Since CpxP has no homologues of known function, we have initially focused on its biophysical and structural characterization. Using multi-angle laser light scattering (MALLS), small-angle X-ray scattering (SAXS) analysis, and formaldehyde-mediated cross-linking experiments, we show that full-length *E. coli* CpxP is a dimer *in vivo* as well as in pathway inactivating (pH 5.8) and activating (pH 8.0) conditions *in vitro*. Far-UV circular dichroism (CD) was used to demonstrate that CpxP is mainly α-helical, while near-UV CD and SAXS revealed that the protein may undergo a small structural adjustment in response to a pathway-inducing stimulus (pH 8.0).

The crystal structure of CpxP, determined to 2.85 Å resolution, revealed an antiparallel dimer of intertwined α-helices with a highly basic concave surface. Each protomer consists of a long, hooked and bent hairpin fold with conserved LTXXQ motifs forming two diverging turns at one end. Three of six previously characterized *cpxP* loss-of-function mutations, $M_{59}T$, $Q_{55}P$, and $Q_{128}H$, likely result from a destabilization of the protein fold, whereas the $R_{60}Q$, $D_{61}E$, and $D_{61}V$ mutations may alter interactions important for the signalling or proteolytic adaptor functions of CpxP.

Keywords: bacterial, biocrystallography, SAXS

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Structure of monomeric and dimeric Sgt1 protein from *Hordeum vulgare* in solution

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Sgt1 (Suppressor of G2 allele of skp1) is a conserved eukaryotic protein that plays many important roles inside the cell [1]. Originally it was discovered as a component of yeast kinetochore assembly and member of SCF ubiquitin ligase complex. Sgt1 is also an interacting partner of Hsp90 molecular chaperone which is important for stability and folding of many key signaling proteins like kinases and steroid hormone receptors. In plants Sgt1 is involved in disease resistance to many pathogens and mutation of Sgt1 gene leads to lost of R protein triggered resistance in many cases. Sgt1 interacts with COP9 signalosome (involved in 26S proteasome protein degradation pathway) and SCF complexes and probably target resistance regulatory elements for degradation in plants. In humans Sgt1 positive regulate Nod1 innate immunity receptor pathway.

Sgt1 consist of five non-enzymatic domains: N-terminal *tetratricopeptide* repeat domain (TPR), middle CS domain, C-terminal Sgt1 specific domain (SGS) and two variable regions (VR1 and VR2) that interacts with many partner proteins[2]. CS domain share structural homology with p23 protein Hsp90 co-chaperone and also interacts with Hsp90. CS domain interacts with CHORD II domain of plant protein Rar1 which is involved in disease resistance. SGS domain interacts with Leucine-rich repeats protein like Barley R protein Mla1 and yeast adenylyl cyclase cdc35p. TRP domain interacts with *Arabidopsis* SRFR1. It is know that Sgt1 form a dimer in low ionic strength solutions and that dimerization is mediated by TPR domains.

Here we present the structure in solution of *Hordeum vulgare* Sgt1 in monomeric and dimeric form using small angle X-Ray scattering data measured at beamline X33 (EMBL c/o DESY, Hamburg) and homology modeling. Using MCR-ALS analysis [3] we were able to separate scattering curves from complex mixture of both species and model them using rigid body modeling. Sgt1 form an extended conformation in solution with disordered variable regions in both forms. Our observation agrees with biological experiments which shows wide spectrum of Sgt1 interacting partners. Such conformation facilitates interaction between proteins. Dimerization may have regulatory role, which depends on physiological state of the cell.

[1] C. Azevedo, A. Sadanandom, K. Kitagawa, A. Freialdenhoven, K. Shirasu, P.Schulze-Lefert, *Science,* **2002***,* 295, 2073-2076 [2] Y. Kadota, K. Shirasu, R. l. Guerois, *Trends In Biochemical Sciences*, **2010**, *35,* 199-207 [3] J. Blobel, P. Bernado, D.I. Svergun, R. Tauler, M.P ons, *Journal of American Chemical Society,* **2009,** *131*, 4378–4386

Keywords: small-angle X-ray scattering, Sgt1, plant disease resistance

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The structural biology knowledgebase – Structures, functions, methods and more

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The Structural Biology Knowledgebase [1] (SBKB, URL: http://sbkb.org) is a free online resource designed to combine all protocols and results of the structural genomics and structural biology efforts with information from the biological community in order to have a better understanding living systems and disease. We will present examples of how to navigate the SBKB and how to use its new interface and tools to enable biological research. For example, a protein sequence or PDB ID search will provide a list of protein structures from the Protein Data Bank, associated biological descriptions (annotations), homology models, structural genomics protein target information, experimental protocols, and the ability to order available DNA clones. Text searches find structures, annotations, publications, and technology reports created by the Protein Structure Initiative's high-throughput research efforts. Web tools that aid in bench top research, such as TargetTrack, the new target and protocol database (formerly TargetDB and PepcDB), and Sequence Comparison and Analysis tool for protein construct design, will also be demonstrated. Created in collaboration with the Nature Publishing Group, the Structural Biology Knowledgebase Gateway provides a research library, editorials about new research advances, news, and an events calendar also present a broader view of structural genomics and structural biology. The SBKB is funded by the Protein Structure Initiative/NIGMS.

