periplakin, and envoplakin. The N-terminus of plectin contains an actin binding domain (ABD) build up of a tandem pair of calponin homology domains (CH1 and CH2) and a ~1000 residue long region termed the plakin domain that is conserved among the protein family. The plakin domain of plectin contains protein-protein interaction sites that are important for the localization of plakins at junctional complexes. We have previously identified eight canonical Spectrin Repeats (SR) in the sequence of the plakin domain of plectin (SR1 to SR5 and SR7 to SR9), and an additional shorter SR-like domain (SR6). The SR2 and SR3 of plectin are connected by a ~20-residues long linker predicted to be non-helical, while repeats SR3 to SR9 occur contiguous in the plectin sequence. Despite the existence of several crystallographic structures of isolated pairs of spectrin repeats, there is not structural information on the global shape and conformation of the plakin domain of plectin. Here, we use Small-Angle X-ray Scattering (SAXS) to generate low resolution models of the whole plakin domain and smaller multi-repeat fragments. The available plectin crystallographic structures are further used to generate pseudo-atomic models, either by docking or rigid-body modelling procedures.

**Keywords: SAXS, plectin, spectrin repeats**

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**Low resolution structure in solution and amyloidogenesis of human cystatin C**

Maciej Kozak,[a] Michał Taube,[a] Magdalena Murawska,[a] Aneta Szymańska,[b] Anders Grubb,[c] *[a]Department of Macromolecular Physics, Faculty of Physics A. Mickiewicz University, Poznań (Poland).* *[b]Department of Medicinal Chemistry, Faculty of Chemistry, University of Gdańsk, Gdańsk (Poland).* *[c]Department of Clinical Chemistry, Lund University, Lund (Sweden).* E-mail: mkozak@amu.edu.pl

Human cystatin C (HCC) is a cysteine proteases inhibitor which inhibits proteins belonging to the papain and legumain families. The general fold of monomeric form of cystatin has been defined by the three-dimensional structure of chicken cystatin. Crystallographic studies of native HCC (tetragonal [2] and cubic [1] crystal forms) have shown that the protein exists in crystal in the form of symmetric three-dimensional domain-swapped dimers. Human cystatin C is also strictly related to the occurrence of Iceland type amyloidosis. HCC Leu68Gln mutation causes protein oligomerization and deposition of amyloid fibrils.

Insoluble amyloid fibrils are found as deposits in patients with a range of conformational diseases, e.g. Alzheimer disease, reactive amyloidosis or cystatin C amyloidosis. [3-5]. Knowledge of the molecular mechanism causing the transition of physiologically normal and soluble proteins to toxic oligomers and insoluble fibrils is essential for the development of treatment strategies for this group of common, but currently incurable, diseases.

In this study we perform a small angle scattering experiment to probe the conformation and low resolution structure of native form of HCC and mutants Val57Pro, Val57Asn, Val57Asp to probe the effect of this mutations on cystatin C structure in solution. The aim of our study was also the characterisation of the formation of HCC oligomers and amyloids in different pH and temperature conditions. SAXS measurements were preformed on the X-33 EMBL beamline at DESY, Hamburg (Germany) using the Pilatus photon counting detector.

Using *ab initio* program DAMMIN [6] we created low resolution 3D models of native and mutated cystatin C in solution. HCC form a dimer in solution with elongated conformation as in tetragonal crystal form. The processes of formation of oligomers and fibrils were also monitored using SAXS in 1h time steps.

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**Keywords: human cystatin C, SAXS, amyloid**

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**Biological small angle scattering - A tool for gaining structural insight when other methods aren't enough**

Angela R. Criswell, Katsunari Sasaki, Mike Degen, Licai Jiang *Rigaku Americas Corporation, 9009 New Trails Dr., The Woodlands TX.* E-mail: angela.criswell@rigaku.com

As the number of protein structures increases so does the complexity of biological systems under study and the difficulty of studying more complex structural problems. Small angle X-ray scattering (SAXS) techniques have emerged as complementary tool for the structural biologist to extract structural information from biological systems. Though the comparative information gleaned from SAXS is of significantly lower resolution, SAXS methods offer an avenue to study macromolecules in near physiological conditions and to glean structural information where other methods, such as X-ray crystallography, fall short. In particular, SAXS has proven an ideal method to study macromolecules in partially disordered environments and to monitor structural changes, including small perturbations due to ligand binding or environmental.

Here we present data collected on the BioSAXS-1000 system, a small angle scattering system designed for macromolecular samples. These studies were aimed at identifying standard proteins for biological SAXS experiments and to evaluate good practices for collecting small angle scattering data on home laboratory systems. These results show that the BioSAXS-1000 system provides synchrotron-quality SAXS in as little as 15 to 30 minutes for most protein samples. The design inherits it strength from a combination of specially designed focusing optics coupled to a microfocus rotating anode and a high sensitivity hybrid pixel array detector. Unlike traditional Kratky systems, this system produces a point focused beam thus eliminating smearing issues common to traditional Kratky camera systems. This presentation will summarize the data collection and processing results for various standard samples collected on the BioSAXS-1000 system.

**Keywords: bioSAXS, small angle X-ray scattering, structural biology**

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**Towards the three-dimensional structure of human small heat-shock proteins B1 and B6**

Sergei V Strelkov,[a] Stephen D Weeks,[a] Ekaterina V Baranova,[a] Steven Beelen,[a] Michelle Heirbaut,[a] Olesya V Bukach,[b] Nikolai B Gusev,[b] *[a]Laboratory for Biocrystallography, Department of Pharmaceutical*