MS11-P8 Hpy188I-DNA structures-snapshots of the GIY-YIG nuclease mediated catalysis. Monika Sokolowska, ^a Honorata Czapinska, ^a Matthias Bochtler, ^{abc} ^aInternational Institute of Molecular and Cell Biology, Trojdena 4, 02-109 Warsaw, Poland, ^bInstitute of Biochemistry and Biophysics PAS, Pawinskiego 5a, 02-106 Warsaw, Poland, ^cSchools of Chemistry and Biosciences, Main Building, Cardiff University, Cardiff CF10 3AT, UK. E-mail: msokolowska@iimcb.gov.pl

The GIY-YIG nuclease domain is present in all kingdoms of life and has diverse functions. It is found in the eukaryotic flap endonuclease and Holliday junction resolvase Slx1-Slx4, the prokaryotic nucleotide excision repair proteins UvrC and Cho, and in proteins of selfish genetic elements. Here we present the structures of the ternary pre- and post-cleavage complexes of the type II GIY-YIG restriction endonuclease Hpy188I with DNA and a surrogate or catalytic metal ion, respectively [1]. Our structures suggest that GIY-YIG nucleases catalyze DNA hydrolysis by a single substitution reaction. They are consistent with a previous proposal that a tyrosine residue (which we expect to occur in its phenolate form) acts as a general base for the attacking water molecule. In contrast to the earlier proposal, our data identify the general base with the GIY and not the YIG tyrosine. A conserved glutamate residue (Glu149 provided in trans in Hpy188I) anchors a single metal cation in the active site. This metal ion contacts the phosphate proS oxygen atom and the leaving group 3'-oxygen atom, presumably to facilitate its departure. Taken together, our data reveal striking analogy in the absence of homology between GIY-YIG and ââá-Me nucleases. Our work has been published back-to-back with the structural studies of Eco29kI restriction endonuclease [2].



- Sokolowska, M., Czapinska, H. & Bochtler, M. (2011). Nucleic Acids Res. 39, 1554-1564.
- [2] Mak, A. N., Lambert, A. R. & Stoddard, B. L. (2010). Structure 18, 1321-1331.

Keywords: GIY-YIG nuclease, catalytic mechanism, restriction enzyme

MS11-P9 Crystal structure and functional characterization of *Helicobacter pylori* DsbG. <u>Hye-Jin Yoon</u>, Ji Young Yoon, Jieun Kim, Sang Jae Lee, Hyoun Sook Kim, Ha Na Im, Kyoung Hoon Kim, and Se Won Suh, *Seoul National University, Korea* E-mail: <u>yoonhj@snu.ac.kr</u>

Dsb proteins play important roles in bacterial pathogenicity. To better understand the role of Dsb proteins in *Helicobacter pylori*, we have structurally and functionally characterized *H. pylori* DsbG (HP0231). The monomer consists of two domains connected by a helical linker. Two monomers associate to form a V-shaped dimer. The monomeric and dimeric structures of *H. pylori* DsbG show significant differences compared to *E. coli* DsbG. Two polyethylene glycol molecules are bound in the cleft of the V-shaped dimer, suggesting a possible role as a chaperone. Furthermore, we have discovered that *H. pylori* DsbG functions as a reductase against HP0518, a putative L,D-transpeptidase.

[1] Yoon JY et al. (2011). FEBS Lett. 585, 3862-3867.

Keywords: DsbG;disulfide bond; reductase activity