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The ββα-Me type II restriction endonuclease Hpy99I from the human pathogen *Helicobacter pylori* has been overexpressed from a synthetic gene in *E. coli* and crystallized in complex with target DNA. The enzyme binds the pseudopalindromic CGWCG| target sequence as a dimer and cleaves two DNA strands with unusual stagger (five nucleotide 3'-overhangs, "|" marks the cleavage site). The Hpy99I protomer consists of an antiparallel β-barrel and two β4α2 repeats. The repeats are poorly conserved on a sequence level but are readily indentified in a structural comparison. Each repeat coordinates a structural zinc ion with four cysteine thiolates in two CXXC motifs positioned in a β-hairpin and at the N-terminal end of an α-helix. The ββα region of the second β4α2 repeat holds the catalytic metal ion (Me) via Asp148 and Asn165 and activates a water molecule with the general base His149. Hpy99I dimer forms a tight ring-like structure around the DNA. Each protomer recognizes one half of the target sequence and contacts the CG/GC base pairs on the major and minor groove side via the first and second β4α2 repeat, respectively. The enzyme interacts with the central symmetry-breaking base pair of the recognition sequence only on the minor groove side, where A:T resembles T:A and G:C is similar to C:G. The Hpy99I-DNA co-crystal structure provides the first detailed illustration of the ββα-Me active site in restriction endonucleases and complements the information on the use of this motif in other groups of enzymes such as homing endonucleases (e.g. I-PpoI, I-HmuI) and Holliday junction resolvases (e.g. T4 endonuclease VII) [1].

[1] Sokolowska M., Czapinska H., Bochtler M. *Nucleic Acids Res.,* **2009**, Apr 20. [Epub ahead of print]

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Structural Basis for Rab-effector Specificity. Amir R. Khan[a], Rosario Recacha[a], Nicholas Jagoe[a]. [a]*School of Biochemistry and Immunology, Trinity College Dublin, Ireland.*
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The Rab family of small GTPases orchestrate the trafficking of vesicular cargo in eukaryotic cells. Rabs have a conserved three-dimensional fold in their active (GTP) state, yet recognize a distinct subset of effector proteins to mediate their biological effects. Unlike Rabs, effector proteins are diverse in size and composition. We have determined the crystal structures of Rab6 and Rab11 with their effectors to gain insight into specificity and subsequent biological function. Our lab and others have observed that Rabs generally bind alpha-helical determinants of cognate effectors via highly conserved residues in switch I, switch II and interswitch regions. However, conformational variability in these conserved regions plays a key role in effector specificity. The seemingly contradictory properties of specificity and promiscuity (Rab6 binding to several unrelated effectors) will be discussed in light of emerging structural data.

[1] Recacha R, Boulet A, Jollivet F, Monier S, Houdusse A, Goud B & **Khan AR** (2009) Structural basis for recruitment of Rab6-Interacting Protein 1 to Golgi via a RUN domain. *Structure*17:21-30.

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