biosensors

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Supplementary Materials

Genetic Programming by Nitric Oxide-Sensing Gene Switch System in Tumor-Targeting Bacteria

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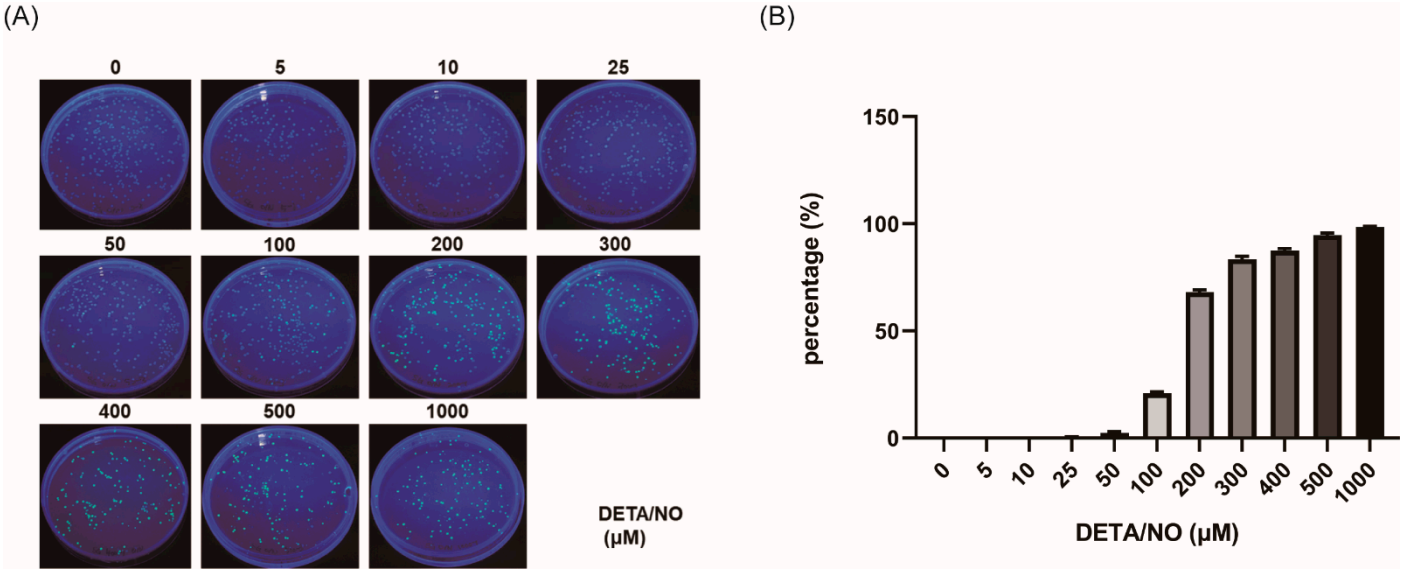

Figure S1. Switch activation percentage under various concentrations of DETA/NO. *SL.* pNASGFP were incubated with the indicated concentrations of DETA/NO for 16 h at 37°C. The bacteria were spread on agar plates containing antibiotics and cultured overnight. (A) Representative UV photos of sfGFP-positive colonies on agar plates. (B) Percentages of activated switching in *SL.* pNASGFP at the indicated concentrations of DETA/NO. Percentages (mean ± SEM) were calculated as the number of sfGFP-positive colonies relative to total colonies. Assays were performed in triplicate.

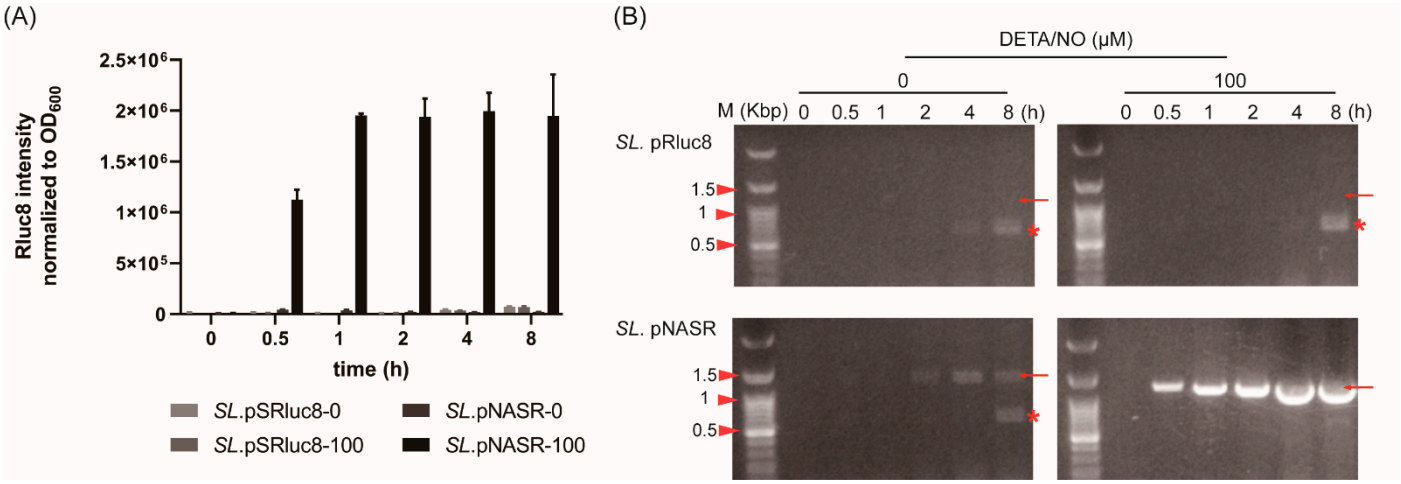

Figure S2. Time-dependent switch event in bacterial cells with the NO−sensing switch system. *SL.* pNASR was cultured for the indicated times in the presence (100 μM) or absence (0 μM) of DETA/NO; *SL.* pSRluc8 was used as the negative control. (A) Rluc8 intensity normalized to number of bacterial cells (OD_{600}). (B) PCR analysis of gene switch events. The switched genes were amplified with ON-state primers (arrows). *, non-specific bands. Results shown as the mean ± SEM of triplicate determinations.

Table S1. Plasmids and *SL*. ΔppGpp transformants.

Plasmid	Constructs	Antibiotic resistance
pFimE	NO inducible FimE expression	Chloramphenicol
pSRluc8	*rluc8* as GOI in *fimS* switch block, with constitutively expressed NorR	Ampicillin
pSGFP	*sfGFP* as GOI in *fimS* switch block, with constitutively expressed NorR	Ampicillin

Strain	Genotype	Purpose
SL. pSRluc8	*SL*. ΔppGpp + pSRluc8	Control
SL. pNASR	*SL*. ΔppGpp + pFimE + pSRluc8	*rluc8* as reporter gene
SL. pNASGFP	*SL*. ΔppGpp + pFimE + pSGFP	*sfGFP* as reporter gene

GOI: gene of interest.

rluc8: *renilla* luciferase variant 8

sfGFP: *super-folder green fluorescent protein*.