MS03 Combining methods in macromolecular structure determination, including special conditions MX

Chairs: Dr. Martin Walsh, Dr. Victor Lamzin

MS03-P07

Scanning electron diffraction reveals the crystalline microstructure of Cellulose Nano Crystals

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All living plants contain cellulose as a crucial part of their structures. It possesses a complex structural diversity, including chirality. The combination of its fascinating structural features and its impressive properties in terms of specific strength and stiffness has triggered the engineering of cellulose-based materials with tailored mechanical and optical properties.

Scanning electron diffraction (SED) is an emerging method developed for the transmission electron microscope (TEM), following developments in electron optics and fast and sensitive detectors. By scanning a small electron probe, down to 1 nm in size, across the specimen and for each raster point a 2D electron diffraction pattern is acquired. In this way it is possible to build up a map revealing the ordering of the atoms in the sample with nanometer resolution. This method is opens up exciting opportunities for specimens, which are very beam sensitive and are difficult to study by other methods.

In this study, SED has been used to shed light on the microstructure of cellulose nanocrystals (CNCs) and to study the local ordering of the polysaccharide chains in the CNCs. Based on mapping of twisted CNCs it is possible to follow the ordering of the chains e.g. as the crystals are twisting.

Keywords: electron diffraction, cellulose, polymer

MS04 Biophysical characterization and crystallization

Chairs: Dr. Andrzej M. Brzozowski, Dr. Pavlina Rezacova

MS04-P14

Structural and biophysical characterisation of titin missense variants in genetic myopathies and cardiomyopathies

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Myopathies and cardiomyopathies are genetic conditions affecting skeletal and cardiac muscle. They are often caused by mutations in sarcomeric genes, such as TTN, which encodes the giant protein titin. Due to its size, TTN gene variants are also found in unaffected individuals (as shown by the 1000 Genomes Project) and it is difficult to assess their impact. In this project, biophysical techniques such as X-ray crystallography, differential scanning fluorimetry (DSF) and 1D Nuclear magnetic resonance (NMR) were applied to determine the structure of single and multiple titin A-band domains, in order to assess the impact of suspected and proven pathogenic variants on their stability and structure. Fibronectin type-3 domains from the titin A-band harbouring rare missense mutations were expressed in E. coli, both in wild-type (wt) and variant forms. AII wt's were confirmed folded by NMR studies, whilst some variants had structural changes induced by the missense mutation. Their DSF melting temperatures were lower by around 10°C, suggesting a reduction in stability caused by the mutation as a common feature of genetically proven pathogenic TTN variants. X-ray structural data elucidated the structural basis of the destabilization, allowing visualization of impact of the missense mutation on the surrounding residues and tertiary structure of the protein.

References:

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