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Report:

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Mutations in DNA mismatch-repair genes predispose for the most prevalent type of familial cancers, Hereditary Non-polyposis Colorectal Cancer (HNPCC). Mismatch DNA repair is highly conserved in organisms ranging from *Escherichia coli* to human. Repair starts by recognition and binding of mismatched DNA by the protein MutS (or its eukaryotic homologs). *E. coli* MutS is a 953 amino acid protein which binds DNA mismatches. We want to determine the crystal structure of MutS with DNA to understand the the binding mode and the mode of action of MutS in mismatch repair.

Crystals were obtained of full-length MutS complexed to a G/T mismatch oligomer. Two crystal forms were found, space group $P2_12_12_1$, (a=89.5Å, b=91.9Å and c=260.2Å) and $C2_12_12_1$ (a=180Å, b=530 Å, c=90Å). The crystals contain ~ 200 kD ($P2_12_12_1$) and 400 kD ($C2_12_12_1$) in the asymmetric unit. Diffraction and resolution are strongly dependent on beam intensity, making synchrotron time obligatory for any test. Thus far the search for suitable derivatives has been hampered by non-isomorphism.

Data collection on MutS crystals has been quite succesful, resulting in 5 potential derivative data sets in the $P2_12_12_1$crystal form as well as a Selenomethionine MAD data set in $C222_1$

	Reso	cell dimensions	mosaic	(degrees) I/sigI in last shell	completeness % (last shell)	Rmerge % (last shell)
$P2_12_12_1$						
I4	2.9	89.9 92.7 261.9	0.66	1.5	99 (55)	07. (30.)
La	3.0	90.3 94.1 262.0	0.72	1.8	99 (59)	09. (44.)
Pt	2.8	90.4 93.3 263.3	0.55	1.5	100 (78)	14. (77.)
I4l	3.7	96.4 93.2 274.9	0.8	1.2	96 (73)	13. (100.)
Tl	3.7	91.3 91.9 270.2	0.7	1.5	86 (72)	14. (72.)
$C222_1$						
Br	3.5	188.0 525.8 91.1	0.6	1.0	92.2	14. (65.0
ndan3	4.0	186.7 525.8 90.6	0.6			
ghh	4.1	187.8 535.1 92.0	1.1	1.1		
peakse	3.5	188.3 535.8 92.0	1.0	1.4	99	10. (60.)
inflse	3.5	188.3 535.9 92.0	1.0	1.7	99	8. (60.)
remse	3.5	188.6 536.7 92.0	1.0	1.3	90	7. (52.)
spekse	3.6	188.7 536.4 92.1	1.0	1.5	100	13. (70.)

Despite the improvement in the data, however, no good derivatives have been identified yet. Large changes in cell parameters give rise to considerable non-isomorphism, making it difficult to identify a good heavy atom derivative. The selenomethionine protein reproducably gives crystals with a shifted spacegroup, by loss of the symmetry. The resulting pseudosymmetry is variable between crystals. In the new space group 100 methionines are available in the crystal and this provides difficulties in finding the sites, given the marginal quality of the data. We are continually improving our crystallizations and we plan to repeat the MAD experiment on ID14-4, with better crystals. We will make use of high redundancy in order to overcome the weakness of the data.