Molecular Buffers Permit Sensitivity Tuning and Inversion of Riboswitch Signals

Predictable integration of foreign biological signals and parts remains a key challenge in the systematic engineering of synthetic cellular actuations, and general methods to improve signal transduction and sensitivity are needed. To address this problem we modeled and built a molecular signal buffer network in Saccharomyces cerevisiae inspired by chemical pH buffer systems. The molecular buffer system context-insulates a riboswitch enabling synthetic control of colony formation and modular signal manipulations. The riboswitch signal is relayed to a transcriptional activation domain of a split transcription factor, while interacting DNA-binding domains mediate the transduction of signal and form an interacting molecular buffer. The molecular buffer system enables modular signal inversion through integration with repressor modules. Further, tuning of input sensitivity was achieved through perturbation of the buffer pair ratio guided by a mathematical model. Such buffered signal tuning networks will be useful for domestication of RNA-based sensors enabling tunable outputs and library-wide selections for drug discovery and metabolic engineering.

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