

MS04.04.03 HIGH RESOLUTION CRYSTAL STRUCTURE OF A GUANINE TETRAPLEX. Ben Luisi, Kathryn Phillips, Zbyszek Dauter*, Alistair Murchie** and David Lilley** Department of Biochemistry, Cambridge Univ., Tennis Court Road, CB2 1QW, U.K. *Department of Chemistry, York Univ. Heslington Y01 5DD, U.K. **Department of Biochemistry, Dundee Univ., Dundee DD1 4HN, U.K.

We report the crystal structure of a DNA molecule with four, parallel strands at 0.95 Å resolution. This tetraplex DNA is formed by d(TGGGGT), and sixteen strands occupy the asymmetric unit. The structure is stabilized by tetrads of hydrogenbonding guanines and by coordination of sodium ions between successive tetrad planes. The high resolution structure permits a detailed analysis of the hydration structure in and around the helical grooves and of the interaction of ions with the phosphate groups. The sugars have a well defined puckering geometry and distinctive base-stacking interactions are observed. Four-stranded DNA structures have been implicated in the organization of telomeres and other large assemblies, and these functions will be discussed.

MS04.04.04 THE THERMODYNAMICS OF CRYSTAL PACKING: SOLVENT VERSUS STERIC EFFECTS. P. Shing Ho and Todd F. Kagawa, Dept. of Biochemistry and Biophysics, Oregon State University, Corvallis, OR 97331

Molecular mechanics methods are useful tools for modeling the atomic details of molecular structures, but have not been generally successful at predicting global conformations. Solvent free energy (SFE) calculations, in contrast, have successfully modeled the contribution of hydrophobicity to the relative stability of protein and DNA conformations, but is not currently applicable at the atomic level. Here, we use a crystallographic assay to study the contribution of competing steric and hydrophobic effects on the orientation of an asymmetric DNA duplex within a crystal lattice. In these studies, we monitor the effect of single methyl groups of cytosine bases on the specific orientations of sequences of the type d(CGCCCG)-d(CGCGCG). From this we determine the free energies of intermolecular interactions in the lattice. Both solvent and steric interactions appear to contribute to assembly of the lattice: the contribution of steric interactions, however, are overestimated by a factor of 4 using standard Lennard-Jones potentials. The assay provides a scaling factor between the two competing interactions and thus a means for incorporating hydrophobicity, as estimated from SFEs, into the more detailed molecular mechanics methods for predicting macromolecular folding and assembly. This should apply to any assembly process, including the packing of helices in polypeptides.

Finally, we have assessed the overall effect of crystal packing on oligonucleotide structures by comparing essentially identical six and eight-base-pair sequences that crystallize as A-DNA. The hexanucleotide conformation of d(GCGCGC) is that of canonical A-DNA, while that of the octanucleotide d(GCGCGCGC) is the longer and narrower structure that is typical of this length. The inter-duplex interactions are identical in both sequences. We therefore have introduced the concept of a "symmetry force" to account for this difference. Here, we define the need for duplexes to adopt a symmetric lattice as the primary force that drives the conformational distortions observed in the oligonucleotide crystals. We estimate the magnitude of these forces by comparing the conformational energies of the six and eight base pair sequences.

MS04.04.05 STRUCTURE OF A DNA-PORPHYRIN COMPLEX Loren Dean Williams, Leigh Ann Lipscomb, Fang Xiao Zhou, Steven R. Presnell, Rebecca J. Woo, Mary E. Peek, School of Chemistry & Biochemistry, Georgia Institute of Technology, Atlanta, GA 30332-0400.

We report the 2.4 Å structure of a complex in which CuTMPyP4 [copper (II) meso-tetra(N-methyl-4-pyridyl)porphyrin] flips a base out of the helical stack of duplex d(CGATCG). The porphyrin system is located within the helical stack, with the copper atom near the helical axis. The porphyrin binds by normal intercalation between the C and G of 5' TCG 3' and by extruding the C of 5' CGA 3'. The DNA hexamer forms a distorted right-handed helix with only four normal cross-strand Watson-Crick base pairs. Two pyridyl rings of the porphyrin are located in each groove of the DNA. The complex appears to be extensively stabilized by electrostatic interactions between positively-charged nitrogen atoms of the pyridyl rings and negatively-charged phosphate oxygen atoms of the DNA. Favorable electrostatic interactions appear to draw the porphyrin into the duplex interior. These favorable interactions offset unfavorable steric clashes between the pyridyl rings and the DNA backbone. We believe these pyridyl-backbone clashes extend the DNA along its axis and preclude formation of van der Waals stacking contacts in the interior of the complex. The unusual lack of van der Waals stacking contacts observed in the porphyrin complex destabilizes the DNA duplex and decreases the energetic cost of local melting. Thus extrusion of a base appears to be facilitated by pyridyl-DNA steric clashes.

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MS04.04.06 CRYSTAL STRUCTURE OF AN END-TO-END BINDING OF TWO NETROPSIN MOLECULES TO THE DNA DECAMER d(C₅I₅). Xin Chen*, S. T. Rao#, K. Sekar#, M. Sundaralingam#. Departments of Chemistry# and Biochemistry The Ohio State University, Columbus, OH 43210

All known minor groove drugs bind DNA in the AT rich region. We recently demonstrated by X-ray crystallography that the alternating DNA octamer d(ICICICIC) binds two distamycin drugs side-by-side simultaneously (Xin Chen, B. Ramakrishnan, S. T. Rao and M. Sundaralingam, *Nature Structural Biology*, **1**, 167, 1994). This binding involves a significant widening of the DNA minor groove. We have also successfully replaced the I's and C's by A's and T's and obtained similar drug DNA complexes (Xin Chen, B. Ramakrishnan and M. Sundaralingam, unpublished results), which indicate that inosine resembles adenine in the minor groove. In our continuing study of the binding of minor groove drugs to other related DNA sequences, we have obtained crystals of the complex between the DNA decamer d(CCCCCIIII) and netropsin. The asymmetric unit contains two DNA duplexes each with two netropsin molecules bound. Interestingly, the structure reveals that the two netropsin molecules are bound in an end-to-end fashion. The details of the binding and the comparison with the side-by-side binding mode will be presented.

The crystals are in space group P1 with cell dimensions $a=32.56\text{Å}$, $b=32.59\text{Å}$, $c=37.63\text{Å}$ and $\alpha=86.30^\circ$, $\beta=84.50^\circ$, $\gamma=68.58^\circ$. The structure was solved by molecular replacement and refined by X-PLOR to a final R-factor of 0.22 using 3688 reflections (78% complete) at 2.5Å resolution.

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