We will present dimeric $P6_1$ structures of "apo" *Ss*APRTase (with SO_4^{2-}) together with the complexes *Ss*APRTase:AMP (product) and *Ss*APRTase:ADP (inhibitor) based on ESRF (Grenoble) synchrotron data to about 2.4 Å resolution. The current work concentrates on obtaining a substrate analog complex with *Ss*APRTase. Large prisms have been obtained by co-crystallization and a $P2_12_12_1$ native data set to 2.0 Å has been recorded at the MAX-lab synchrotron (Lund). The substrate complex is not an easy molecular replacement task.

Keywords: adenine, phosphoribosyltransferase, archaea

FA1-MS01-P09

Crystal Direct : A new system for automatic crystal mounting and diffraction analysis. Florent Cipriani, José A. Márquez. *European Molecular Biology Laboratory, Grenoble Outstation, France.*
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The introduction of automated crystallization and data collection systems has had a significant impact in structural biology both by helping speed up the process of solving structures and enabling very challenging projects. At the same time this has resulted in a significant increase in the number of crystallization experiments performed and the number of crystals tested. However, recovering crystals from crystallization plates and mounting them on the supports used at data collection stations remains still a manual operation. Attempts to automate this step by emulating the manual mounting process with the use micromanipulators have been made, however this still requires significant human intervention. We have recently developed a new concept for protein crystallization and crystal recovery. This system is based on a new crystallization support that can be excised to recover the crystalline material. This system is also compatible with standard cryoprotection and other sample manipulations allowing X-ray diffraction measurements both at RT and under cryocooling conditions. Among the advantages of this new approach is the absence of mechanical stress for the crystals during the mounting process and that it facilitates handling of microcrystals, which can be challenging with standard methods. Moreover this system is designed to enable full automation of the crystal mounting process. The Crystal Direct system will be presented along with examples from a number of samples analyzed so far.

Keywords: Biological-Crystallography, High-througput-Crystallization, Automation, Crystal-mounting, Data-collection

FA1-MS01-P10

Incorporation of Extra-domain B into Fibronectin mediates local dimerization. André Schiefner, Michaela Gebauer, and Arne Skerra, *Department of Biological Chemistry, Technical University Munich, Emil-Erlenmeyer-Forum 5, 85350 Freising-Weihenstephan,Germany*
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Fibronectin (Fn) is an abundant and ubiquitous component of the extracellular matrix (ECM). It mediates a wide variety of cellular interactions, which are essential for cell adhesion, migration, proliferation, and differentiation. Many different cell lines synthesize and secrete Fn as a large glycoprotein consisting of repeating Fn type I, II, and III domains [1]. These domains comprise functional units that mediate interactions with other ECM components such as collagen and heparin, with cell surface receptors such as integrins, and with Fn itself. Fn is encoded by a single gene, which is spliced and expressed in a tissue-specific and developmental stage-dependent manner. The most prominent splice variants are observed in the type III set of domain repeats, termed EDA, EDB, and IIICS. While IIICS and EDA appear to be associated with Fn dimer secretion and cell differentiation, respectively, the function of EDB remains elusive. EDB gets incorporated between the domains Fn7 and Fn8 during embryogenesis, wound healing, and neoplastic vascularization but is absent in normal adult tissue. Thus, its presence serves as a marker for tumorigenesis that can be targeted by antibody fragments, which are currently subject to clinical studies [2]. In order to investigate the role of EDB on Fn structure, we determined the crystal structure of the four-domain Fn fragment Fn7-EDB-Fn8-Fn9 at 2.4 Å resolution. As previously described for Fn7-Fn8-Fn9-Fn10 [3], the individual FnIII domains are assembled in an extended rod-like structure. However, the relative orientations of Fn7-EDB and EDB-Fn8 are significantly different compared to Fn7-Fn8 in Fn7-Fn8-Fn9-Fn10. Unexpectedly and in contrast to the Fn7-Fn8-Fn9-Fn10 structure, Fn7-EDB-Fn8-Fn9 shows an, extended, anti-parallel contact between two Fn molecules in the crystal packing. This interaction is dominated by H-bonds and electrostatics with a total buried surface area of 1800 $Å^2$ for each molecule. Dimerization of Fn7-EDB-Fn8-Fn9 has been confirmed by size exclusion chromatography. Thus, our data suggest that the incorporation of EDB influences the affinity of Fn molecules for each other, which likely affects Fn mediated cell-cell adhesion.

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Keywords: fibronectin, alternative splicing, crystal structure

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PSPF - A protein sample production facility for structural biologists Anja Schütz[a], Joop van den Heuvel[b], Volker Jäger[b], Konrad Büssow[b], Dirk Heinz[b] and Udo Heinemann[a] [a]*Max Delbrück Center for Molecular Medicine, Berlin,German,* [b]*Helmholtz Centre for Infection Research, Braunschweig, Germany*
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The Protein Sample Production Facility (PSPF) was established in 2007 as the result of an international review of the major bottlenecks and needs for the structural biologists within the Helmholtz Association. The platform is dedicated to develop and provide infrastructure for the production of biomacromolecules for structural biology research. The decentralised facility is shared between the Helmholtz Centre for Infection Research (HZI) in Braunschweig and the Max-Delbrück-Centre (MDC) in Berlin. Since 2010 the PSPF is additionally integrated within the ESFRI project as an