Nanoscale Imaging of Neurotoxic Proteins

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Misfolding and aggregation of peptides and proteins is a characteristic of many neurodegenerative disorders, including Parkinson's Disease (PD) and Alzheimer's (AD). Their common feature is that normally unstructured and soluble proteins, misfold and aggregate into insoluble amyloid fibrils, which make up the deposits in the brains of patients suffering from these devastating illnesses. A key requirement to gain insight into molecular mechanisms of disease and to progress in the search for therapeutic intervention is a capability to image the aggregation process and structure of ensuing aggregates *in situ*.

In this talk I will give an overview of research to gain insight on the aggregation state of alpha synuclein (relevant to PD) beta-amyloid and Tau (relevant to AD) *in vitro* [1], in cells [2, 3] and in live model organisms [4].

In particular we wish to understand how these and similar proteins nucleate to form toxic structures and to correlate such information with phenotypes of disease [3]. I will show how direct stochastic optical reconstruction microscopy, *d*STORM, and multiparametric imaging techniques, such as spectral and lifetime imaging, are capable of tracking amyloidogenesis *in vitro,* see figure 1, and *in vivo*, and how we can correlate the appearance of certain aggregate species with toxic phenotypes [5].

Using multiparametric imaging methods we follow the trafficking of aggregates between cells and see how the misfolded state propagates from cell to cell. I will show how such information at the molecular level guides our understanding of disease pathology in humans.

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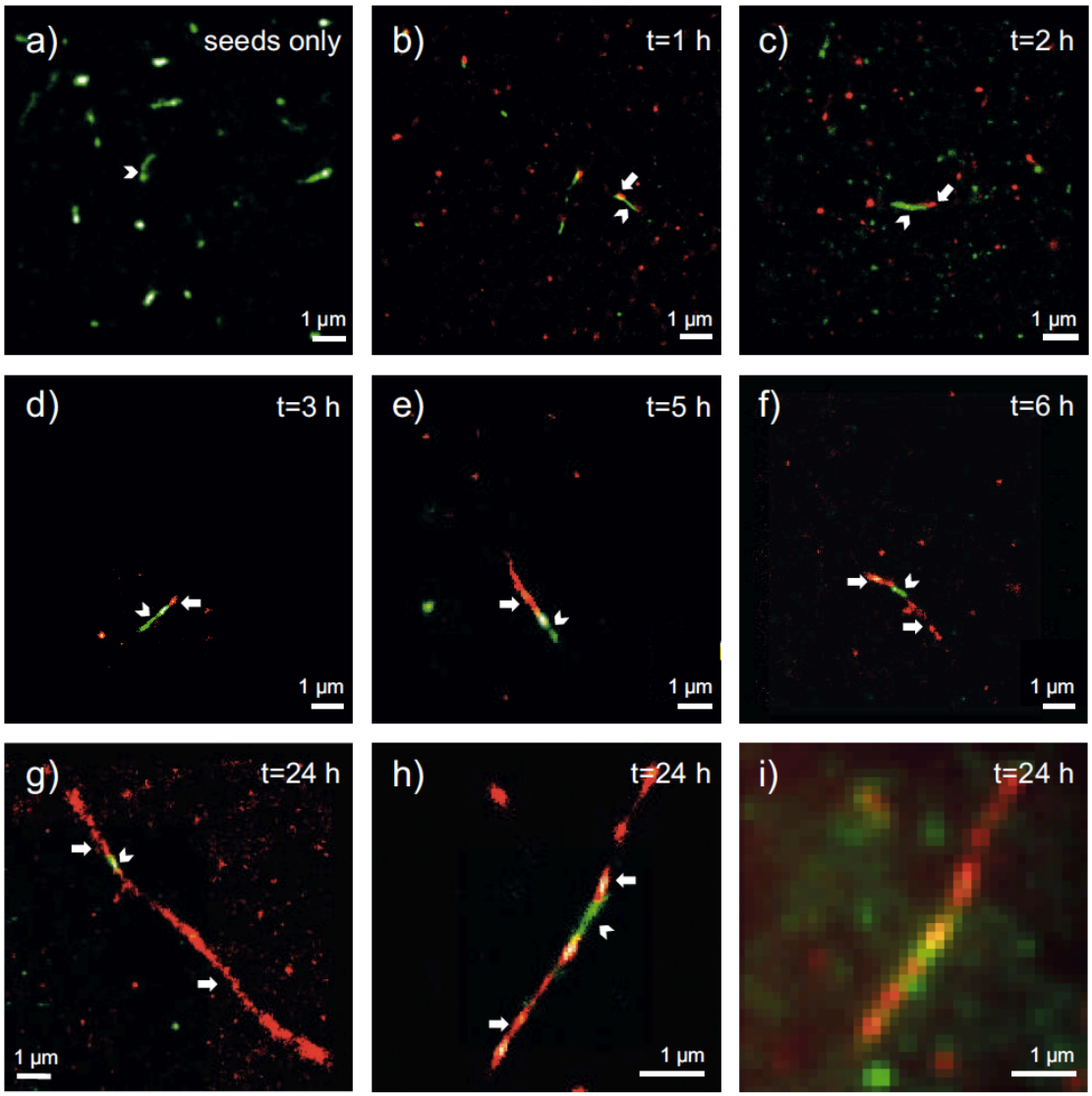

Fig. 1: α-synuclein elongation assay performed with dSTORM super-resolution microscopy. Small fibril seed species, shown in green, are incubated in solution containing monomeric α-synuclein, shown in red. α-synuclein seeds were covalently labelled with Alexa Fluor® 568 and monomer with Alexa Fluor® 647 dyes, respectively. The images show the time-sequenced growth of individual α-synuclein fibrils. Clearly, growth takes place from both ends of the seed fibril, extending to several micrometers in length after 24 hours. The last image shows a conventional fluorescence microscopy image, blurred by optical diffraction. Adapted with permission from27, copyright 2014 American Chemical Society.