Comprehensive Modeling and Validation of Glucose and Temperature Compensation of the *Neurospora* Circadian Clock

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***Short Abstract* — Recently, a transcription repressor, CSP-1, was identified as a component of the circadian clock in *Neurospora crassa* functioning in a negative feedback loop on a circadian transcription factor WC-1. This feedback mechanism is suggested to maintain the circadian period in a wide range of glucose concentrations, which is referred to as glucose compensation. Here, we studied a mathematical model of the Neurospora circadian clock incorporating the above WC-1/CSP-1 feedback loop, and investigated molecular mechanisms of glucose and temperature compensation. Our model shows that glucose compensation is achieved by balancing the activation rates of *csp-1* and *wc-1*.**

***Keywords* — Neurospora, Circadian clock, Glucose compensation, Temperature compensation.**

Autonomous circadian oscillations arise from transcriptional-translational feedback loops of core clock components. The period of a circadian oscillator is relatively insensitive to changes in physiological temperature and nutrients (e.g., glucose), which is referred to as temperature and nutrient compensation, respectively. Recently, a transcription repressor, CSP-1, was identified as a component of the circadian system in *Neurospora crassa*. The transcription of *csp-1* is under the circadian regulation [1]. Intriguingly, CSP-1 represses the circadian transcription factor, WC-1, forming a negative feedback loop that can influence the core oscillator [2]. This feedback mechanism is suggested to maintain the circadian period in a wide range of glucose concentrations. In this work, we modified the existing mathematical model of the *Neurospora* circadian clock [3] incorporating the above WC-1/CSP-1 feedback loop, and investigated molecular mechanisms of glucose and temperature compensation.

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Our model shows that glucose compensation exists within a narrow range of parameter space where the activation rates of *csp-1* and *wc-1* are balanced with each other, and that temperature compensation can be achieved by an intricate balance of synthesis and degradation of FRQ and WC-1. More importantly, we experimentally validated loss of glucose compensation in the *wc-1*^{ov} mutant, and maintenance of the abundance of nuclear FRQ as a function of temperature as predicted in the model.

Furthermore, our stochastic simulations demonstrate that the CSP-1-dependent negative feedback loop functions in glucose compensation, but does not enhance the overall robustness of oscillations against molecular noise. Our work highlights predictive modeling of circadian clock machinery and experimental validations employing *Neurospora* and brings a deeper understanding of molecular mechanisms of glucose and temperature compensation.

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