

Workshop

Title:	Live Cell Imaging of a translocation biosensor
Date, duration:	On demand, 1 day
Location:	Institute of Cell Biology, Baltzerstrasse 4, 3012 Bern
Lecturer(s):	Prof. Dr. Olivier Pertz (ICB) Benjamin Grädel (ICB)
Number of participants:	2 – 4 students
Target audience:	Master and PhD students of the University of Bern. Lecture Series on Advanced Microscopy plus exam (KSL 9256)
Registration:	Send request to Olivier Pertz olivier.pertz@izb.unibe.ch cc: Benjamin Grädel benjamin.graedel@students.unibe.ch
KSL:	471134
Reward:	0.5 ECTS
Costs:	100 CHF per student - PhD students enrolled in the Graduate School for Cellular and Biomedical Sciences (GCB) can apply for refund at the PhD program Cutting Edge Microscopy - Amount accounts for students of the University of Bern. Other participants, please request quote.
Learning goals:	Quantitative live cell imaging. High-throughput analysis of biosensor activity using ilastik, Cellprofiler and R.
Description:	Most of what is known about signalling pathways has been derived from population averages of a multitude of cells analysed by methods such as western-blot or proteomics. However, in recent years it became clear that population averages do not necessarily offer a complete picture of the pathway activity observed in cells. One way to study this is by analysing genetically-encoded fluorescence biosensors. These reporters come in a variety of types such as FRET or translocation sensors. Here, we will focus on a translocation sensor called ERK-KTR (ERK-kinase translocation reporter). With this method, the pathway activity of thousands of individual cells can be observed simultaneously and quantitatively by live cell fluorescence microscopy. Once the images have been acquired, automated image analysis allows us to extract single-cell trajectories of ERK activity from all the cells in a population. ERK, which is part of the mitogen-activated



protein kinase pathway, is a prime example in which population averages (as measured by western blot by example) occlude a large part of the picture. Recent studies have observed random pulses of ERK activity of conserved amplitude and duration in individual cells of a population. The frequency of these ERK pulses can specify if cells will die, survive or proliferate. In this course people will learn to time-lapse living cells that express the ERK-KTR biosensor. They will then learn how to apply automated image analysis to extract ERK activity trajectories in hundreds of single cells.

Course structure:

Morning: Students will learn how to acquire a time-lapse movie of cells expressing an ERK translocation biosensor
Afternoon: Students will learn how to perform automated image analysis to extract ERK activity fluctuations from the images (Cellprofiler, Ilastik, R)

Assessment:

pass/fail