

## Supplementary Information

# Design of Oscillatory Networks through Post-translational Control of Network Components

Brianna E.K. Jayanthi <sup>1</sup>, Shridhar Jayanthi <sup>2</sup> and Laura Segatori <sup>1,2,3,4,\*</sup>

<sup>1</sup> Systems, Synthetic, and Physical Biology Graduate Program, Rice University, Houston, TX 77005, USA

<sup>2</sup> Department of Bioengineering, Rice University, Houston, Texas 77005, USA

<sup>3</sup> Department of Chemical & Biomolecular Engineering, Rice University, Houston, Texas 77005, USA

<sup>4</sup> Department of BioSciences, Rice University, Houston, Texas 77005, USA

\* Corresponding author. E-mail: segatori@rice.edu (L.S.)

**Table S1.** Parameters used in activator-repressor simulations.

Parameter	Description	Value	Source
$p_{AT}$	Activator gene DNA concentration	1 nM	This work
$p_{BT}$	Repressor gene DNA concentration	1 nM	This work
$p_{NT}$	NanoDeg gene DNA concentration	1 nM	This work
$K_A$	Activator-operator equilibrium dissociation constant	3 nM	[1]
$K_B$	Repressor-operator equilibrium dissociation constant	3 nM	[1]
$\delta_A$	Activator degradation rate	4 h	This work
$\delta_B$	Repressor degradation rate	4 h	This work
$\delta_N$	NanoDeg degradation rate	0.9 h	[1]
$\alpha_1$	Activator synthesis rate with maximum self-activation	112.5 h <sup>-1</sup>	[2]
$\alpha_2$	Activator synthesis rate with leaky self-activation	1 h <sup>-1</sup>	Modified from [2]
$\beta_1$	Activator synthesis rate with maximum repression	0.04 h <sup>-1</sup>	Modified from [2]
$\beta_2$	Activator synthesis rate with leaky repression	1.8 h <sup>-1</sup>	[2]
$m$	Activator Hill coefficient	2	This work
$n$	Repressor Hill coefficient	2	This work
$k_5$	Repressor synthesis rate with maximum activation	36 h <sup>-1</sup>	[2]
$k_6$	Repressor synthesis rate with leaky activation	0.05 h <sup>-1</sup>	Modified from [2]
$k_{on}$	NanoDeg-activator association rate constant	0.6264 nM h <sup>-1</sup>	[1]
$k_{off}$	NanoDeg-activator dissociation rate constant	2.7648 h <sup>-1</sup>	[1]
$k_N$	NanoDeg synthesis rate	5 h <sup>-1</sup>	This work

**Table S2.** Parameters used in Goodwin oscillator simulations.

Parameter	Description	Value	Source
$p_{AT}$	Repressor gene DNA concentration	1 nM	This work
$p_{NT}$	NanoDeg gene DNA concentration	1 nM	This work
$K_A$	Repressor-operator equilibrium dissociation constant	3nM	[2]
$\delta_A$	Repressor degradation rate	11 h	This work
$\delta_N$	NanoDeg degradation rate	0.9 h	[1]
$\beta_1$	Repressor synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$\beta_2$	Repressor synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$m$	Repressor Hill coefficient	2	This work
$k_{on}$	NanoDeg-mature repressor association rate constant	0.6264 nM h <sup>-1</sup>	[1]
$k_{off}$	NanoDeg-mature repressor dissociation rate constant	2.7648 h <sup>-1</sup>	[1]
$k_N$	NanoDeg synthesis rate	5 nM/h	This work
$\tau$	Repressor maturation time	0.5 h	This work

**Table S3.** Repressilator with a common NanoDeg.

Parameter	Description	Value	Source
$p_{AT}$	Repressor A gene DNA concentration	1 nM	This work
$p_{BT}$	Repressor B gene DNA concentration	1 nM	This work
$p_{CT}$	Repressor C gene DNA concentration	1 nM	This work
$p_{NT}$	NanoDeg gene DNA concentration	1 nM	This work
$K_A$	Repressor A-operator equilibrium dissociation constant	3 nM	[2]
$K_B$	Repressor B-operator equilibrium dissociation constant	3 nM	[2]
$K_C$	Repressor C-operator equilibrium dissociation constant	3 nM	[2]
$\delta_A$	Repressor A degradation rate	11 h	This work
$\delta_B$	Repressor B degradation rate	11 h	This work
$\delta_C$	Repressor C degradation rate	11 h	This work
$\delta_N$	NanoDeg degradation rate	0.9 h	[1]
$k_1$	Repressor A synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_2$	Repressor A synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$k_3$	Repressor B synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_4$	Repressor B synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$k_5$	Repressor C synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_6$	Repressor C synthesis rate with leaky repression.	181 h <sup>-1</sup>	[2]
$m$	Repressor A Hill coefficient	4	This work
$n$	Repressor B Hill coefficient	4	This work
$r$	Repressor C Hill coefficient	4	This work
$k_{on}$	NanoDeg-repressor association rate constant	0.6264 nM h <sup>-1</sup>	[1]
$k_{off}$	NanoDeg-repressor dissociation rate constant	2.7648 h <sup>-1</sup>	[1]
$k_N$	NanoDeg synthesis rate	6.5 nM/h	This work

