**INVITED SPEAKER**

# REGULATION OF SMALL GTPASE SIGNALING AT THE PERIPHERY OF MEMBRANE

*M. ZEGHOUF [1], J. CHERFILS [1], A. NAWROTEK [1]*

*[1]CNRS-ENS Paris-Saclay - Gif-Sur-Yvette (France)*

How do circuits of protein enable cells to monitor information from their environment and to make decisions to mount efficient responses? Small GTPases are central players in the regulation of cellular signaling thanks to their ability to function as molecular switches that alternate between an inactive and an active state, in response to received signals. This apparently simple mechanism is in reality extremely complex. Multiple signals converge towards each small GTPase through its numerous activators (guanine nucleotide exchange factors or GEFs, which stimulate their GTP loading) to generate a unique response by selecting specific effectors. The level of active GTPases is balanced by the inhibitory activities of their GTPases-activating proteins (or GAPs), which inactivate them by stimulation of GTP hydrolysis. The regulation of cellular signaling by GTPases occurs at the periphery of membranes, to which small GTPases anchor by lipidic modifications, and regulators and effectors associate by membrane-binding domains. Deregulation of any member of the small GTPase machinery leads to diseases including cancers or developmental diseases. In that context, we are interested in how multi-protein platforms assemble on membranes and regulate the level of activation and the specificity of small GTPase signaling. We show that the protein-membrane interaction is determinant for GEF [1] and GAP activities and could remodel intramolecular interactions to prime for full activity toward small GTPase [2]. We also demonstrate that the protein-membrane interaction is an attractive target for inhibition of small GTPase signaling in cancers [3,4]. Taking together our results decipher the regulatory network that control the signaling output and reveal an innovative strategy to modulate small GTPase activation.

1. Karandur, D., Nawrotek, A., Kuriyan, J. & Cherfils, J. Multiple interactions between an Arf/GEF complex and charged lipids determine activation kinetics on the membrane. Proc Natl Acad Sci U S A 114, 11416-11421 (2017).

2. Das, S. et al. Structural Organization and Dynamics of Homodimeric Cytohesin Family Arf GTPase Exchange Factors in Solution and on Membranes. Structure 27, 1782-1797 e7 (2019).

3. Nawrotek, A. et al. PH-domain-binding inhibitors of nucleotide exchange factor BRAG2 disrupt Arf GTPase signaling. Nat Chem Biol 15, 358-366 (2019).

4. Nawrotek, A., Zeghouf, M. & Cherfils, J. Protein-membrane interactions in small GTPase signalling and pharmacology: perspectives from Arf GTPases studies. Biochem Soc Trans 48, 2721-2728 (2020).