02.1-15 THE THREE-DIMENSIONAL STRUCTURE OF TRICHOSANTHIN. By Pan Kezhen, Lin Yujuan, Fu Zhuji, Zhou Kangjing, Cai Zhaoping, Chen Zhiwei and Zhang Yongmao, Fujian Institute of Research on the Structure of Matter, Academia Sinica, Fuzhou, China, Dong Yicheng, Wu Shen, Ma Xingqi, Wang Yaoping, Chen Shizhi, Wang Jiahuai and Zhang Xuejun, Institute of Biophysics, Academia Sinica, Beijing, China, Ni Chaozhou, Zhang Zhiming, Xia Zongxiang, Fan Zhaouchang and Tian Gengyuan, Shanghai Institute of Organic Chemistry, Academia Sinica, Shanghai, China. (The authors are listed by alphabetical order of their institutes).

In previous work on the structure of trichosanthin at 4Å resolution we reported the location of the molecular boundary and the tracing of the polypeptide chain (Pan Kezhen et al., *Scientia Sinica* B25 1982, 730; B29 1986, 26). In the present work the resolution has been extended to 3Å, then further to 2.6Å. A model for each of the two molecules in the asymmetric unit of the crystal has been constructed. Each molecule is made up of two domains. The larger domain contains 169 residues starting from the N-terminus and the smaller domain, of 65 residues, includes the C-terminus. The two domains are linked by a β-sheet formed by strands β_{13} near the C-terminus and strands β_2 and β_3 of the large domain. This forms an important non-covalent interaction between the domains. The molecule may be described as a core of eight α-helices surrounded by four β-sheets composed of thirteen strands. One helix (α_5) is almost completely buried in the center of the molecule. Two aspects of the secondary structure are particularly noteworthy. Firstly, at residue Gly 179 in helix α_7 the turn angle is 70°. Such a large turn angle is rarely seen in protein structures. Secondly, the β-sheet composed of the six strands β_1, β_4 ... β_8 shows a clear example of the typical left-handed twist.

Despite their overall similarity there are some differences between the two non-crystallographically related molecules. The most obvious are conformational changes in the C-terminal strand and in some irregular loops joining elements of secondary structure. The two trichosanthin molecules in the asymmetric unit interact with each other forming a boundary surface parallel to the *c*-axis. The interaction is formed principally by the large β-sheet (β_1, β_4 ... β_8) in molecule A and the β-sheet composed of β_2, β_3 and β_{13} (including the c-terminal strand) as well as the loops between strands β_3 and β_4 in molecule B. At the interface many residues are interdigitated forming salt bridges, hydrogen bonds and hydrophobic interactions.

02.1-16 CRYSTALLOGRAPHIC STUDY OF A MONOCLONAL Fab FRAGMENT SPECIFIC FOR HPr OF THE PHOSPHOENOLPYRUVATE: SUGAR PHOSPHOTRANSFERASE SYSTEM (PTS) OF E. COLI. L. Prasad, M. Vandonselaar, J.S. Lee and L.T.J. Delbaere, Department of Biochemistry, University of Saskatchewan. Saskatoon, Saskatchewan, CANADA, S7N OWO.

It is possible to study a specific protein-protein interaction by determining the structure of the interacting proteins. A monoclonal antibody was prepared against HPr, the histidine-containing protein of the PTS system of E. coli. Papain digestion of the purified IgG antibody gave a good yield of Fab fragments which were purified by DEAE-Sephacryl chromatography and isoelectric focusing. These proteins are suitable for the crystallographic study of protein recognition, since the structure of HPr has been solved, the Fab fragments form crystals suitable for diffraction, and the Fab:HPr complex is stable at neutral pH and also forms crystals. Crystals of the Fab fragment were grown from solutions containing 10-20 mg/ml Fab, 15% PEG 6000, 0.05M phosphate buffer (pH 8.0) and 0.2 M NaCl. The crystals are monoclinic $P2_1$, with a = 117.48, b = 66.56, c = 67.31Å and β = 118.71°, and Z = 4. Four isomorphous heavy-atom derivatives were obtained with sodium p-chloromercuribenzene sulfonate, dichloro-(ethylenediamine)platinum, potassium mercury iodide and uranyl nitrate. X-ray intensity data for the native protein and derivatives were collected using an Enraf-Nonius CAD4 diffractometer. Attempts are being made to solve the structure by the molecular replacement method, using the coordinates for the HED10 Fab fragment as model (Cygler et al., 1987), and also by the multiple isomorphous replacement method.
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02.1-17 CRYSTALLOGRAPHIC ANALYSIS OF THE CALCIUM-CALMODULIN/TRIFLUOPERAZINE COMPLEX. L.T.J. Delbaere, L.M.B. Gehrig, M. Vandonselaar, J.W. Quail, and R.A. Hickie, Departments of Biochemistry, Chemistry, and Pharmacology, University of Saskatchewan, Canada, S7N OWO.

Calmodulin is a widely distributed, acidic, heat-stable calcium-binding protein which activates many calcium-dependent enzymes and modulates various physiological processes. Cancer cells have abnormally high levels of calmodulin; this factor is believed to contribute to the unregulated growth of neoplastic cells. Trifluoperazine (TFP) binds to the active form of calmodulin and blocks the activation of calmodulin-dependent enzymes. In fact, TFP and other anti-calmodulin agents have been shown to inhibit the growth of neoplastic cells.

Crystals of the calmodulin-TFP complex were grown from solutions containing 10 mg/ml calmodulin, 8 mM TFP, 26% PEG 4000, 0.01M sodium cacodylate buffer pH 5.2, 20 mM $CaCl_2$ and 20 mM $MgCl_2$. The crystals are trigonal, space group $P3_121$ or $P3_221$ with unit cell dimensions a = b = 40.73(3)Å, c = 179.2(2)Å. Heavy-atom derivatives were prepared with phenylmercuric acetate, mersalyl and dichloro(ethylenediamine)platinum. Intensity data from native and derivative crystals were collected and analysis of 2.8Å data is currently in progress. The three-dimensional structure of our trigonal crystal form will provide the tertiary structure of calmodulin and the specific interactions with an antipsychotic drug that inactivates this important protein.
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