p53 pulses diversify and coordinate target gene expression

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***Short Abstract* — Cells employ complex systems to sense environmental conditions and execute appropriate responses. In response to DNA double-strand breaks, p53, a tumor-suppressing transcription factor, is expressed in a series of pulses. These pulses influence the fate of the cell, but their mechanism of action is unclear. We show that p53 pulses give rise to different target gene expression profiles, which can be predicted by the genes' mRNA decay rates, consistent with a mathematical model. Furthermore, we show that p53 coordinates expression of certain target genes and that this coordination changes as the DNA damage response progresses.**

***Keywords* — p53, pulsing, dynamics, signal transduction, DNA damage response, gene expression**

I. Introduction

As methods for observing and quantifying intracellular signaling improve, we can increasingly appreciate that cells transmit information in the dynamics (changes in time) of molecules involved in signaling [1]. In particular, several recently discovered signaling pathways contain components that pulse in time [2,3]. Pulsing is believed to serve different purposes in different contexts, including coordinating gene expression, keeping track of time, generating diverse patterns of gene expression or phosphorylation, and improving signal-to-noise ratios in information transmission.

The tumor suppressor protein p53 pulses as part of its response to DNA double-strand breaks [4]. Since p53 is a transcription factor, regulating over 100 genes [5], this pulsing potentially impacts numerous downstream processes, including apoptosis, cell cycle control, DNA repair, and metabolism. p53 pulsing is ultimately linked to cell fate, as suppressing pulsing pharmacologically while keeping p53 at a constant high level leads to changes in cell fate patterns [6]. The direct mechanistic consequences of p53 pulsing, however, are unknown. Here we investigated two hypotheses about what p53 pulsing accomplishes mechanistically.

II. p53 Pulsing Diversifies Target Gene Dynamics

We had previously proposed that p53 pulsing could enable a wider range of target gene expression dynamics than would be possible if p53 were raised to a constant high level [4]. To test this, we treated MCF-7 breast carcinoma cells with a drug to induce DNA double-strand breaks, then measured expression of 93 p53 target genes over a 10-h period. We found that target gene expression profiles clustered into distinct groups, including those which pulsed with p53 and those which simply rose in response to p53. Moreover, we found that a gene's mRNA decay rate was a significant predictor of whether its expression would pulse or rise, consistent with a simple mathematical model of target gene activation by a transcription factor. These findings suggest that each target gene acts as a low-pass filter for p53 pulses, tuned by its mRNA decay rate.

III. p53 Pulsing Coordinates Target Gene Expression

We also investigated whether p53 coordinates expression of its target genes, similar to the pulsing transcription factor Crz1 in yeast [7]. We performed single-cell transcriptional profiling on cells treated to induce DNA double-strand breaks, then looked for correlations in the expression of pairs of genes. This revealed two coordinated subsets of p53 target genes. One subset was p53-independent and largely composed of DNA repair genes; these genes maintained correlation throughout the DNA damage response. The other subset was p53-dependent and largely composed of genes with pulsing dynamics. The "pulsing" subset peaked in correlation after the third p53 pulse, at which time it became negatively correlated with the "repair" subset.

IV. Conclusion

Our results suggest that p53 pulsing generates diversity in target gene dynamics, enabling a complex response to DNA damage, and coordinates expression of a subset of its target genes in a time-varying manner, likely driving the DNA damage response through different stages.

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