[1] M.J. Gabanyi, P.D. Adams, K. Arnold, L. Bordoli, L.G. Carter, Flippen-J. Andersen, L. Gifford, J. Haas, A. Kouranov, W.A. McLaughlin, D.I. Micallef, W. Minor, R. Shah, T. Schwede, Y.P. Tao, J.D. Westbrook, M. Zimmerman, H.M. Berman *J Struct Funct Genomics.* **2011**, in press.

Keywords: database, function, methodology

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Combination of in-situ optical spectroscopy and macromolecular crystallography

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Complementing structural data obtained by X-ray diffraction with optical spectroscopic techniques has become a growing interest in structural biology. In-situ spectroscopy can reveal the nature of chemical species that remain ambiguous in the electron density maps.

Here we present some of the results obtained by the use of the on-axis micro-spectrophotometer developed at beamline X10SA of the Swiss Light Source [1].

The on-axis geometry of the micro-spectrophotometer is perfect for studying radiation damage and/or the X-ray induced phenomena. Photo-reduction of the copper centers in Copper nitrite reductase from *Achromobacter cycloclastes* has been monitored using UV-Vis absorption spectroscopy. A 'low-dose' data set with the Cu centers still oxidized has been collected and the structure has been validated by spectroscopy.

Raman spectroscopy under resonant conditions (in the Soret absorption band), has been carried out on two different hemoproteins: myoglobin from horse heart and cytochrome c' from *Alcaligenes xylosoxidans* by the use of laser probes at either 405 or 413 nm. In both cases vibrational spectroscopy results complement the active site picture provided by X-ray diffraction.

Non-resonant Raman experiments, with an excitation wavelength in the near infra red domain (785 nm), have been successfully performed on horse heart insulin and hen egg-white lysozyme. For these two proteins, the disulfide bond breakage due to X-ray exposure has been followed by the decreasing intensity of the S-S stretch band.

[1] R.L Owen, A.R. Pearson, A. Meents, P. Boehler, V. Thominet, C. Schulze-Briese, *Journal of Synchrotron Radiation* **2009**, *16*, 173-182.

Keywords: spectroscopy, Raman, UV-Vis absorption

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The HC1 at Diamond, setup, use and first successful results

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A sample humidity control device (HC1) [1-3] has been integrated on the beamlines of the Macromolecular Crystallography Village at Diamond Light Source. The device is primarily used to improve the diffraction quality of crystals via controlled dehydration but is also used for room temperature data collection, as mounting samples is very simple and easy. The current implementation allows easy transfer between the different beamlines and even the beamline laboratories to suit user demand permitting a very efficient use of beamtime. Users can characterise their crystals and optimise their dehydration protocols in a few hours of beamtime. Later, with the device off-line, they can apply these protocols, conditioning and cryo-cooling as many samples as needed in order to obtain the desired dataset. We'll give examples of use that will include some of the most recent results our users have achieved.

[1] J. Sanchez-Weatherby *et al*. *Acta Crystallographica D Biological Crystallography* **2009**, *65*, 1237-46. [2] S. Russi, *et al*. *Journal of Structural Biology* **2011**, doi:10.1016/j.jsb.2011.03.002 [3] J. Kadlec, *et al*. *Nature Structural & Molecular Bilogy* **2011**, *18*, 142-149.

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More flux, less background: Improvements in low power X-ray beam delivery systems

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Microfocus sealed tube sources coupled to advanced x-ray optics provide a high brighness beam in a low maintenance package. These systems are increasingly used in single cristal diffraction applications due to increased performance compared to traditional rotating anode generators in particular for small crystal analysis. Small Angle X-ray Scattering applications require sample illumination with a high brilliance x-ray beam having a well controlled spatial and angular distribution. Indeed high intensity at the sample is required with small beam expansion towards the detector to achieve low values of wave vector.

We will present new developments in the field of beam delivery and beam conditioning systems enabling the optimum use of low power high brightness microfocus sources. These developments include both aspheric multilayer optics with increased capture angle as well as new collimation devices for reduced background signal.

Application data illustrating the capabilities of this new generation X-ray beam delivery system (the GeniX^{3D}) will be presented. The results include diffraction measurements on tiny and poorly diffracting crystals and aperiodic crystal analysis. First results on the combination of the GeniX^{3D} with new hybrid pixel detectors will also be presented highlighting the capabilities of these systems for fast fine slicing measurements.

Small angle X-ray scattering requires well controlled beam propagation with low parasitic scattering while maintaining a high intensity at the sample. We will present how the unique combination of new scatterless collimation with aspheric multilayer optics impacts the useful flux for SAXS applications in particular on diluted solutions.

Keywords: source, optics, saxs

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Copper(I) Anilide Complex $[Na(phen)_3][Cu(NPh_2)_2]$—an Intermediate of Copper-Catalyzed *N*-Arylation of *N*-Phenylaniline Reaction

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Complex $[Na(phen)_3][Cu(NPh_2)_2]$ (**2**), containing a linear bis(*N*-phenylanilide)copper(I) anion and a distorted octahedral tris(1,10-phenanthroline)sodium counter cation, has been isolated from the catalytic C-N cross coupling reaction based on the CuI-phen-*t*BuONa catalytic system. Complex **2** can react with 4-iodotoluene to produce 4-methyl-*N,N*-diphenylaniline (**3a**) with 70.6 % yield. In addition, **2**