

## **“Cell signalling by image flow cytometry (AMNIS): an innovative tool to combine flow cytometry with microscopy”**

Cells of the innate and adaptive immune system work in concert to protect the body from pathogenic attack. The many different types of immune cells can be distinguished from one another not only by cell surface markers but also by their diverse cellular responses to different pathogens or stimuli. By combining quantitative image analysis tools with the large sample sizes common to flow cytometry, the ImageStream®xII uniquely enables multiple applications within the field of Immunology, including:

- measurement of nuclear translocation of transcription factors,
- T:APC interactions and accumulation of proteins at the immune synapse,
- chemokine-induced shape change, and
- phagocytosis.

In lecture and course we will introduce the basic principles of Image Stream flow cytometry technique and we will give an overview about different possible applications. In addition, the course will provide a hands-on training in acquiring different samples and analyzing the data with specific software. The “IDEAS®” software offers powerful tools for creating histograms and scatter plots. To each dot seen in a scatter plot one can see the corresponding cell's image. Numerical scoring of parameters such as size, shape, texture, colocalization and intensity allow quantification of visual data.

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### Literature:

- Khalil, A. M., Cambier, J. C., & Shlomchik, M. J. (2012). B cell receptor signal transduction in the GC is short-circuited by high phosphatase activity. *Science*, 336(6085), 1178-1181.
- Li, S., Dislich, B., Brakebusch, C. H., Lichtenthaler, S. F., & Brocker, T. (2015). Control of Homeostasis and Dendritic Cell Survival by the GTPase RhoA. *J Immunol*.
- Ling, G. S., Bennett, J., Woollard, K. J., Szajna, M., Fossati-Jimack, L., Taylor, P. R., et al. (2014). Integrin CD11b positively regulates TLR4-induced signalling pathways in dendritic cells but not in macrophages. *Nat Commun*, 5, 3039.