Engineering an NADPH/NADP⁺ Redox Biosensor in Yeast

Genetically encoded biosensors have emerged as powerful tools for timely and precise in vivo evaluation of cellular metabolism. In particular, biosensors that can couple intercellular cues with downstream signaling responses are currently attracting major attention within health science and biotechnology. Still, there is a need for bioprospecting and engineering of more biosensors to enable real-time monitoring of specific cellular states and controlling downstream actuation. In this study, we report the engineering and application of a transcription factor-based NADPH/NADP⁺ redox biosensor in the budding yeast Saccharomyces cerevisiae. Using the biosensor, we are able to monitor the cause of oxidative stress by chemical induction, and changes in NADPH/NADP⁺ ratios caused by genetic manipulations. Because of the regulatory potential of the biosensor, we also show that the biosensor can actuate upon NADPH deficiency by activation of NADPH regeneration. Finally, we couple the biosensor with an expression of dosage-sensitive genes (DSGs) and thereby create a novel tunable sensor-selector useful for synthetic selection of cells with higher NADPH/NADP⁺ ratios from mixed cell populations. We show that the combination of exploitation and rational engineering of native signaling components is applicable for diagnosis, regulation, and selection of cellular redox states.