**Table S4.** Parameters used in Repressilator with individual NanoDegr simulations.

Parameter	Description	Value	Source
$p_{AT}$	Repressor A gene DNA concentration	1 nM	This work
$p_{BT}$	Repressor B gene DNA concentration	1 nM	This work
$p_{CT}$	Repressor C gene DNA concentration	1 nM	This work
$p_{NTA}$	NanoDeg A gene DNA concentration	1 nM	This work
$p_{NTB}$	NanoDeg B gene DNA concentration	1 nM	This work
$p_{NTC}$	NanoDeg C gene DNA concentration	1 nM	This work
$K_A$	Repressor A-operator equilibrium dissociation constant	3 nM	[2]
$K_B$	Repressor B-operator equilibrium dissociation constant	3nM	[2]
$K_C$	Repressor C-operator equilibrium dissociation constant	3nM	[2]
$\delta_A$	Repressor A degradation rate	11 h	This work
$\delta_B$	Repressor B degradation rate	11 h	This work
$\delta_C$	Repressor C degradation rate	11 h	This work
$\delta_N$	NanoDeg degradation rate	0.9 h	[1]
$k_1$	Repressor A synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_2$	Repressor A synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$k_3$	Repressor B synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_4$	Repressor B synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$k_5$	Repressor C synthesis rate with maximum repression	1.8 h <sup>-1</sup>	[2]
$k_6$	Repressor C synthesis rate with leaky repression	181 h <sup>-1</sup>	[2]
$m$	Repressor A Hill coefficient	4	This work
$n$	Repressor B Hill coefficient	4	This work
$r$	Repressor C Hill coefficient	4	This work
$k_{on}$	NanoDeg-repressor association rate constant	0.6264 nM h <sup>-1</sup>	[1]
$k_{off}$	NanoDeg-repressor dissociation rate constant	2.7648 h <sup>-1</sup>	[1]
$k_{NA}$	NanoDeg A synthesis rate	5 nM/h	This work
$k_{NB}$	NanoDeg B synthesis rate	5 nM/h	This work

$k_{NC}$	NanoDeg C synthesis rate	5 nM/h	This work
----------	--------------------------	--------	-----------

**Table S5.** Parameters used in NanoDeg repressor simulations.

Parameter	Description	Value	Source
$p_{AT}$	Repressor A gene DNA concentration	1 nM	This work
$p_{BT}$	Repressor B gene DNA concentration	1 nM	This work
$p_{NT}$	NanoDeg gene DNA concentration	1 nM	This work
$K_A$	Repressor A-operator equilibrium dissociation constant	3 nM	[2]
$K_B$	Repressor B-operator equilibrium dissociation constant	3 nM	[2]
$\delta_A$	Repressor A degradation rate	11 h	This work
$\delta_B$	Repressor B degradation rate	11 h	This work
$\delta_N$	NanoDeg degradation rate	0.9 h	[1]
$k_1$	Repressor A synthesis rate	18.1 h <sup>-1</sup>	This work
$k_2$	Repressor B synthesis rate with maximum repression	0.018 h <sup>-1</sup>	Modified from [2]
$k_3$	Repressor B synthesis rate with leaky repression	18.1 h <sup>-1</sup>	Modified from [2]
$k_4$	NanoDeg synthesis rate with maximum repression	0.018 h <sup>-1</sup>	Modified from [2]
$k_5$	NanoDeg synthesis rate with leaky repression	18.1 h <sup>-1</sup>	Modified from [2]
$m$	Repressor A Hill coefficient	10	This work
$n$	Repressor B Hill coefficient	10	This work
$k_{on}$	NanoDeg-Repressor A association rate constant	0.6264 nM h <sup>-1</sup>	[1]
$k_{off}$	NanoDeg-Repressor A dissociation rate constant	2.7648 h <sup>-1</sup>	[1]

## References

1. Zhao, W.; Pferdehirt, L.; Segatori, L. Quantitatively Predictable Control of Cellular Protein Levels through Proteasomal Degradation. *ACS Synth. Biol.* **2018**, *7*, 540–552.
2. Zhao, W.; Bonem, M.; McWhite, C.; Silberg, J.J.; Segatori, L. Sensitive detection of proteasomal activation using the Deg-On mammalian synthetic gene circuit. *Nat. Commun.* **2014**, *5*, 3612.