Frequency-Modulated Localization Bursts Coordinate Gene Regulation

Long Cai, Chiraj Dalal, and Michael Elowitz

***Short Abstract* — In yeast, the transcription factor Crz1 is dephosphorylated and translocates into the nucleus in response to extracellular calcium. Using time-lapse microscopy, we found that Crz1 exhibited short bursts of nuclear localization (~2 minutes) that occurred stochastically in individual cells and propagated to the expression of downstream genes. Strikingly, calcium concentration controlled the frequency, but not duration, of localization bursts. Using an analytic model, we found that this frequency modulation (FM) of bursts ensures proportional expression of multiple target genes across a wide dynamic range of expression levels, independent of promoter characteristics. We experimentally confirmed this theory with natural and synthetic Crz1 target promoters. Another stress response transcription factor, Msn2, exhibits similar, but largely uncorrelated, localization bursts under calcium stress. These results suggest that FM regulation of localization bursts may be a general control strategy utilized by the cell to coordinate multi-gene responses to external signals.**

***Keywords* — Nuclear Localization Bursting, Frequency Modulation, Stochasticity, Coordination of Gene Expression.**

I. Purpose

Cells respond rapidly to many stresses by post-translationally modifying transcription factors and mobilizing them to the nucleus, where they can activate the expression of a multitude of target genes [1-3]. In the budding yeast, *Saccharomyces cerevisiae,* extracellular calcium stress affects the activity of Crz1, the calcineurin-responsive zinc-finger transcription factor, through phosphorylation and dephosphorylation [4], causing changes in its nuclear localization, rather than its abundance However, it has remained unclear how the post-translational regulation of this transcription factor encodes extracellular calcium levels, and how it operates dynamically at the single-cell level to control expression of the more than 100 genes necessary for calcium adaptation [5]. To address these issues, we acquired time-lapse movies of Crz1 localization dynamics, using a strain in which the Crz1 protein was tagged with GFP [6].

II. Results

We observed that, in individual cells, Crz1 responded to step changes in the extracellular calcium concentration with rapid and intense nuclear localization burst that occurred stochastically across the population.

A. Nuclear localization bursts are Frequency Modulated

Increase in calcium concentration resulted in increases in only the burst frequency, and not the burst durations. Thus, cells employ Crz1 localization bursts in a Frequency Modulated (FM) fashion to encode and respond to extracellular calcium. This FM response is propagated to downstream target genes.

B. Localization bursts coordinate downstream genes

We observed transcriptional bursting of Crz1 target genes following Crz1 localization bursts. We showed that FM regulation of nuclear localization bursts allow transcription factors to modulate the expression of multiple target genes in concert, keeping their relative abundances fixed over a wide dynamic range, regardless of the shapes of their input functions. This mode of regulation may be applicable beyond transcriptional networks to the general problem of coordination in cells.

III. Conclusion

We observed similar bursting behavior with Msn2 and Mig1 proteins, suggesting that FM regulation may be broadly employ by cells to encode and coordinate responses to external signals.

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Division of Biology, California Institute of Technology, Pasadena, CA USA. E-mail: lcai@caltech.edu