

Supplementary Materials

Development and characterization of indole-responsive whole-cell biosensor based on the inducible gene expression system from *Pseudomonas putida* KT2440

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Supplementary Tables

Supplementary Table S1. Bacterial strains used in the study.

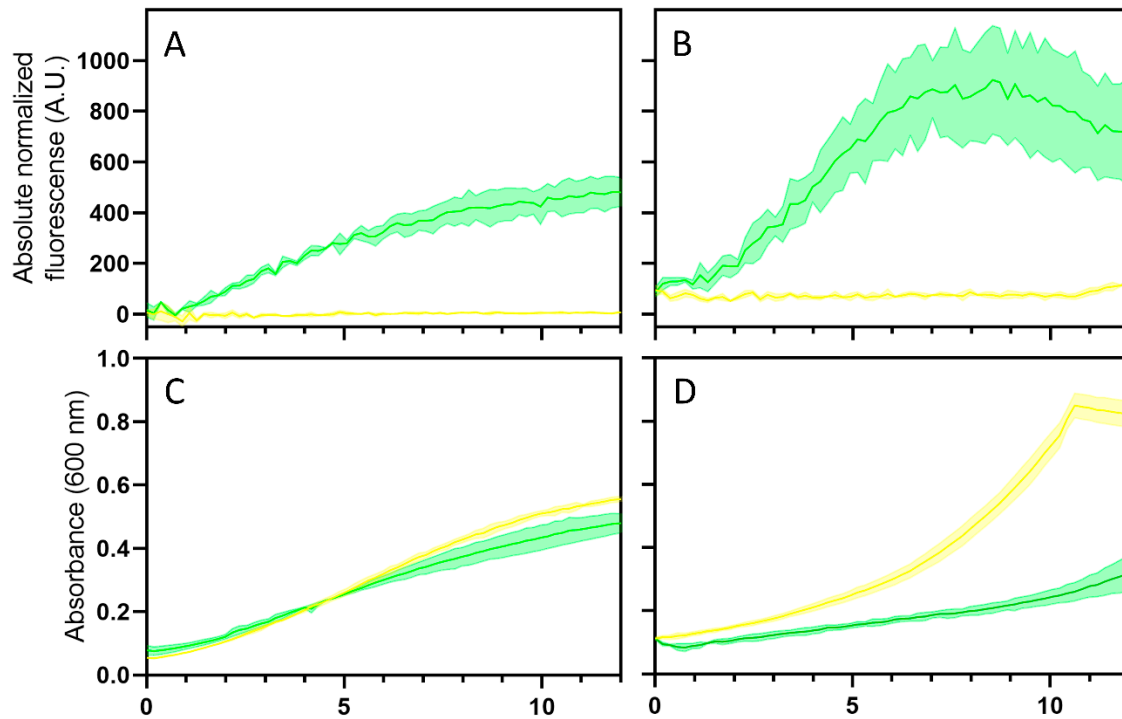
Strain	Characteristics	Reference or source
<i>Escherichia coli</i> TOP10	F ⁻ <i>mcrA</i> Δ (<i>mrr-hsdRMS-mcrBC</i>) Φ 80 <i>lacZ</i> Δ M15 Δ <i>lacX74</i> <i>recA1</i> <i>araD139</i> Δ (<i>araleu</i>)7697 <i>galU</i> <i>galK</i> <i>rpsL</i> (StrR) <i>endA1</i> <i>nupG</i>	Thermo Fisher Scientific, Waltham, MA, USA
<i>Cupriavidus necator</i> H16	Wild-type strain	DSM 428 (DSMZ, Braunschweig, Germany)

Supplementary Table S2. Oligonucleotide primers used in the study.

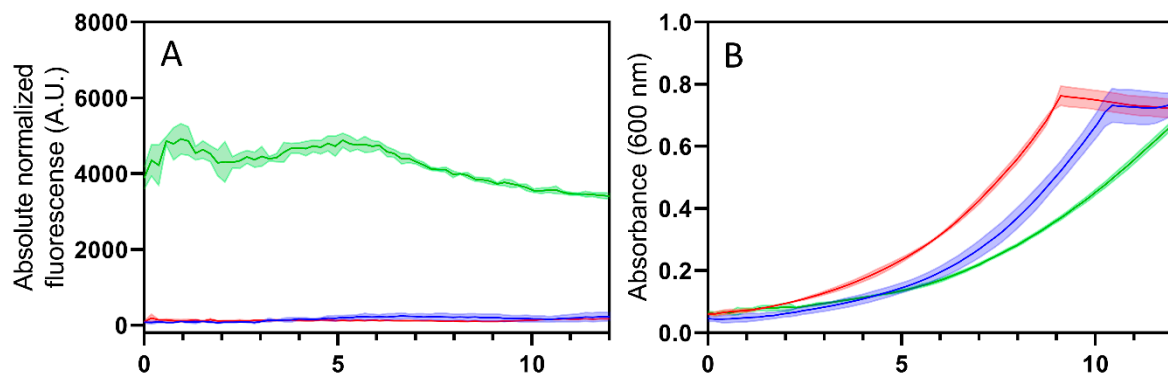
Name	Oligonucleotide primer sequence (5' → 3')
P015	gggccttgcgtttatgacgtctgatatgcaagcgaactaagcgctctgc
P016	cgtcttcgctactcgccatatgggcgctccaaggcgtag
P015TgSr	gggccttgcgtttatgacgtcttacctgtgagttttctgacaagtttac
P065	tcactccaccggtgcttaagcaaaagatcttttaagaaggagatatacatatggaaaactttaacatctccctgaaccg
P066	tccttactcgagtttggatccttaaaactctttaagtttgcggtgaagtgc

