Group of Proteases of Human Pathogens

"We offer a multidisciplinary approach to* in vitro *and cellular assays development and adaptation as well as testing compounds of interest with subsequent advanced analyses of interaction target protein – ligand."

Offer

Ibodies

iBodies are fully syntetic antibody mimetics based on pHPMA scaff old which is decorated with:

- small molecule targeting ligand
- affinity anchor and
- fluorescence labelling.

iBodies are useful various biochemical assays:

- ELISA
- surface plasmon resonance (SPR)
- flow cytometry
- fluorescence microscopy etc.

Know-how & Technologies

- We use iBodies for reveling binders/receptors/enzymes of biologically active compounds with unknown targets
- We set up the stochastic modification of those active small molecules to attach the handle to all possible sites on small mollecules, conjugate it to iBodies and after pull-down we identify proteins using mass spectrometry
- We routinely use iBody against GCPII (PSMA), FAP, aspartic proteases, His-tagged proteins and many others are in development

"From ligands of proteases to future first-class early lead compounds."

Main Capabilities

- Protein expression and purification
- Small molecule inhibitor design
- Assay development *in vitro* and *ex vivo*

Content of Research

- In the viral proteases research, we integrate the tools of classical biochemistry and structural biology with methods of cellular and molecular biology for in house development of *in vitro* and cell-based enzymological assays for search of early lead compounds with non-canonical mechanisms of action.
- In the field of proteases participating in cancer development, we characterized Prostate specific membrane antigen (PSMA) and fibroblast activated protein (FAP) in terms of 3D structure, substrate specificity and inhibitor design.
- We study viral proteases is focused mainly to HIV, Zika and Dengue NS2B-NS3 proteins as models for proteases autoactivation and activity modulation. We also study proteases involved in malignancies (PSMA, FAP and others).
- We develop novel assays for enzyme detection and characterization.

Key Research Equipment

Konvalinka's lab is a joint laboratory between the Department of Proteases of Human Pathogens at the Institute of Organic Chemistry and Biochemistry of the Academy of Science (IOCB) and Department of Biochemistry. Extensive set of instrumentation at IOCB is available to interested collaborators, including state of the art FPLC instrument, protein microcalorimetry, SPR, qPCR equipment, protein crystallization and others.

Partnerships & Collaborations

Academic Partners

- Hans-Georg Kräusslich: Department of Virology, University of Heidelberg, Heidelberg, Germany (In vitro analysis of polyprotein processing, culture testing of protease inhibitors)
- Alex Wlodawer: National Institutes of Health, Bethesda, USA (Structural biology of HIV PR and its mutants)
- Monique Nijhuis: Eijkman-Winklar Centre for Medical Microbiology, Infectious Diseases and Inflammation

Private and Public Sector

- AIDS Center, Clinic of Infectious Diseases, Faculty Hospital Bulovka: Long term clinical study on the development of resistance against antiretroviral treatment in HIV positive patients

Main Projects

- **2012–2018**: Controlling Structure and Function of Biomolecules at the Molecular Scale: Theory Meets Experiment. Grant Agency of the Czech Republic; project P208/12/G016 (principal coinvestigator)
- **2012–2014**: Glutamate carboxypeptidase II and its role in tumor development. Grant Agency of the Czech Republic; project P304/12/0847 (principal investigator)
- **2013–2015:** Assembly of HIV virions as a therapeutic target. Grant Agency of the Czech Republic; project GA13-19561S (principal investigator)

Achievements

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- Schimer J., Pávová M., Anders M., Pachl P., Šácha P., Cígler P., Weber J., Majer P., Řezáčová P., Krausslich H.G., Muller B., Konvalinka J. (2015) **Triggering HIV polyprotein processing by light using rapid photodegradation of a tight-binding protease inhibitor.** Nature Communications 6: 6461
- Tykvart J., Bařinka C., Svoboda M., Navrátil V., Souček R., Hradílek M., Šácha P., Lubkowski J., Konvalinka J. (2015) **Structural and biochemical characterization of human N-acetylated alpha-linked acidic dipeptidase-like (NAALADase L) protein, a novel aminopeptidase from human intestine.** Journal of Biological Chemistry 290: 11321–11336
- Tykvart J., Schimer J., Jančařík A., Bařinková J., Navrátil V., Starková J., Šrámková K., Konvalinka J., Majer P., Šácha P. (2015) **Design of Highly Potent Urea-Based, Exosite-Binding Inhibitors Selective for Glutamate Carboxypeptidase II.** Journal of Medicinal Chemistry 58: 4357–4363

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