

APPLIED MICROBIOLOGY AND BIOTECHNOLOGY

Supplementary material for:

Enhanced rhamnolipid production in *Burkholderia thailandensis* transposon knockout strains deficient in polyhydroxyalkanoate (PHA) synthesis

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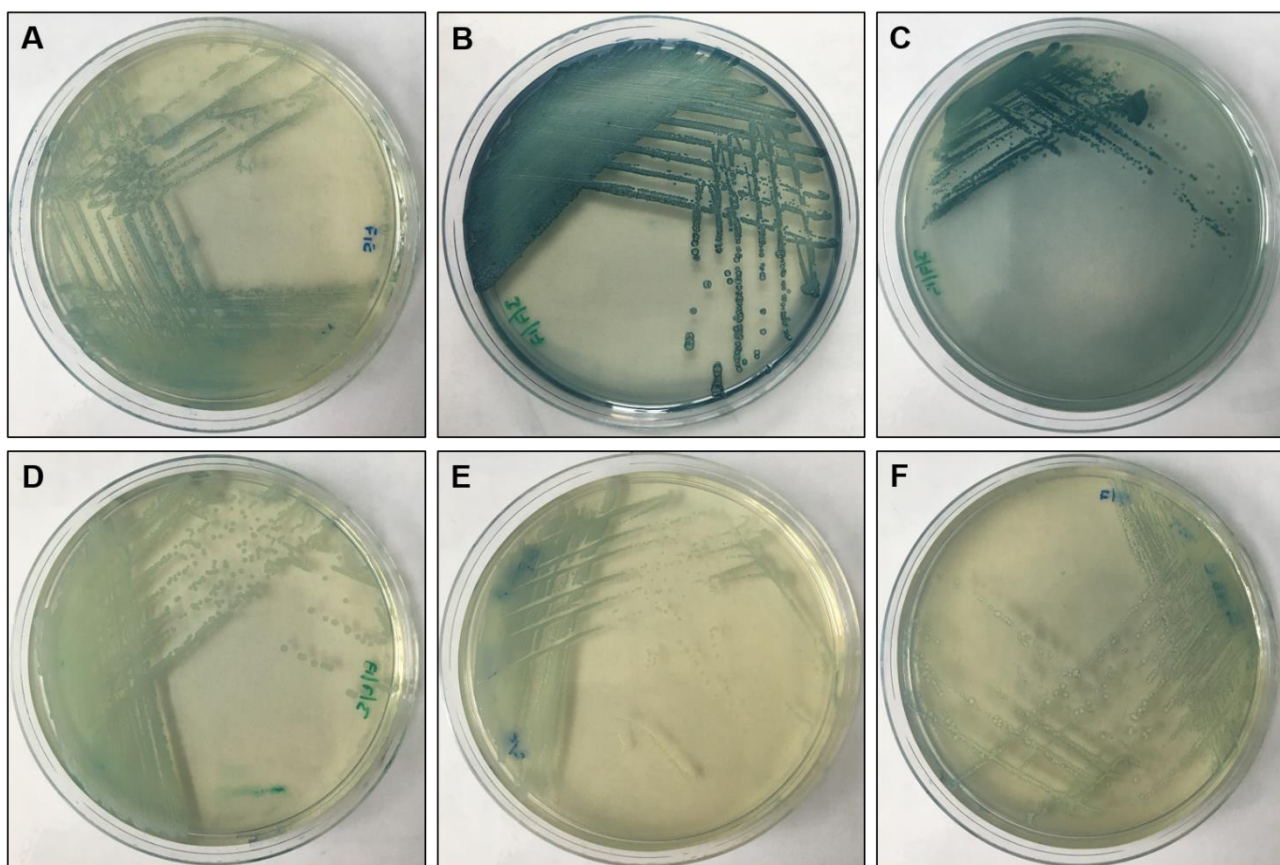


