CRYSTALLOGRAPHIC STUDIES OF DNaseI: OLIGONUCLEOTIDE COMPLEXES. By A. Lahm and D. Suck, Biological Structures Division, EMBL, Heidelberg, West Germany.

Interactions of proteins with double-stranded DNA vary from being strictly sequence-specific to being completely unspecific. Examples of the former are the interaction of repressors with operator sequences or of restriction endonucleases with their cognate recognition sequences; an example of the latter is the histone-like DNA binding protein II from bacteria. Structural models have been proposed for either class and will be discussed briefly. A third class of proteins with properties in between these two extremes bind to DNA in a sequence-dependent, rather than a sequence-specific or completely random manner. These proteins somehow recognize sequence-dependent variations in DNA conformation. Bovine pancreatic DNaseI belongs to this group of proteins and accordingly shows a sequence-dependent DNA-cutting pattern. Based on the 3D-structure of DNaseI we have proposed a model for binding to and cutting ds-DNA. Subsequently, we have succeeded in co-crystallizing a series of self-complementary oligonucleotide duplexes with DNaseI. HPLC-analysis of the dissolved crystals showed that the bound oligonucleotides were partially degraded in a sequence-dependent manner. 1.5 A data have been collected for one of these complexes and its structure was solved by the molecular replacement method using the refined DNaseI co-ordinates, and observed amplitudes to 3 A resolution. A piece of DNA was built into the resulting difference density and is presently being refined. At the present state of the analysis it is clear that the oligonucleotide adopts a somewhat distorted B-type DNA conformation. In agreement with the predicted model (Suck and Gefener, Nature, 1986, 321, 620-25) an exposed loop of the dissolved crystals interacts in a minor groove of B-DNA and electrostatic contacts are formed between several Arg and Lys residues with phosphates of both DNA strands across the minor groove. The mechanism of sequence-dependent cutting of ds-DNA by DNaseI will be discussed on the basis of the 3D-structure of the complex.