Practical considerations when performing quantitative or super resolution microscopy

Abstract:
The combination of light microscopy and fluorescent reporters offers an unparalleled view into the function of intact cells. Recently, the scientific community has witnessed many major innovations in fluorescence microscopy that have paved the way for exciting new discoveries. These include the development of microscopy platforms that enable improvements in either throughput or quantification with high content imaging and analysis or in resolution through super resolution microscopy. In this two part seminar, we will discuss considerations when performing these exciting new approaches. In the first seminar we will present the advantage of high content imaging and analysis in terms of throughput, objectivity and ease of quantification. These advantages will be further highlighted using studies drawn from broad ranging areas such as stem cell biology or internalization and activity of antibody-drug conjugates. Applications of high content imaging and analysis to investigate neuronal function, differentiation and development will be shown. Finally we will show the use of high-content imaging and analysis in screening applications, with emphasis on screening siRNA libraries and phenotypic profiling of CRISPR edited cells. The second seminar will introduce the diffraction-limit with respect to fluorescence microscopy, followed by means to bypass the diffraction limit-so called ‘super resolution microscopy’ approaches. We will present the theory behind STED, SIM, single-molecule localization approaches (for example PALM, STORM, GSDIM) as well as fluorophore selection for each modality. Lastly, practical considerations when labeling samples for super resolution microscopy will be presented.

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