

02.6-2 CRYSTALLISATION AND PRELIMINARY X-RAY DATA FOR TET-REPRESSOR AND THE TET/TETRACYCLINE COMPLEX: By H.E. Parge, M. Schneider, U. Hahn, W. Hillen\*, and W. Saenger, Institut für Kristallographie, Freie Universität Berlin, Takustr. 6, D-1000 Berlin 33; \*Institut für Organische Chemie und Biochemie der TH, Petersenstr. 22, D-6100 Darmstadt, FRG.

The TET-repressor (encoded by the *E. coli* transposon Tn 10) controls the expression of tetracycline resistance as well as its own synthesis. On repressor/tetracycline complex formation the two genes are transcribed in divergent directions. The two corresponding operators are separated by 11 base pairs, each binding one TET-repressor (2 x 23,000 Daltons).

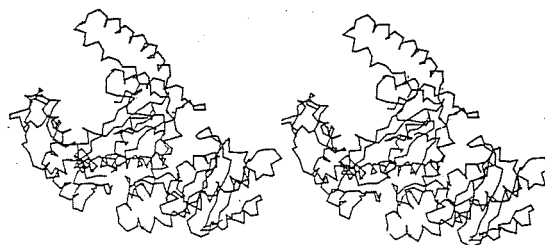
The TET-repressor has been crystallised in two forms, I (P4<sub>2</sub>2<sub>1</sub>2 or P4<sub>1</sub>2<sub>1</sub>2, a = b = 74.2, c = 94.4 Å, V<sub>M</sub> = 2.82) and II (P2<sub>1</sub>2<sub>1</sub>2, a = 73.5, b = 58.5, c = 39.9 Å, V<sub>M</sub> = 1.72) using phosphate as precipitant by both vapour diffusion and microdialysis techniques.

Crystals have also been obtained of the TET/tetracycline complex, III, which are nearly isomorphous with the tetragonal form of the native repressor with cell dimensions, a = b = 73.3, c = 94.7 Å.

The present resolution on still photographs of both the native and complex crystals is 3.0 Å X-ray structural analysis is in progress.

02.6-4 STRUCTURE OF THE LARGE FRAGMENT OF *E. coli* DNA POLYMERASE I COMPLEXED WITH dCMP. By D. Ollis,\* P. Brick,\*† R. Hamlin,‡ N.G. Xuong‡ and T.A. Steitz,\* \*Dept. of Molecular Biophys. and Biochem., Yale, New Haven, CT, USA. †Present Address: Biophysics Group, Imperial College, London, SW7 2AZ UK. ‡Dept. of Physics, Univ. of Calif., La Jolla, CA USA.

A model of the large fragment of *E. coli* DNA polymerase complexed with d-CMP has been built by fitting its amino acid sequence into a 3.0 Å electron density map. The polymerase structure consists of two domains, the smaller of which binds Zn-dCMP. The larger domain has two large protrusions giving it a "claw-like" shape. The location of an amino acid that is altered in a mutant defective in DNA binding as well as the size and shape of the cleft suggests that double stranded DNA may bind between the "claws".



02.6-3 THE STRUCTURE OF A PROKARYOTIC DOUBLE-STRANDED DNA BINDING PROTEIN. by K.Appelt, I.Tanaka, S.W.White and K.S.Wilson\* of Max-Planck Institute for Molecular Genetics, Abt.Wittmann,Imnestrasse 63-73, 1000 Berlin 33.

DNA binding protein II (DNABPII) binds non-specifically to double-stranded DNA inducing the formation of bead-like structures similar to those formed by histones in eukaryotes. The protein has a molecular weight of 9,500 and forms dimers (and possibly higher order oligomers such as tetramers) in solution. DNABPII appears to be ubiquitous in the eubacterial kingdom and also occurs in archeobacteria. It is present at the level of up to 100,000 copies per cell. Sequences have been determined for DNABPII's from several sources and their homology established.

We have crystallised DNABPII from the thermophilic bacterium *Bacillus stearotherophilus*. The crystals are monoclinic in spacegroup P2<sub>1</sub>, and there are 3 dimeric molecules in the unit cell. The structure has been determined at 3 Å resolution using a single isomorphous derivative with K<sub>2</sub>UO<sub>2</sub>F<sub>6</sub>. The three dimers in the cell each sit on independent 2-fold axes, and the molecules related by non-crystallographic symmetry were assumed to have identical structures and averaged in the X-ray analysis.

The monomer is composed of a three-stranded anti-parallel beta pleated sheet and two alpha helices. The dimer is stabilised primarily by a hydrophobic core. Extending from either side of the sheets are two 'arms' which are disordered in our crystals.

We have investigated possible interactions of the protein with DNA using a graphics system, and propose a model for DNA binding in which the two arms of DNABPII lie in the complementary grooves on either side of a B-DNA helix.

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02.7-1 CRYSTALS OF TOMATO ASPERMY VIRUS By H.S. Savithri, Department of Biochemistry and M.R.N. Murthy, Molecular Biophysics Unit, Indian Institute of Science, Bangalore-560 012, Karnataka, India.

Tomato aspermy virus belongs to the cucumo class of plant viruses. Cucumo viruses contain 180 copies of protein subunits arranged in a T=3 icosahedral surface lattice. We have grown crystals of tomato aspermy virus in 0.1M sodium citrate buffer, pH 5-6, by vapour diffusion technique using polyethylene glycol 6000 as a precipitant. The crystals grown in the absence of a reducing agent breakup into small fragments, presumably due to the oxidation of sulphhydryl groups. However, the crystals grown in the presence of dithiothreitol are stable. Three crystal morphologies, square pyramidal, triangular and octahedral were observed. The square pyramidal crystals diffracted X-rays only to 10 Å resolution. The triangular plates and the octahedral crystals diffract to 3 Å resolution and do not show significant radiation damage for 30 hours of exposure. Examination of the latter two forms revealed that they both belong to Rhombohedral R3 space group with cell constants a = 290 Å and α = 60°. The unit cell is compatible with only one virus particle. Hence, the particle three fold is coincident with the crystal three fold. Approximately, 26% of the unit cell volume is occupied by the solvent. Three-dimensional data collection using screenless oscillation photography is in progress.