Restriction endonuclease recognition sites are underlined.

Supplementary Figures



Supplementary Figure S1. Induction dynamics of *PpTrpI/P_{PP_RS00425}*-based *E. coli* and *C. necator* biosensors. Absolute normalized fluorescence (A, B) and absorbance (C, D) obtained using *E. coli* (A, C) and *C. necator* (B, D) harbouring inducible system *PpTrpI/P_{PP_RS00425}* (pPM0081). Measurements were performed over the period of 12 hours in MM supplemented with 1 mM (green) and without indole (yellow). Data are mean \pm SD, $n = 3$.



Supplementary Figure S2. Screening and characterization of *tnaA*-positive strains using *C. necator*/PpTrpI/P_{PP_RS00425} biosensor. Absolute normalized fluorescence (A) and absorbance (B) of *C. necator* harbouring inducible system PpTrpI/P_{PP_RS00425} without (pPM0081; control, blue) or with *tnaA* gene (pPM0083; red and green). Data are mean \pm SD, n=3.

A

AatII

gacgtctgatatgcaagcgaa**acta**agcgctctgccgcgaagtccgcccctaaccactgcgccaactgctcggcgcgcccgtctcggcgcgcgctggcaccacaatgc
end of *trpI* (PP_RS00425)

cagcgccgcctgggtaggtgaaaagccccaaaggcgcaactcaatcgccccccgaggtcatcggcgaccagcggtcggtgctatcgccacgcccacatcgggccacg
gcagcctccagcaggtatatacaggtgctcgaaggcctggccatagtgaaatgccgcggctccaggccctgttcaccacccaggtcgccaggtctgtggcggtgacgt
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gcgcagggcattgaggggagggaggtcgtgggc**cat**tt**ac**ctgtgagttt**ctgacaagtttacgcaatcttatcggttttcagccgagcttgccgtggttagagtaaagcc**

start of *trpI*

NdeI

catcactcattcactacgccttgagcgcccatatg****

start of *rfp*

B

AatII

gacgtctt**ac**ctgtgagttt**ctgacaagtttacgcaatcttatcggttttcagccgagcttgccgtggttagagtaaagcccatcactcattcactacgccttgagcgcc**

NdeI

catatg

start of *rfp*

C

BglII

agatctttaagaaggagatatacat**atg**gaaaaactttaacatctccctgaaccgttcgcattcgtgtattgagccagtaaaacgtaccactcgcgcttatcgtgaagaggc
start of *tnaA*

aattattaaatccggtatgaaccggttctgctggatagcgaagatgttttatcgatttactgaccgacagcggcaccggggcggtgacgcagagcatgcaggctgcgatga
tgcgcggcgacgaagcctacagcggcagtcgtagctactatgcgttagccgagtcagtgaaaaatatcttgggtatcaatacaccattccgactaccaggggcgtggcgca
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gttgcgcataatccggcagaccagttcccggcacaggcgctggcctgcgagctgtataaagtcgcgggtatccgtgcggtgaaattggctcttctgttaggcgcgcatc
cgaaaaccggtaaacactgccatgccggctgaactgctgcgtttaaccattccgcgcgaacataactcaaacacatatggacttcatttgaagcctttaacatgtgaa

BamHI

agagaacgcggcgcaatattaaaggattaacctttacgtacgaaccgaaagtattgctgctacttcaccgcaaaacttaagaagtt**taaggatcc**

end of *tnaA*

Supplementary Figure S3. Nucleotide sequences of genetic elements of the indole-inducible system. (A) Nucleotide sequence containing *trpI* gene, encoding LysR family transcriptional regulator (locus tag *PP_RS00430*), and intergenic region *PP_RS00430/PP_RS00425*, assembled in pPM0081 and corresponding to the schematic representation in Figure 1B. (B) Sequence of intergenic region *PP_RS00430/PP_RS00425* (in pPM0082 and Figure 1C). (C) Nucleotide sequence of *tnaA* gene encoding tryptophanase (in pPM0083 and Figure 1D). The start and end of open reading frames are displayed in bold. AatII, NdeI, BglII, and BamHI restriction sites are underlined. The nucleotides of intergenic region *PP_RS00430/PP_RS00425* are highlighted in green.