02.2-2 A 3D MODEL OF PHOSPHORIBOSYL TRANSFERASE by B.Busetta and M.Hospital, Laboratoire de Cristallographie UA 144 CNRS - Université de Bordeaux I - 33405 Talence Cedex, France.

The sequences of seven phosphoribosyl transferases were aligned using amino-acid homologies and secondary structure predictions. An average "folding pattern" [1] was computed for the transferase family and compared with the "folding patterns" of nucleotide binding proteins of known X-Ray structures:

- the NAD dehydrogenases (ie Lactate dehydrogese).
- the adenylate kinase.
- the GTP binding proteins (ie elongation factor Tu).

The folding pattern of the transferase family appeared related to the correspondent pattern of adenylate kinase and subsequent amino acid homologies were found with this protein. From this homology, we were able to build a 3D-model of phosphoribosyl transferase.

The analysis of conservative residues on this model allows to define the locations of both associated substrates and to propose a possible mechanism of the enzymatic reaction.

[1] BUSETTA,B.(1986),Biochem.Biophys.Acta 870,327-338.

02.4-1 CRYSTALLIZATION OF THE COMPLEX OF MOMODICA CHARANTIA L TRYPSIN INHIBITOR (MTI-1) WITH PORCINE TRYPSIN. By Huang Qichen, Chen Zhongguo, Li Genpei, Tang Youchi, Institute of Physical Chemistry, Peking University; Qian Ruiqing, Zeng Fuyao and Wang You, Shanghai Institute of Organic Chemistry, Academia Sinica.

Three trypsin inhibitors were isolated from Momodica charantia L seeds, their amino acid sequences were determined, and their protease-inhibiting properties were characterised. Among them, MTI-1 (MW 9000, 77 residues) is a Bowman-Birk type inhibitor, and we have succeeded in preparing rather promising crystals of MTI-1 and porcine trypsin complex for an X-ray structural study. A 'hanging drop' vapor diffusion method was adopted, and crystals usually appeared within two weeks with maximum dimensions of 0.8x0.8x0.3 mm. Precession photographs showed that the crystals belong to the trigonal system with the space group $P3_121$ (or $P3_221$) and unit cell dimensions a=b=62.6, c=124.3Å, and $V=4.2\times10^5$Å^3, and diffract X-ray to a resolution better than 1.9Å. We have collected 2.0Å resolution on a Huber Rotation Camera using graphite monochromatized CuKα radiation, and 3.0Å resolution data on a Rigaku automatic diffractometer (AFC-5). Further studies of molecular replacement method (rotation and translation function) and energy refinement are in progress.

02.4-2 CRYSTAL STRUCTURE DETERMINATION AND MOLECULAR MODEL OF MUNG BEAN TRYPSIN INHIBITOR LYS ACTIVE FRAGMENT-BOVINE TRYPSIN (MBILF-BTRY) COMPLEX AT 3.0 Å RESOLUTION. By Chen Zhongguo, Li Genpei, Zeng Jie, Tang Youchi, Lu Guangying, Wei Xincheng, Institute of Physical Chemistry and Department of Biology, Peking University; Lin Guangda, Zhang Rongguang, Xuan Jiancheng, Chi Zhengwu, Tsao Tienchin, Institute of Biochemistry, Academia Sinica, Shanghai, China.

The crystal structure of the MBILF-BTRY complex was determined by molecular replacement and its 3-D molecular model was thereby derived. Mung bean trypsin inhibitor belongs to the Bowman-Birk inhibitor group, which is by far the most complicated among the ten fundamental groups of serine protease inhibitors (Laskovski and Kato, 1980). The crystallographic data for the MBILF-BTRY complex are a=62.9, b=63.5, c=69.7 Å, space group $P2_12_12_1$ with Z=4. Its molecular weight is about 27500. On a Rigaku AFC-5 Four-circle diffractometer attached to a RU-300 rotating anode generator, 5142 independent reflections of up to 3Å resolution were collected. Relative orientation angles between the model BTRY and BTRY in our crystal, α=235°, β=46°, and γ=75° (expressed in terms of Crowther angles), were obtained by using Crowther's fast rotation function with diffraction data of 4Å resolution. Translational functions were calculated by applying Lattman's program. From the three Harker sections, the relative translational components between the model BTRY and that in our crystal were found to be ΔX=-10.1, ΔY=-3.0 and ΔZ=-13.3Å. Group refinement (Huber and Schneider, 1985) improved the above rotational and translational parameters to α=236.4°, β=45.7°, γ=74.4° and ΔX=-9.8 Å, ΔY=-3.0 Å, ΔZ=-13.1 Å. After the model BTRY molecule was oriented and located in the unit cell at the correct position using the refined parameters, the R-factor dropped to 0.39. Four cycles of EREF refinement (Jack and Levitt, 1978) of the BTRY molecule improved the R-factor to 0.353. A Sim-weighted (Sim, 1958) electron density map both of 3Å resolution were calculated. Dense contour levels apparently attributable to the MBILF could be seen clearly near the active center of BTRY molecule. The molecular size of MBILF was estimated at 15x15x25 Å. It could be unambiguously seen that the electron density corresponding to the long side chain of Lys 20 of MBILF molecule extends deeply into the specific pocket and terminates near ASP 189 OD1 of BTRY. The polypeptide folding could be traced in a Fourier map with reference to a difference Fourier. A preliminary stereoscopic model of MBILF has been constructed. Five cycles of EREF refinement of the intact complex molecule has reduced the R-factor to 0.346. Further refinement of the MBILF-BTRY complex is now in progress. The 3-dimensional structure of MBILF-BTRY complex reveals the first Bowman-Birk type inhibitor complex which may lead to structure-function correlation studies.