Laboratory of Signal Transduction

Plasma membrane signalosomes, immunoreceptor signalling, mast cell activation, chemotaxis

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The main research interest of our laboratory has been traditionally focused on understanding molecular mechanisms governing signal transduction from the high affinity IgE receptor (FcεRI) and other plasma membrane receptors to the cytoplasm. We continued in functional studies on the role of transmembrane adaptor proteins in regulation of such important antigen-mediated events as calcium response, degranulation and chemotaxis. Especially, we attempted to solve the problem we noticed in our previous studies that NTAL has a negative regulatory role on FcεRI activation in mast cells isolated from NTAL knockout mice, but positive regulatory role in human mast cells with NTAL knockdown (through RNA interference). We found that the observed discrepancies did not reflect compensatory developmental alterations in mouse cells, as expected, but rather different roles of NTAL in FcεRI-induced signalling pathways in human and mouse cells. To gain more insight into NTAL function, we also examined gene expression profiles in resting and antigen-activated mast cells with NTAL knockout and knockdown and corresponding controls and we identified several genes that were differentially expressed in NTAL deficient and wild-type cells. We also continued in studies on stromal interacting molecule 1 (STIM1) in mast cell signalling and found that STIM1 is required for formation of microtubule protrusions in antigen-activated cells and for chemotaxis towards antigen.

Several other molecules were analysed (Csk, PAG, STAT5, CD9, ORMDL3, FcεRI) to get better insight into the signalling pathways involved in mast cell signalling. Significant effort was devoted to identifying new signalling molecules by high-throughput screening using RNA interference libraries.

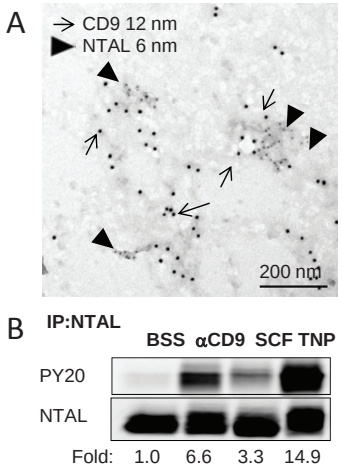

Fig. 1. CD9-NTAL crosstalk. A. Membrane topography of tetraspanin CD9 and transmembrane adaptor protein NTAL on isolated plasma membrane sheets as detected by electron microscopy. B. phosphorylation of NTAL after activation of mast cells with CD9-specific antibody, stem cell factor (SCF) and antigen (TNP) as detected by immunoblotting of immunoprecipitated NTAL.

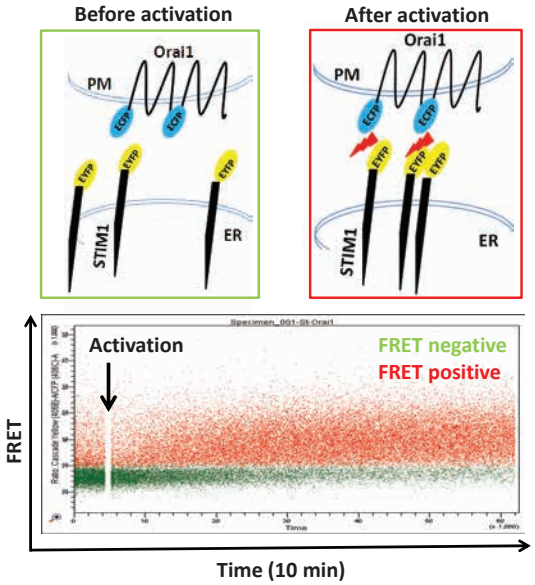

Fig. 2. Dynamics of STIM1-Ora1 crosstalk. Interactions between STIM1 and Orai1 were determined by fluorescence resonance energy transfer (FRET). FRET (in red) occurs between STIM1-EYFP and Orai1-ECFP in cells after activation with thapsigargin that releases calcium from endoplasmic reticulum. FRET is determined by flow cytometry.

- AS CR, M200520901 - New components of immunoreceptor signaling in mast cells, 2009-2012, Pe. Dráber
- AV CR, KAN200520701 - Nano-PCR - ultrasensitive test for detection of specific proteins in body fluids, 2007–2011, Pe. Dráber
- GA CR, GA301/09/1826 - Topography and function of Csk-binding proteins of the plasma membrane in mast cells, 2009-2013, Pe. Dráber
- GA CR, GAP302/10/1759 - Function-structure relationships between transmembrane adaptor-based signalosomes in mast cells, 2010-2013, L. Dráberová
- Ministry of Industry and Trade of the Czech Republic, FR-TI3/067 - Genetically modified polymerases and their use for amplification of unpurified DNA, 2011-2014, Pe. Dráber
- TA CR, TA01010436 - New generations of DNA aptamers, 2011-2013, Pe. Dráber, L. Dráberová
- Ministry of Education, Youth and Sports of the Czech Republic, LD12073 LD-COST CZ - Membrane signalosomes of mast cells and basophils - targets for innovative therapies, 2012-2014, P. Dráber

1. Rashid A, Housden JEM, Helm BA, Draber P. Fc receptor- γ subunits with both polar or non-polar amino acids at position of T22 are capable of restoring surface expression of the high-affinity IgE receptor and degranulation in γ subunit-deficient rat basophilic leukemia cells. **Mol Immunol 2012** 53(3): 270-273.
2. Halova I, Draberova L, Draber P. Mast cell chemotaxis – chemoattractants and signaling pathways. **Front Immunol 2012** 3: 119.
3. Hájková Z, Bugajev V, Dráberová E, Vinopal S, Dráberová L, Janáček J, Dráber P, Dráber P. STIM1-directed reorganization of microtubules in activated mast cells. **J Immunol 2011** 186(2): 913-923.
4. Horáková H, Polakovičová I, Shaik GM, Eitler J, Bugajev V, Dráberová L, Dráber P. 1,2-propanediol-trehalose mixture as a potent quantitative real-time PCR enhancer. **BMC Biotechnol 2011** 11: 41.
5. Potůčková L, Franko F, Bambousková M, Dráber P. Rapid and sensitive detection of cytokines using functionalized gold nanoparticle-based immuno-PCR, comparison with immuno-PCR and ELISA. **J Immunol Methods 201**1 371(1-2): 38-47.

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