

Master thesis

Topic: "Application of *Tobacco etch virus* recombinant protease for the preparation of fusion proteins".

Background

The recombinant TEV protease is originally derived from *Tobacco etch virus* (TEV). It is a highly position-specific cysteine protease. Its sequence specificity makes it a very powerful reagent for removing fusion tags from recombinant proteins after protein purification. Its ease of extraction and robustness of use make it a widely used tool in protein biochemistry.

The aim of this work is to establish a workflow for the use of the TEV protease on the membrane proteins FoF1-ATP synthase and neurotensin receptor 1 that we use. Our research group studies conformations and modes of operation of individual proteins (membrane transporters, motors, receptors), e.g. FoF1-ATP synthases, G-protein coupled receptors, and others. To this end, we use and develop confocal microscopy methods for single-molecule Förster resonance energy transfer (FRET) methods. We analyze the conformational dynamics of these proteins '*in vitro*', with spatial resolution below a nanometer and temporal resolution below a millisecond. To maximize single molecule observation times in solution, we use 'single molecule traps' that counteract Brownian molecular motion in real time (ABEL trap).

Tasks

- Preparation and implementation of TEV protease expression using *Escherichia coli* on a laboratory scale
- Protein purification of TEV protease using affinity chromatography
- Optimization of reaction conditions for cutting TEV protease on fusion proteins
- Quantitative comparison between commercial and self-produced TEV protease.

Requirements

- advanced master's degree in biochemistry, biotechnology, bioanalytics or comparable studies
- first experience in a biochemical laboratory and with microbiological work
- basic methodological knowledge of recombinant protein expression, affinity chromatography and protein biochemistry
- very good language and writing skills in German or English
- motivation and ability to work independently and enjoy contributing own ideas

We offer

- small interdisciplinary team with close supervision and extensive experience
- practical application and deepening of the methods learned during the studies
- looking beyond the horizon: insight into current biophysical research in the field of fluorescence spectroscopy

If you are interested or have any questions, please email Prof. Dr. Michael Börsch. (michael.boersch@uni-jena.de).