A potential growth rate sensing mechanism in budding yeast

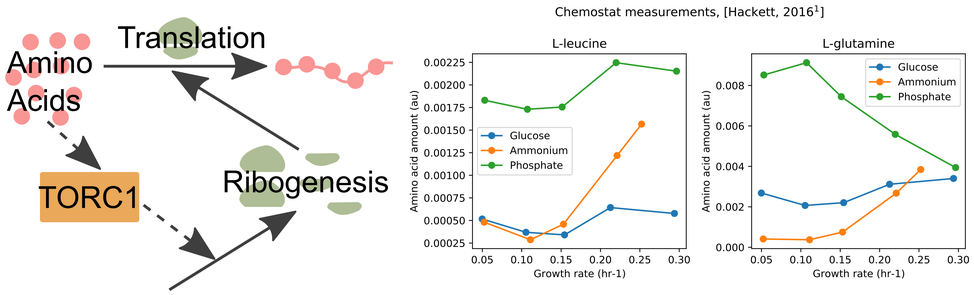
Amogh Prabhav Jalihal[1], Julius Palme[1], Michael Springer[1]

[1]Department of Systems Biology, Harvard Medical School, Boston

Background

TORC1 the global regulator of growth in eukaryotes senses intracellular amino acid concentrations to coordinate growth rate. We expect concentration sensing to pose two drawbacks: 1. Activation of TORC1 signaling causes an increase in cellular translation, which is expected to change the intracellular amino acid levels, thus affecting the relationship between nutrient state and growth control. 2. A change in nutrient sensor levels will affect the strength of the signal conveyed to TORC1. If TORC1 instead sensed the magnitude of amino acid flux into translation, it could achieve robust growth control. Based on evidence from the GAL pathway, we propose that TORC1 can achieve flux sensing via the leucine sensor Cdc60. In preliminary work, we have built a toy model to construct experiments that can prove this hypothesis.

Concentration sensing model



Model

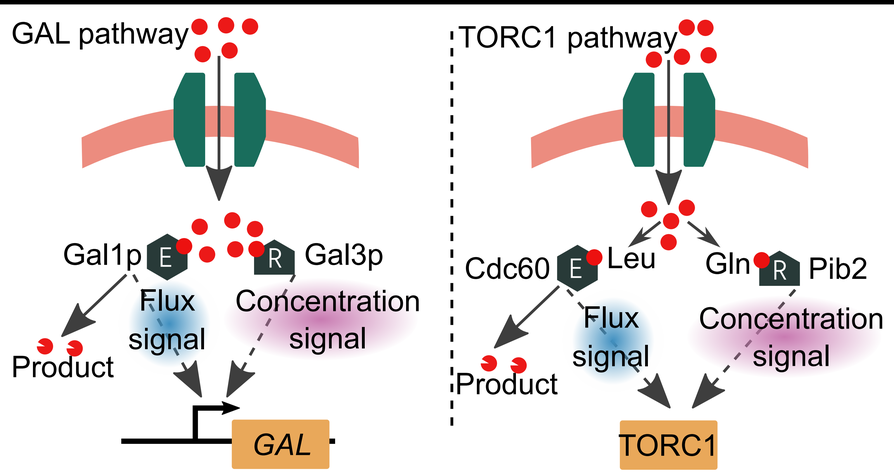
We model the accumulation of nutrients inside the cell N_i and track the response of a flux sensor and a concentration sensor. **An enzyme that signals when it is catalytically active can signal flux through a reaction**

$$\frac{dN_i}{dt} = (J_{in} - J_{out}) - \frac{vEN_i}{k_m + N_i}$$

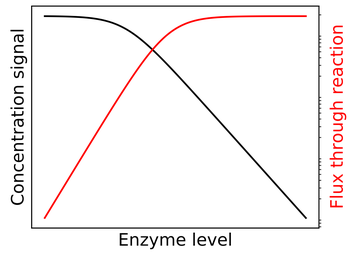
$$S_{\text{flux}} = \frac{v_e E N_i}{k_m + N_i}, E \quad \text{is the enzyme}$$

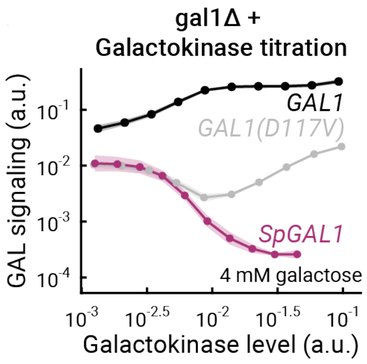
$$S_{\text{conc}} = \frac{v_c R N_i}{k_{mc} + N_i}, R \quad \text{is the receptor}$$

