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High-end Solution for In-house Protein Crystallography. Marianna Biadene[a], Matt Benning[b], Vernon Smith[a]. [a]*Bruker AXS GmbH, Karlsruhe, Germany.* [b]*Bruker AXS Inc, Madison, WI, USA.*
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The ability to collect quality diffraction data in-house provides a an improvement in productivity and efficiency, reducing the reliance on synchrotron sources. High-end in-house solutions provide the maximum flexibility for the scientist. Due to advances in optics and the introduction of microfocus rotating anode generators, there has been a remarkable increase in the performance of home laboratory X-ray systems. When combined with an ultra sensitive CCD detector, these systems can produce data comparable to that collected at synchrotron beamlines. The overall performance of these solutions allows for a number of experiments which are hardly possible on many of the currently installed equipments:

- data sets suitable for in-house SAD phasing exploring the anomalous signal
- high quality, high-resolution data set with a resolution better than 1.2 Å
- complete data collection within less than a minute X-ray exposure time

We will present data on crystals of a number of number of proteins to demonstrate the exciting capabilities of a high-end systems such as the X8 PROTEUM (figure 1).

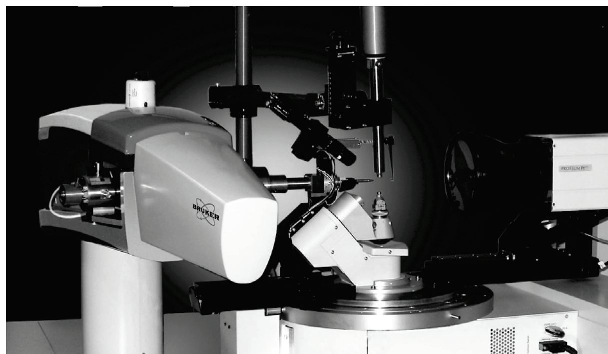

Figure 1: High-end In-house solution (X8 PROTEUM).

Keywords: SAD-phasing, high-resolution data, data collection time

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How do S-layers bind to bacterial cell walls. [a]A. Đordić[a], T. Pavkov[a], E. M. Egelseer[b], U. B. Sleytr[b], W. Keller[b]. [a]*Institute of Molecular Biosciences, K.F. University Graz, Austria.* [b]*Center for Nanobiotechnology, University of Natural Resources and Applied Life Sciences, Vienna, Austria.*
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Monomolecular paracrystalline surface layers (S-layers) are composed of a single (glyco)protein and are the most commonly observed cell surface structures of bacteria and archaea. Because of their diverse properties S-layers have various potential applications in nanobiotechnology [1]. However, detailed structural information on S-layer proteins is very scarce. In order to determine the structure-function relationship of SbsC, the S-layer protein from *Geobacillus stearothermophilus*, deletion mutants were produced. It was shown that the N-terminal part is responsible for binding to the secondary cell wall polymer (SCWP) and that the C-terminal part is essential for self-assembly [2]. Recently, the crystal structure of the C-terminally truncated form $rSbsC_{(31-443)}$ was solved to 2.4 Å [3].
To further characterize the interaction of SbsC with SCWP, binding studies with ITC and Thermofluor were performed. For some of the derivatives strong binding could be shown. To structurally describe the exact binding site(s), soaking and co-crystallization experiments of potential $rSbsC_{(31-443)}$-ligand complexes were set up. Crystals were obtained by co-crystallization with one of the SCWP derivatives. The complex crystallizes in the same space group as the native $rSbsC_{(31-443)}$ ($P2_1$) but with some changes in the unit cell parameters. The resolution could be improved to 1.8 Å and the structure was solved by molecular replacement. Comparing the two structures, with and without ligand, we observed a slight domain movement, which is necessary for the binding of the ligand to take place. The residues of $rSbsC_{(31-443)}$ involved in ligand binding are accommodated in domain I and positioned along the positively charged cleft. The crystal structure of the SbsC-ligand complex confirms the previous prediction of the SCWP binding site within the N-terminal domain of SbsC protein.

[1] Sleytr, U.B., Sára, M., J Bacteriol 182 (2000) 859. [2] Jarosch M., EgelseerE.M., Mattanovich D., Sleytr U.B., Sara M., Microbiology 147 (2001) 1353. [3] Pavkov,T., Egelseer E.M., Tesarz,M., Svergun,D.,M., Sleytr,U.B., Keller,W., Structure 16 (2008) 1226.

Keywords: S-layer, co-crystallization, protein-ligand complex

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A Comparison of Models for Giant Haemoglobins Derived from Different Techniques. Helmut Durchschlag[a], Peter Zipper[b], Angelika Krebs[c]. [a]*Institute of Biophysics and Physical Biochemistry, University of Regensburg, Germany.* [b]*Physical Chemistry, Institute of Chemistry, University of Graz, Austria.* [c]*Bioinformatics and Structural Biology, Medical University of Graz, Austria.*
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Extracellular haemoglobins (Hb) found in many invertebrates are giant multisubunit molecules of hexagonal bilayer (HBL) appearance [1]. The most prominent example is *L.t.* Hb; it is a 3.6 MDa haemoprotein, a 180-mer consisting of 144 globin and 36 linker chains. Existence, nature and complexity of these large oxygen carriers have been enigmatic for nearly two centuries. The most intriguing questions addressed in the last years refer to the central cavity within the HBL structure, the precise composition of haeme-containing globins and haeme-free linkers, the exact 3D structure of the complex, its constituents and preferentially bound water molecules, and the biological significance of such huge assemblies.
The 2.6 Å resolution crystal structure of the dodecameric subunit of *L.t.* Hb and data for the 3.5 Å architecture of the complex became available recently [2, 3]. The combination of previous physicochemical data [electron microscopic (EM) reconstructions, SAXS, hydrodynamics] and currently available crystal data allows to scrutinize earlier established