Seminar/Talk

The Life of an Organelle - Studying Synaptic Vesicles Through Life-span Tracking and X10 Expansion Microscopy

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Host: Johann Georg Danzl

Synaptic vesicles are arguably the most well-characterized organelle model of cell biology. Their quantitative composition is known, the synaptic environment has been reconstructed, and their molecular interactions are characterized in near-atomic detail. However, like most organelles, synaptic vesicles are usually conceptualised as static, not changing their behaviour over time. The assumption is that synaptic vesicles are produced and trafficked to the synapse, where they are selected randomly for release/recycling until degradation ~4 days later. We tracked synaptic vesicles throughout this life-cycle, using live antibody-tagging and correlative metabolic imaging (nanoSIMS and FUNCAT), and found that only young vesicles release neurotransmitter, for ~12 hours. Combined pHluorin- and Ca2+-imaging further allowed us to determine that synaptic vesicle usage is limited to ~200 release events during their entire life-span, which is not exceeded even under increased demand. How is such a tight control of synaptic vesicle usage achieved? Super-resolution STED microscopy revealed that ageing synaptic vesicles become contaminated with SNAP25 from the cell membrane. On the vesicle, SNAP25 interferes with the quantitatively scarce CSP-alpha, hindering further release events. This inactivation is timed to precede damage accumulation on synaptic vesicles, likely to ensure that only pristine synaptic vesicles are employed in neurotransmission, to minimize damage-induced errors. This suggests that cells can track the functional age of organelles and manage their usage through stochastic timer mechanisms. In addition to this functional investigation of synaptic vesicles, I will present a new way to achieve multi-colour super-resolution imaging on conventional epifluorescence microscopes, using a refined version of expansion microscopy, termed X10. X10 allows a one-step expansion of biological samples by ~10-fold, achieving a resolution of ~25 nm, and will hopefully further advance the use of super-resolution microscopy in cell biology.

Monday, November 13, 2017 10:00am - 11:00am
Mondi Seminar Room 1, Central Building

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