

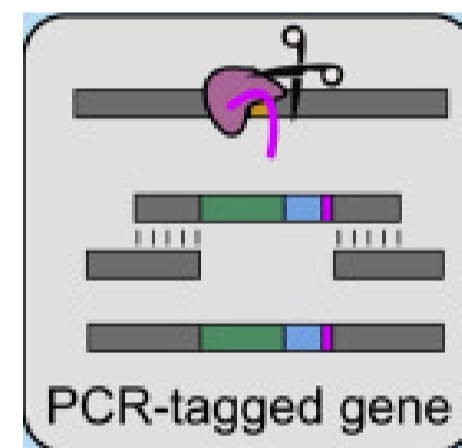
## Analysis of stable EGFR-mEos4b cell lines

### Motivation

The epidermal growth factor receptor (EGFR) belongs to the transmembrane receptor tyrosine kinase family and is involved in cellular processes like cell differentiation. It is involved in many types of cancer, e.g. if overexpressed or mutated. For studying the activation of EGFR upon ligand binding and its oligomeric state, single molecule localization microscopy methods like photoactivated localization microscopy (PALM) can be used. Quantitative PALM requires stoichiometric fusion of the protein of interest to a fluorescent protein. Genetic fusion on a genomic level can be accomplished by CRISPR/Cas editing.

### Task Description

We have used the recently developed CRISPR/Cas12a-assisted PCR tagging method for the generation of a stable cell line expressing EGFR tagged with the photoconvertible fluorescent protein mEos4b. Your task will be the analysis of clones generated with this method. For this you will work in the cell culture and use methods like PCR, DNA sequencing, western blot, and single particle tracking to study the tagging efficiency and the effect of the ligand binding on the mobility of EGFR.



### Key References

1. Ran et al. (2013) Nature Protocols 8, 2281, DOI: 10.1038/nprot.2013.143
2. Fueller et al. (2020) J. Cell Biol. 219, e201910210, DOI: 10.1083/jcb.201910210
3. Sauer and Heilemann (2017) Chem. Rev. 117, 7478, DOI: 10.1021/acs.chemrev.6b00667

### Work Area

Laboratory	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Microscopy	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Data Analysis	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Programming	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>

### Time

#### Possible Start

Sept. 2020

#### Duration

4 weeks

### Contact

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**Language** German, English