

Supplementary Information

Dual promoters of the major catalase (KatA) govern distinct survival strategies of *Pseudomonas aeruginosa*

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Table S1. Primers used in this study.

<i>Primer^a</i>	<i>Sequence (engineered enzyme site)</i>	<i>Purpose</i>
<u>Gene mutation and S1 mapping</u>		
anr-N1	5'-GCGTCGCGAATTCGAGCCC-3' (<i>EcoRI</i>)	<i>anr</i> deletion
anr-C1	5'-GCCCAGCCCAGAAAGCTTCCA -3' (<i>HindIII</i>)	<i>anr</i> deletion
anr-UC	5'-ACTGCTGGGCGGAGAACAGCGAATCCATGTCT-3'	<i>anr</i> deletion
anr-DN	5'-AGACATGGATTCGCTGTTCTCCGCCAGCAGT-3'	<i>anr</i> deletion
katA-N10	5'-CCGAATAAGGCATCTGCTGC-3'	<i>katAp</i> mutation/ S1 mapping
katA-S1C1	5'-ACCACACGTCCTGCAGCAAC -3' (<i>PstI</i>)	<i>katAp</i> mutation/ S1 mapping
katAp1m-UC	5'-CAGGTTAGGTACCAATGACAGCCCTCCAACAATC-3' (<i>KpnI</i>)'	<i>katAp1m</i> mutation
katAp1m-DN	5'- TGTCATTGGTACCTAACCTGCTTTTACGAAAAGC-3' (<i>KpnI</i>)'	<i>katAp1m</i> mutation
katAp2m-UC	5'-AATCATCGGATCCGACCAGGGATTGGCGGAGGA-3' (<i>BamHI</i>)	<i>katAp2m</i> mutation
katAp2m-DN	5'-CCTGGTCGGATCCGATGATTTCCGTGTAGCC-3' (<i>BamHI</i>)	<i>katAp2m</i> mutation
<u>LacZ transcriptional fusion</u>		
katA-N3	5'-CGTAGATCTGGTTGATCGTC-3' (<i>BglII</i>)	LacZ fusion
pQF50-lacZ-C1	5'-CAGCAGGATATCCTGCACC-3' (<i>EcoRV</i>)	LacZ fusion
katA-lacZ-UC ^b	5'-TTAATACCCTCTAGCTAGAACGTGCTATGAAGCGAA-3'	LacZ fusion
katA-lacZ-DN ^b	5'- TTCGCTTCATAGCACGTT CTAGCTAGAGGGTATTAA-3'	LacZ fusion

a: N and C refer to the forward and reverse primers for each gene, respectively.

b: The *katA* promoter region is indicated (bold).

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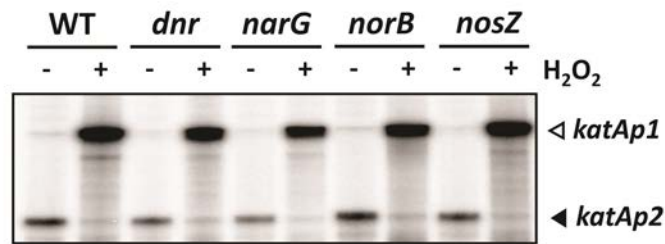


Fig. S1. Transcription profiles of *katA* promoters in various mutants.

The transcription patterns were assessed by low-resolution S1 nuclease assay with H₂O₂ treatments in some mutants for dissimilatory nitrate respiration grown in LB. Total RNA (50 µg) that had been prepared from the wild type (WT) and the mutant (*dnr*, *narG*, *norB*, and *nosZ*) cells with (+) or without (-) 1 mM H₂O₂ treatment for 10 min at OD₆₀₀ of 0.5 were subjected to S1 nuclease assay. The two transcriptional start sites of the *katA* gene are indicated by open (*katAp1*) and closed (*katAp2*) arrows.

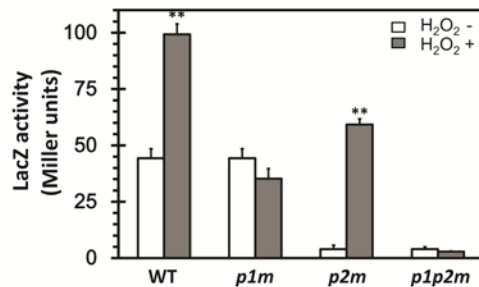


Fig. S2. H₂O₂-induced transcription of the *katA* promoter fusions.

H₂O₂-induced *katA* promoter activities were determined in the wild type cells containing one of the promoter fusions (WT, *p1m*, *p2m* and *p1p2m*). The cells were grown in LB with 15 mM KNO₃ to the mid-logarithmic growth phase, and then treated with (filled bar) or without (empty bar) 1 mM H₂O₂ for 10 min. The aliquots from the cells were subjected to β-galactosidase (LacZ) assay. The error bars represent the standard deviations from five independent experiments (two cultures per experiment). Statistical significance based on the Student's *t*-test is indicated (**, *p* < 0.005).

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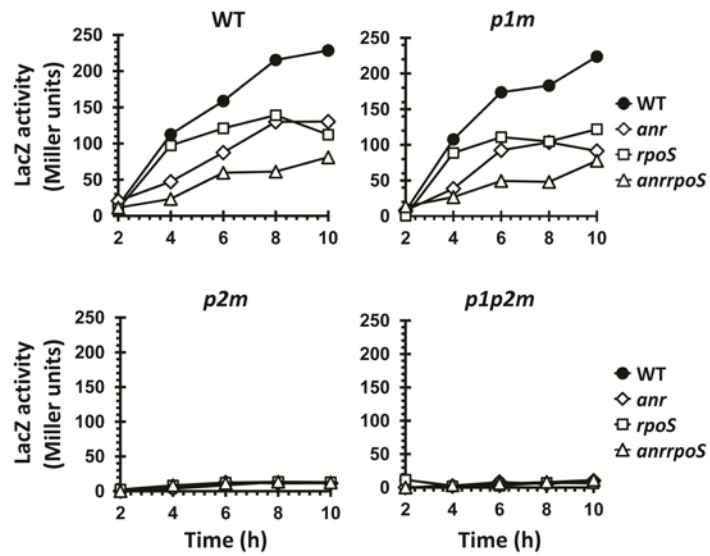


Fig. S3. Stationary phase-induced transcription of the *katA* promoter fusions.

The growth phase-dependent transcriptions of the *katA* promoter fusions were assessed using PA14 (●, WT) as well as in its isogenic *anr* and *rpoS* null mutants (◇, *anr*; □, *rpoS*; △, *anr rpoS*), which harbor one of the *katA* promoter fusions (A, WT; B, *p1m*; C, *p2m*; D, *p1p2m*). The culture aliquots were harvested at every 2 h from 2 to 10 h post-inoculation, and then subjected to LacZ assay. The data represent the average of the means of three independent experiments (two cultures per experiment).