzed on the basis of the refined structure models of Trichosanthin at 1.7Å resolution (Gao Ben, et al., Science Sinica B, 1993, 4, in press) and α-Momordicinin at 2.0 Å resolution provided by Ben Jinghong and Wang Yangping (Private communication). The superposition of the two models was performed using the main-chain atoms and the RMS deviation for all the main-chain atoms of the 246 residues is 0.8 Å. Dividing the two models into many pairs of different polypeptide fragments, the superpositions of the pairs of different polypeptide fragments have been done in turn and the RMS deviations for the main-chain atoms of four fifth polypeptide fragments are smaller than 0.2 Å. The RMS deviations for the side-chain atoms of more than half of all residues are smaller than 0.5 Å and the great majority of these residues form six hydrophilic cores in the interior of the two proteins. These results indicate that the backbones of the two proteins have very similar three-dimensional arrangements. There are three domains of the greatest deviation for the main-chain conformations of the two proteins, A1-A2, 172-182, and 216-222, which are the flexible loops on the surfaces of the two proteins and corresponding to the sequence fragments with the greatest residue differences. Therefore, this result indicates that the residue differences have evidently brought about the three-dimensional conformational differences. There are the residue differences corresponding to primary amino acid sequences for one third residues of the two models. As the result of those residue differences, the differences were found to exist not only in the main-chain conformations but also evidently in the secondary structures and in distributions of the other hydrogen bonds relative to the main-chain atoms and booted waters which form hydrogen bonds to the main-chain atoms. The patterns of thirteen percent of the hydrogen bonds for the α helix of the two models, that of seventeen percent of the hydrogen bonds for the β sheets, and that of thirty-eight percent of the hydrogen bonds for the turns are different from each other, respectively. The patterns of thirty percent of the other hydrogen bonds relative to the main-chain atoms and thirty areas of the waters bound to the main-chain atoms are different, respectively. 

Ten highly conserved residues among primary amino acid sequences of 17 RIPs (Funatrus, C., et al., Ibechino, 1991, 73, 1157-1161, Gao Ben, et al., Science Sinica B, 1993, 4, in press) were analyzed and those corresponding to Trichosanthin are 14lys, 22arg, 25lys, 11lys, 12lys, 32leu, 106gly, 161ala, 163arg, and 197asp. Superposition of ten residues of Momordicinin on those corresponding to Trichosanthin was done together using the all-booted atoms of the main-chain. The RMS deviation for the main-chain atoms is 0.8 Å and that for the side-chain atoms and thirty areas of the waters bound to the main-chain atoms are different, respectively. 

Trichosanthin is a toxic protein (Mr 27000) used as a traditional Chinese drug for inducing abortion and recently found to be an anti-human immunodeficiency virus agent. Tri- 
chosanthin is type I ribosome-inactivating protein (RIP) with the activity of RNA N- 
glycosidase, and it was reported that ricin, a type-II RIP, catalyzes the cleavage of the N- 
glycosidic bond of a specific adenine within 28S rRNA, resulting in the inhibition of protein 
synthesis (Endo et al., J. Biol. Chem., 1991, 322, 1128-1130). We have determined at 3Å resolution the three-dimensional structure of trichosanthin crystallizing in monoclinic space group P2(1) (Xia et al., Chin. J. Chem., 1991, 9, 565-567) and it has been refined at 2.0 Å resolution (Xia et al., Abstracts of 6th FABCON Congress, 1992, 41-42, 99). The molecule shows a cleft near the interface of the two domains and the cleft is likely to be the active site region in which several absolutely conserved residues are located.

The complex of trichosanthin with nicotinamide adenine dinucleotide phosphate (NADPH), a substrate analogue, was prepared and crystallized in space group P2(1) with unit cell dimen- 
sions a=38.3Å, b=76.8Å and c=79.9Å, similar to orthorhombic native crystals. The diffraction data up to 1.7Å resolution were collected on an X-003 X-ray area detector. The three- 
dimensional structure of the complex has been solved by molecular replacement method (program MERCKOT) using one molecule of the mono- 
clinic trichosanthin structure as the search 
molecule. The complex structure was refined at 1.7Å resolution, using program PROFFIT, in 
which 170 bonded water molecules were included but NADPH was absent in the model, giving an 
R-factor of 18.9% in the resolution range 0. 

2.0Å with the rms deviation of 0.02Å from 
ideal bond lengths. The resulting (2Fo-Fc) map shows excellent electron density for the protein and an additional piece of continuous electron density. The NADPH has been fitted into it with the adenine ring in the strong and flat electron density which is located between the aromatic rings of Try70 and 
Try71. The adenine interacts with Arg161 which is absolutely conserved and located in the 
depth center of the cleft, Ser159 which is conserved in some of the RIPs, and the main- 
chain of the protein. The phosphate at the po- 
tion 0' of the ribose of the adenine interacts with several conserved residues in the 

cleft and is important for stabilizing the complex, as shown by the fluorescence 
spectra. The further refinement with NADPH 
present in the model is in progress.
03-Crystallography of Biological Macromolecules

PS-03.07.09 STRUCTURE OF OLIQU-1,6-GLUCOSIDASE REPLICATED AT 2.3Å RESOLUTION: A STRONG CORRELATION BETWEEN THEIR PROTEIN LADDER AND THERMOSTABILITY

In order to elucidate the contribution of protein to their thermality, we have analyzed the structure of oligo-1,6-glucosidase (OGL) and its thermophilic homologs. The enzyme function enzima was assessed by examining its thermal stability, pH sensitivity, and stability. The structure showed a strong correlation between their protein ladders and thermostability.

![Structure of OGL](image1)

**PS-03.07.11 STRUCTURE ANALYSIS OF PHOSPHOLIPASE A2 FROM VENOM OF TRIMERUS PLATYCEPHALUS**

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The structure analysis was conducted with the molecular replacement method using the modified structure of PLAP2 from the Western Diamondback rattlesnake. The structure model was refined with the program X-PLOR and the resolution extended to 2.1 Å. The α-helix and β-strand motifs were distinguished into three domains. The α-terminal domain has (β/α)− barrel with four additional α-helices. The β-strand domain is a three-strand β-sheet, and the α-helix motif is a four-strand β-sheet. The structure of PLAP2 is similar to those of other α-helical enzymes and can be subdivided into three domains. The structure was determined at 2.1 Å resolution.

![Structure of PLAP2](image2)

**PS-03.07.10 CRYSTAL STRUCTURE OF ALKALINE SERINE PROTEASE OF BACTERIAL ORIGIN**


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The crystal structure of alkaline serine protease from a Bacillus strain was solved at 2.6 Å resolution. The enzyme is homologous to the Bacillus cereus protease and can be classified as a serine protease.

![Crystal Structure](image3)