Mechanism of Snf1/AMPK autoregulation linked to subcellular localization in *Saccharomyces cerevisiae*

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In the budding yeast *Saccharomyces cerevisiae*, protein kinase Snf1/AMPK is a heterotrimeric complex made by the catalytic α subunit Snf1, a regulatory β subunit (alternatively Gal83, Sip1 and Sip2, involved in the modulation of its subcellular localization) and the γ subunit Snf4. As in mammalian cells, in low glucose concentration or upon stress, Snf1/AMPK complex is activated through phosphorylation of the α subunit, by one of the three upstream kinases Sak1, Tos3 and Elm1, among which Sak1 is the major one. On the other side, in response to high glucose concentrations Snf1/AMPK is inactivated through dephosphorylation by the PP1 protein phosphatase Glc7/Reg1 (1). These regulatory events are accompanied by changes in the nucleo-cytoplasmic distribution of the complex, as Snf1/AMPK operates within distinct subcellular pools, each specialized for unique regulatory functions.

Using a phospho-proteomic approach, we reported that Snf1/AMPK phosphorylates multiple proteins belonging to its own regulatory complex (2). We generated mutants in Snf1-dependent phospho-sites of Sak1 and Reg1 and by combining nucleo-cytoplasmic fractionation and microfluidic analysis with single-cell live imaging we identified new mechanisms which regulate Snf1 localization as a function of to its activity under low glucose conditions. Our findings highlight that Snf1 is able to regulate its own subcellular localization, which is critical for fine-tuning its activity and downstream signaling during metabolic stress. This localization-driven regulatory mechanism involves Sak1-mediated phosphorylation, which promotes Snf1 activation and nuclear enrichment, counterbalanced by cytosolic Reg1-dependent dephosphorylation to maintain proper Snf1 distribution and signaling.

(1) Coccetti et al., 2018 Microbial Cell, doi: 10.15698/mic2018.11.655.
(2) Caligaris et al., 2023 eLife, doi: 10.7554/eLife.84319.