EUniWell 3DEM Workshop Program

27th April, 2023

Individual talks are planned to take 20min + 10min of questions.

10:15h-10:30h (CET)	**Opening** (Antonio Martinez Sanchez)
10:30h-12:00h	**First Session: Challenging problems in Structural and Cell Biology** (chair Yves-Henri Sanejouard)
10:30h-11:00h Yves-Henri Sanejouard (University of Nantes)	**On the unknown structures of unknown proteins** Thanks to artificial intelligence algorithms like Alphafold we now have accurate 3D models for a wide variety of protein targets. However, there is still a lot of work for structural biologists. Noteworthy, for performing its predictions Alphafold relies on what is currently known at both the sequence and structural levels. I will illustrate this point by showing what Alphafold can tell about unknown proteins, that is, proteins with no known homologue.
11:00h-11:30h Olga Mayans (University of Konstanz)	**Molecular mechanoreceptors: elastic sensory components of the cell cytoskeleton** The cytoskeleton plays a crucial role in the generation and transmission of force in the living cell. Variations in its mechanical load trigger cellular responses that are deterministic of cell fate, such as proliferation, differentiation, apoptosis, resorption, and functional remodelling by hypertrophy or atrophy. Using striated muscle as model system, our laboratory studies the molecular mechanisms mediating the sensing of mechanical forces in the cell and their translation into intracellular signalling events. Our interest is to characterize protein components that mediate the communication of the contractile sarcomere with other cellular organelles in function of mechanical activity. Our ultimate goal is to understand how the dysfunction of this sensory system leads to cardiac and skeletal muscle disease. (Cardio)myopathy is closely linked to human demographics and of major socio-economical significance. In this research, we apply an integrative approach to structural biology that combines biochemical, biophysical, computational and structural methodologies, with emphasis on X-ray crystallography.
11:30h-12:00h María Senena Corbalán García & Jesús Baltanás Copado. (University of Murcia)	**In pursue of the 3D structure determination of the 135 kDa PKCα-Fascin1 complex by means of CryoEM.** Cancer is one of the leading causes of death worldwide. PKC is a family of highly homologous serine/threonine kinases involved in diverse cellular functions. Since the 1980s, many studies have shown they are involved in tumorigenesis, converting them into important targets for therapy. So far, this group of enzymes is a high potential target for anti-cancer drugs, but no successful approach is available up to date. The lack of structural 3D information of full-length PKCs is one of the big challenges we still face, and perhaps, one of the reasons why most of the PKC inhibitors entering clinical trials turn out to be ineffective. The aim of this project is to collect 3D structural information of PKCα in a complex with one of its substrates, Fascin1. The development of this challenging project will provide an invaluable tool to design new therapeutic strategies with an important impact on health, solving a very important puzzle in PKC-mediated signal transduction.
12:00h-13:00h	**Second Session: Cryo-Electron Microscopy Infrastructures** (chair Elmar Bhermann)
12:00h-12:30h Elmar Bhermann	**Cryo-EM in Cologne** Despite being one of the largest universities in Germany, the University of Cologne did not have access to high-resolution electron microscopy for a long time. In

(University of Cologne)	2017, funding from the DFG made it possible to establish a platform for molecular cryo-EM, which became operational in 2019. Since then, cryo-EM has contributed to both individual and collaborative research efforts, but the microscopes are not yet fully utilized. I will talk about the capabilities of our platform in Cologne, how we organize access to the infrastructure, and how it could be used in a larger research consortium.
12:30h-13:00h Marco Laurati (University of Florence)	**Cryo-EM infrastructure in Florence and recent applications in Soft Matter** I will present an overview of the instrumental infrastructure of the Florence Center for Electron Nanoscopy, funded in 2020. Beyond setups for Cryo-EM and Cryo-ET, a Ceta-D camera for microcrystal electron diffraction was recently installed on our instrument and is available for external users based on a proposal system. While the instrument design is optimized for structural biology studies, I will show results of Cryo-EM and Cryo-ET in the field of Soft Condensed Matter, concerning the investigation of lipid-based nanoparticles and self-assembled structures.
13:00h-14:30h	**Lunch Break**
14:30h-16:00h	**Third Session: Cryo-Electron Tomography** (chair Antonio Martinez-Sanchez)
14:30h-15:00h Marion Jasnin (Helmholtz Pioneer Campus Munich)	**Revealing the structural principles governing actin functions using in situ cryo-electron tomography** Actin contributes to an exceptionally wide range of cellular processes through the assembly and disassembly of highly dynamic and ordered structures. An integrated view of these different structures is needed to understand how distinct properties emerge from the same pool of accessory proteins, and how forces are generated and transmitted from molecules to cells. In recent years, cryo-electron tomography (cryo-ET) has become the method of choice for structural analysis of the cell interior at the molecular scale. In this talk, I will introduce the cryo-ET workflow and present our work on two actin-based force-producing cellular structures, cardiac sarcomeres and macrophage podosomes, and show how cryo-ET has begun to provide structural information about actin functions in cells across scales.
15:00h-15:30h Naoko Mizuno (NIH)	**Molecular Neurobiology by cryo-ET** Neurons are highly polarized cells forming an intricate network of dendrites and axons. They are shaped by the dynamic reorganization of cytoskeleton components and cellular organelles. Axon growth is controlled by the external stimuli. Axon branching allows the formation of new paths and increases circuit complexity. Finally, neuronal maintenance, the process of regeneration and degeneration are critical points for the remodeling of the circuit. However, our understanding of the neuronal morphogenesis is sparse at a molecular level due to the lack of direct in-depth observations. Using in situ cellular cryo-electron tomography on primary neurons, we aim to understand the molecular actions of the remodeling of organelles and cytoskeleton structures during the critical events of neuronal morphogenesis. In this talk, I will discuss about our recent molecular findings of neuronal regeneration.
15:30h-16:00h Antonio Martinez-Sanchez (University of Murcia)	**Computer methods for cryo-electron tomography: current challenges** The recent advancements in cryo-electron tomography (cryo-ET) hardware for data collection have enabled the production of a large number of tomograms. Consequently, a major bottleneck in cryo-ET is the lack of computer methods for productive processing of these massive datasets. In this presentation, I will make a brief review of the current state-of-the-art of computer methods for quantitative data analysis in cryo-ET, with a focus on the main challenges that need to be addressed.
16:00h-17:00h	**Round table**