

BBB seminar (BMED380)



Thursday, October 20, 14:30 at the BBB, Auditorium 4

Cryo-electron microscopy methods to explore the nano-universe of cells and molecules

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Cryo electron tomography (cryo-ET) provides us with possibilities to visualize the architecture and molecular machines driving bacterial growth and development. The bacterial cytoplasm and plasma membrane is a crowded environment, filled with biomolecules, with unique spatial organization and dynamic properties, we aim to envision. However, bacteria dimensions and the electron density of biomolecules under native conditions, makes the transmission electron microscopy signal to noise ratio less favorable for cryo-ET of bacterial specimen. Recent developments of cryo focused ion beam (cryo-FIB) preparation, correlative fluorescence and electron microscopy as well as genetically modifies mini-cells in combination with cryo-ET has shown great importance for visualization of cell volumes at near native state [1].

We use correlative approaches including cryo-ET to study the filamentous bacterium *Streptomyces coelicolor* intermediate filament like protein FilP. Bacterial intermediate filament like cytoskeleton proteins can be identified by their secondary and tertiary structural characteristics and chemical properties. FilP is biochemically homologous to the eucaryotic intermediate filament Lamin, but a genetic conservation cannot be confirmed. We found that purified FilP polymerize *in vitro* into thick, branched, repeatedly segmented filaments and networks with a 60 nm repetitive unit. We further studied the *in vitro* assembly of FilP and were able to build a 3D-model of the filament bundle structure [2]. The heteromorph appearance of FilP filaments, in analogy to intermediate filaments, makes ET and 3D reconstruction methods crucial to characterize the intracellular spatial organization.

References:

- [1] N. Söderholm, B. Singh, BE. Uhlin and L. Sandblad, *Curr Opin Struct Biol*, 23;64:166-173 (2020)
- [2] A. Javadi, N. Söderholm, A. Olofsson, K. Flärdh and L. Sandblad, *Life Sci Alliance*, 26;2(3) (2019)

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