GAL and TORC1 pathways



Enzyme titration[2]



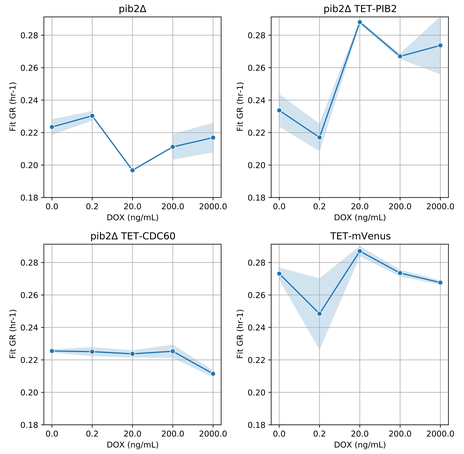


Hypothesis

TORC1 has two well characterized nutrient sensors: Cdc60 (leucine tRNA synthetase), and Pib2 (glutamine sensor). Cdc60 only signals to TORC1 during the aminoacylation reaction.

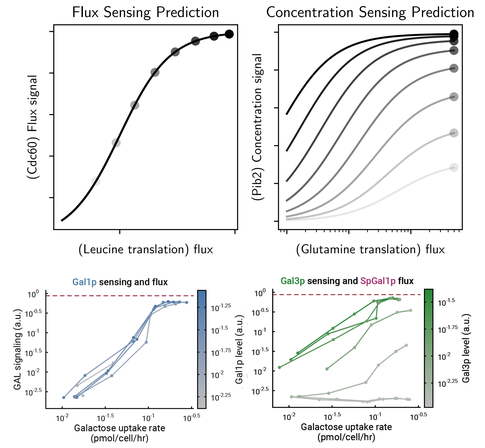
This mechanism provides a basis for the hypothesis that Cdc60 might function as a flux sensor. [4]

Sensor titrations

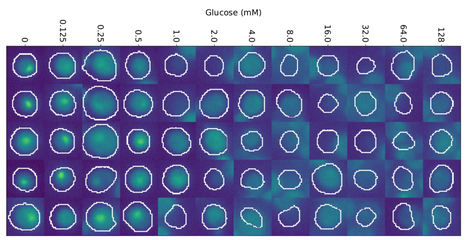


Overexpressing a concentration sensor is expected to increase TORC1 signaling.

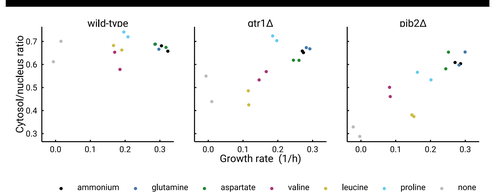
Flux-Sensor[2]



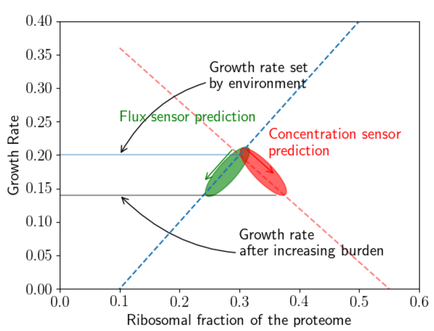
Stb3 localization



Growth rate sensing



Properties



Challenges

Distinguishing flux and concentration sensing requires measuring intracellular amino acids concentrations and flux into translation.

TORC1 signaling directly controls ribosomal expression. Measuring the ribosomal fraction of the proteome is limited in throughput.

Cdc60 is an essential gene. Since tRNA synthetases are highly conserved, species allele swaps will have to be tested for functionality in budding yeast.

Future Plans

Use carbon, nitrogen limited chemostats to characterize growth rate sensing.

Compare other transcription repressors, validate with established readouts.

Test FRET sensors to monitor leucine and glutamine concentrations in real time.

Summary

TORC1 mediated growth control is expected to be unstable if it senses intracellular amino acid concentrations. Cdc60 can sense flux of leucine into translation, and might serve as a readout for cellular growth rate.

References

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[2] Palme, J., Li, A., Springer, M. *Unpublished*

[3] Scott, M., (2010). Interdependence of cell growth and gene expression: origins and consequences. Science, 330(6007), 1099âĂŞ1102.

[4] Bonfils, G. (2012). Leucyl-trna synthetase controls torc1 via the ego complex. Molecular Cell, 46(1), 105âĂŞ110.

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Email: amogh_jalihal@hms.harvard.edu

Website: https://springerlab.org/research/