bound and unbound forms by the circular dichroism analysis. In order to investigate the molecular mechanism for recognizing its effecter specifically, the crystal structure of MobR in complex with 3-hydroxybenzoate has been determined at 2.25 Å resolution. Diffraction data were collected on the BL44XU at SPring-8. The structure showed that MobR forms homo dimer and the subunit has a winged helix-turn-helix DNA binding domain as well as the other members of the MarR family. Furthermore, the effecter-binding pocket is distant from DNA binding domain and 3-hydroxybenzoate was recognized by side chains H31, R37 and Q134 in the pocket.

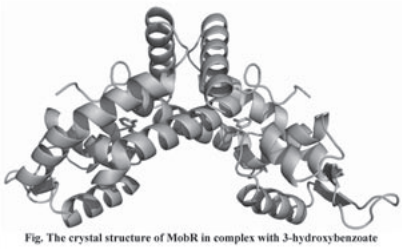

Fig. The crystal structure of MobR in complex with 3-hydroxybenzoate

Keywords: X-ray crystallography, DNA-binding proteins, transcription factor structure

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Structure of human RECQ1 helicase: Identification of a putative DNA strand separation pin

Ashley CW Pike[1], Binesh Shrestha[1], Nicola Burgess-Brown[1], Laura Muzzolini[2], Alessandro Vindigni[2], Opher Gileadi[1]
[1]Structural Genomics Consortium - University of Oxford, SGC, Old Road Campus Research Building, Headington, Oxford, Oxfordshire, OX3 7DQ, UK, [2]International Centre for Genetic Engineering and Biotechnology (ICGEB), I-34012 Trieste, Italy, E-mail：ashley.pike@sgc.ox.ac.uk

RECQ-like helicases are ATP- and Mg^{2+}-dependent enzymes that are involved in maintaining genome integrity. The RECQ helicase family has five representatives in the human genome. Here we describe the 2Å crystal structure of human RECQ1 in complex with Mg-ADP. Overall, the structural architecture closely resembles that of bacterial RECQ albeit with altered relative domain positioning. All domains are conserved, including two RecA-like modules, the RECQ-specific zinc-binding and a winged-helix (WH) domain. The orientation of the two RecA domains, believed to harbour the helicases' ATP-dependent translocation activity, exhibits considerable variability as adjudged from the overall conformation adopted by the protein in multiple crystal forms. The C-terminal WH domain is positioned in a novel orientation in the human enzyme resulting in a more elongated molecule. This domain also exhibits a prominent beta-hairpin structural element, not seen in the bacterial enzyme, that is reminiscent of the DNA strand separation pin of other DNA helicases. The role of this pin is to act as an unwinding element by displacing the individual strands of duplex DNA. Mutation of the Tyr residue (Y564) that caps the separation pin as well as shortening the beta-hairpin abolishes DNA-unwinding activity confirming that this structural element plays a key role in DNA strand separation. The probable DNA-binding mode of RECQ1 can be inferred by comparison with other DNA-helicase complexes. The structure will be presented in detail along with implications for recognition and binding of DNA. In addition, progress on the structural characterisation of the other members of RECQ family will also be summarised.

Keywords: DNA replication, protein-DNA interactions, structural genomics

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A new nicking enzyme is developed from a mutant of the modified type II restriction enzyme scPvuII

Chrysi Meramveliotaki[1,2], Maria Androulaki[1,2], Elias Eliopoulos[2], Michael Kokkinidis[1]
[1]University of Crete, Greece, Biology, Vassilika Vouton, Heraklion, Crete, 71409, Greece, [2]Agricultural University of Athens, Iera Street 75, Athens, 11855, Greece, E-mail：chrysamer@biology.uoc.gr

PvuII is the first restriction endonuclease (nuclease component of one of the type II restriction-modification systems of Proteus vulgaris) 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 [1]. The DNA cleavage activity of the enzyme is thereby largely retained. The determined crystal structures (from twinned and un-twinned crystal forms) [2] show that the apo scPvuII adopts a more compact conformation compared to the wild-type form. Four mutants of scPvuII, which address specific aspects of its interactions with DNA have been crystallized and studied with similar results. In contrast, in equilibrium in solution, scPvuII and the mutants adopt two conformations, as proved from gel-filtration [3] and SAXS measurements. As proved from the crystal structure, the peptide linker forms new H-bonds in that area of the protein, which are possibly responsible for the two conformations of the apo enzyme. Several attempts for the co-crystallization of the scPvuII or a mutant - DNA complex were not successful, probably because of the serious aggregation problem of the proteins, as studied by Dynamic Light Scattering techniques [4]. The complex formation was also studied by SAXS method and it is actually formed in the case of scPvuII in the expected 1:1 molar stoichiometry. Later studies based on FRET technique proved that the DNA molecule in the complex is not bended [4], as it is also the case for the wtPvuII, but not for other type II restriction enzymes. The proteins - DNA interactions were also studied by EMSA techniques and the most outstanding result was that the D34G/K70A mutant of scPvuII proved to be a nicking enzyme.

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Keywords: scPvuII, nicking enzyme, mutant

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Structural characterization of ANAC019, a member of the NAC family of plant transcription factors

Ditte H Welner[1], Heidi A Ernst[1], Addie N Olsen[1], J. G Grossmann[2], Charlotte Helgstrand[1], Karen Skriver[1], Leila L Leggio[1]
[1]University of Copenhagen, Dept. of Chemistry, Universitetsparken 5, Copenhagen OE, Denmark, 2100, Denmark, [2]School of Biological Sciences, The University of Liverpool, Liverpool L69 7ZX, UK, E-mail：dittewelner@gmail.com

The NAC proteins constitute a large group of plant specific transcription factors which play important roles in biotic and abiotic stress responses and plant development (Olsen et al, 2005, Trends in Plant Science, 10:79-87). NAC proteins consist of two regions: a conserved N-terminal region (NAC domain) with DNA binding and oligomerization abilities, and a diverse C-terminal region which functions as a transcriptional activator. We have previously determined the first crystal structure of a conserved DNA-binding