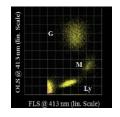


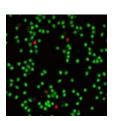
Metrology for Measurement and Monitoring of Immunity

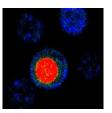
Rainer Macdonald

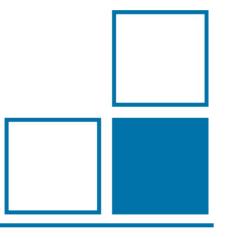
Fachbereich Biomedizinsche Optik rainer.macdonald@ptb.de











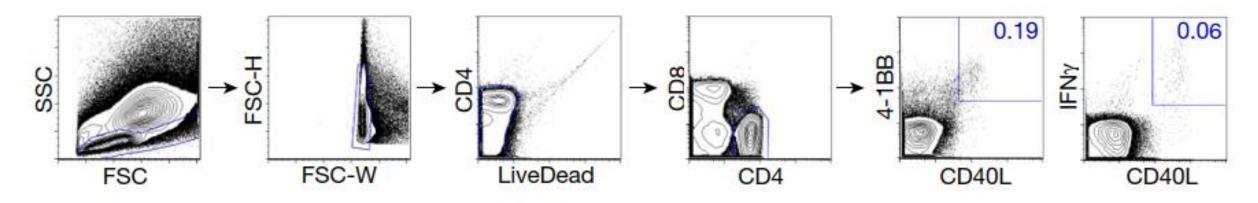
Cellular Phenotyping and Immunity



- Sensitive multiplex panel for target cells needed
 - Quantification of low expression levels (e.g. T cell activation)
 - Multi parameter analysis
- Complex Assay using in vitro stimulation necessary

Needs:

- ➤ Protocol for assay development and validation, incl. standardization, EQA
 - Antibody selection/validation
 - Guide to select fluorochromes and instrument settings
 - Absolute quantification



Braun et al. 2020 Nature 10.1038/s41586-020-2598-9

Measuring Antibody Binding Capacity

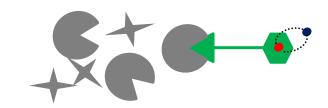


- Multicolor detection and characterisation of specific cell states and/or responses
- > Antibody Binding Capacity, protein expression level
 - quantification and localisation of antigenes, e.g.
 - HIV: changes in CD38 expression, indicator for progression
 - ❖ CLL: CD3/CD4 on T-cells, CD20/CD22 on B-cells
 - Also relevant for bead assays etc.
- ➤ Correlation with ID-MS → traceability

Calibration Free Antibody Quantification



absolute quantification by Xe NMR spectroscopy



- xenon exchange with biosensor detected by NMR
- hyperpolarization boosts detection sensitivity -> NMR competitive to optical assay
- \succ instrumentation independent results ightarrow high comparability
- no biological reference material needed

applications

- orthogonal quantitative approach
- > traceability due to traceable xenon solubility
- quantification when RM not available/quantification of RM