Sructure Determination of sc*Pvu*II by Crystallographic and SAXS Methods

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*Pvu*II is the first restriction endonuclease which has been converted from its wild-type (wt) homodimeric form into a single chain (sc) protein by tandemly joining the two subunits through the peptide linker GlySerGlyGly. The DNA cleavage activity of the enzyme is thereby largely retained [1]. The single-chain enzyme provides a scaffold for the development of asymmetrically modified restriction endonucleases.

Crystals of sc*Pvu*II, obtained by the hanging drop vapor diffusion method, were measured at EMBL/DESY (X11 beamline). The crystals diffract to a resolution of 2.35Å and belong to space group $P4_2$ with the unit cell parameters a=b=102.0, c=100.3 Å and two molecules in the asymmetric unit. The crystal structure was determined by molecular replacement method using the AMORE program, using the DNA-binding subdomain (residues 36-157) of the wt*Pvu*II monomer [2] as search model. The crystal structure shows that sc*Pvu*II adopts a more compact conformation compared to the wt form. SAXS measurements at EMBL/DESY (X33 beamline) confirm this result.

[1] Simoncsits A., et al., *J. Mol. Biol*., 2001, **309**, 89-97. [2] Athanasiadis A., et al., *Stuct. Bio.,* 1994, **1**, 469-475.

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