M2 intership offer

Effet of rate increase of osmotic stress on yeast growth and density

Context

Cells are naturally confined by their environment. Cell proliferation in a spatially-limited environment leads to the progressive emergence, at the population level, of growth-induced pressure. This growth-induced pressure is generated by cells through the accumulation of intracellular osmolytes, at a rate that matches cell growth. The typical response to growth-induced pressure is a reduction of cell proliferation, through the concomitant accumulation of macromolecules, in parallel to osmolytes generating pressure: the accumulation of macromolecules increases intracellular density and viscosity, decreasing macromolecule diffusion, and thus limiting reaction rates, and ultimately growth [1]. This viscosity change can be measured by the diffusion of nanoparticles, called GEMs [2].

Growth-induced pressure has a lot of similarities with osmotic stresses. An instantaneous osmotic stress leads to the decrease in cell volume through water efflux, leading to an increase in intracellular density and crowding, measured by a decreased diffusion of GEMs. During an osmotic stress, yeast cells initiate an adaptive osmoregulation through the MAPK Hog1, which triggers both directly and transcriptionally the production of intracellular osmolytes. This production leads to cell swelling, and recovery of intracellular density.

The Hog1 pathway activation under osmotic stress depends not only on the magnitude but also on the rate of stimulation. When osmotic pressure gradually increases, Hog1 pathway initiation is delayed or does not even occur [3]. This is reminiscent of the fact that cells do not exploit specific stress response pathways during growth-induced pressure development. The question arises here about how cells achieve osmotic equilibrium without Hog1-mediated osmoadaptation under slow increases in osmostress: *Does the cell viscosity gradually increase as well? Or does the slow stimulation rate allow the cell enough time to secure another osmolyte pool, such as amino acids supply from protein degradation and ions from the outside?*

Objectives of this internship

The goal of this project is to study how the rate of osmotic stress increase, in both Hog1-GFP and Hog1 KO mutants, impacts cell growth and viscosity. We will inject and mix both iso-osmotic and a hyper-osmotic media inside a microfluidic device through the use of syringe pumps to control the mixing rate and thus the rate of osmotic stress, and evaluate optically Hog1 response, cell growth, and intracellular density change.

How to postulate

For this essentially experimental internship, we are primarily looking for a candidate with knowledge in cell biology and/or microfluidic, imaging. The candidate must have a strong will to work at the interface between physics and biology. The student will be trained in microbe cell culture and imaging, as well as microfabrication principles.

For more information or to apply, please send your motivation letter and a CV to hyojun.kim@laas.fr morgan.delarue@laas.fr.

[1] B. Alric, C. Formosa-Dague, E. Dague, L. J. Holt, and M. Delarue, “Macromolecular crowding limits growth under pressure,” *Nature Physics,* 2022.

[2] M. Delarue *et al.*, “mTORC1 Controls Phase Separation and the Biophysical Properties of the Cytoplasm by Tuning Crowding,” *Cell*, 2018.

[3] A. N. Johnson *et al.*, “A rate threshold mechanism regulates MAPK stress signaling and survival,” *Proceedings of the National Academy of Sciences*, 2020.