Figure S1

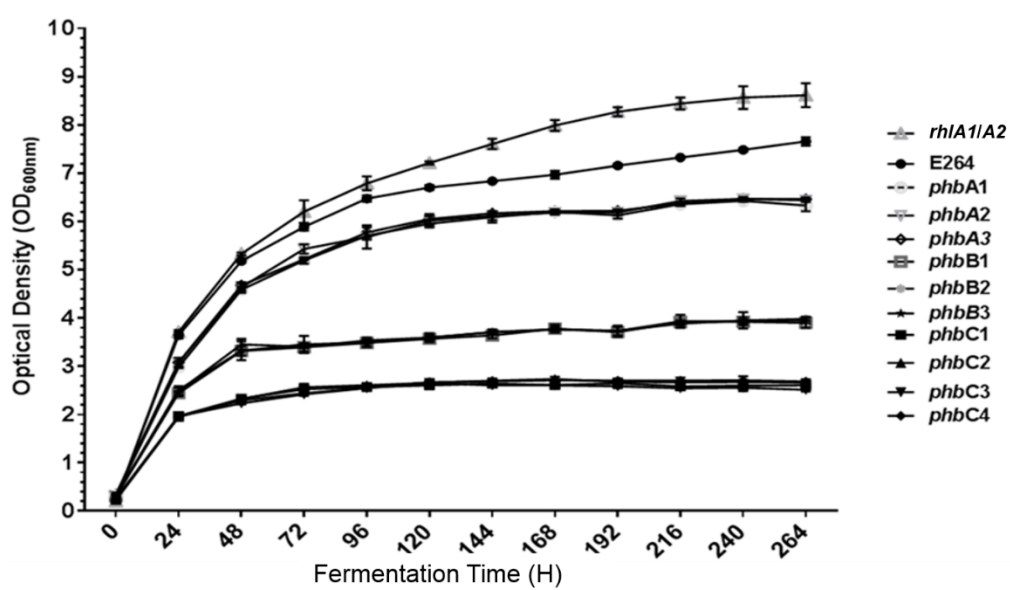


Figure S2

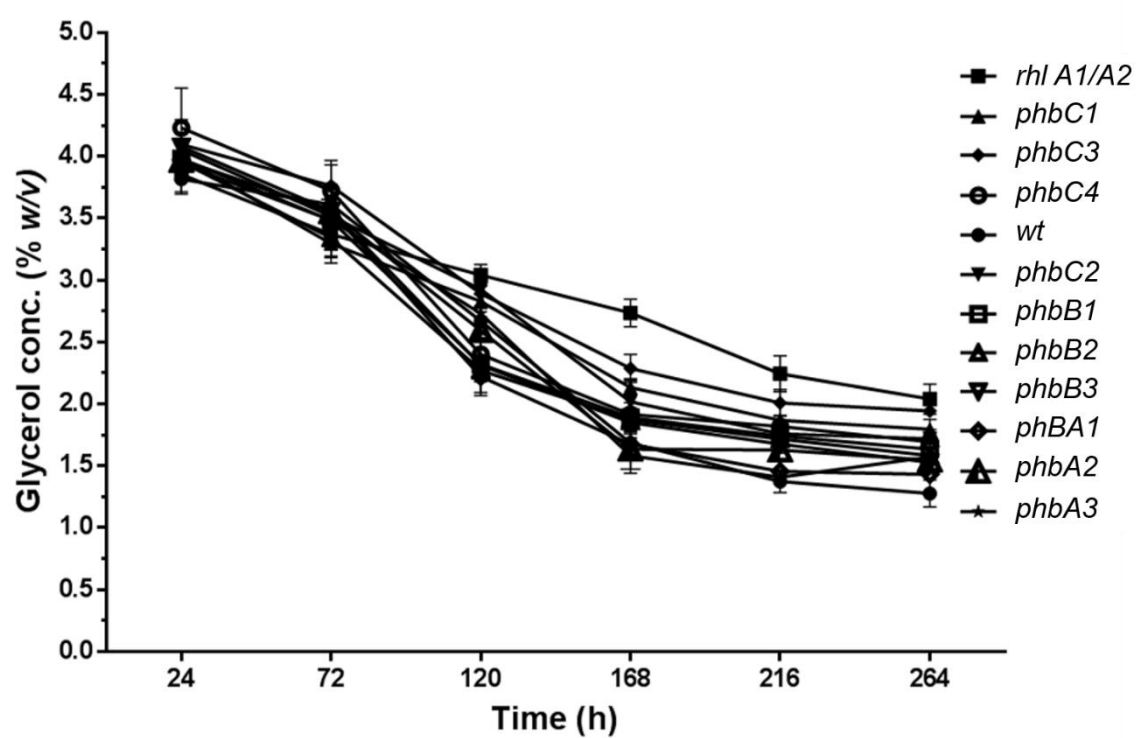


Figure S3

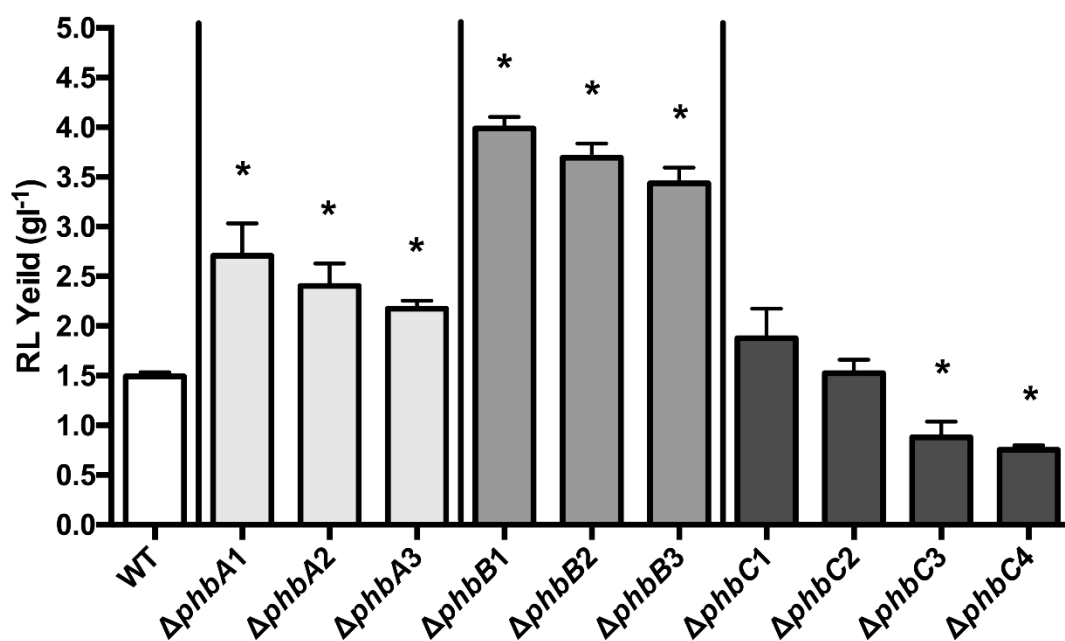


Figure S4

Table S1. List and description of *B. thailandensis* transposon mutants in PHA synthesis genes used in this study.

Species	Strain Name	Strain Description	Transposon Mutant Library Designation (Gallagher <i>et al.</i> 2013)
<i>B. thailandensis</i>	E264 <i>phbC1</i>	T23 transposon mutant in BTH_I2255 (<i>phbC</i>) encoding a poly(R)-hydroxyalkanoic acid synthase, class I	tnbt1_er100504p04q148
<i>B. thailandensis</i>	E264 <i>phbC2</i>	T23 transposon mutant in BTH_I2255 (<i>phbC</i>) encoding a poly(R)-hydroxyalkanoic acid synthase, class I	tnbt1_er100421p06q104
<i>B. thailandensis</i>	E264 <i>phbC3</i>	T23 transposon mutant in BTH_I2255 (<i>phbC</i>) encoding a poly(R)-hydroxyalkanoic acid synthase, class I	tnbt1_er100421p06q152
<i>B. thailandensis</i>	E264 <i>phbC4</i>	T23 transposon mutant in BTH_I2255 (<i>phbC</i>) encoding a poly(R)-hydroxyalkanoic acid synthase, class I	tnbt1_er100408p04q145
<i>B. thailandensis</i>	E264 <i>phbA1</i>	T23 transposon mutant in BTH_I2256 (<i>phbA</i>) encoding a acetyl-CoA acetyltransferase	tnbt1_er100412p05q191
<i>B. thailandensis</i>	E264 <i>phbA2</i>	T8 transposon mutant in BTH_I2256 (<i>phbA</i>) encoding a acetyl-CoA acetyltransferase	tnbt1_2r100310p04q118
<i>B. thailandensis</i>	E264 <i>phbA3</i>	T23 transposon mutant in BTH_I2256 (<i>phbA</i>) encoding a acetyl-CoA acetyltransferase	tnbt1_er100329p06q190
<i>B. thailandensis</i>	E264 <i>phbB1</i>	T23 transposon mutant in BTH_I2256 (<i>phbB</i>) encoding a acetylacetyl-CoA reductase	tnbt1_er100329p01q164
<i>B. thailandensis</i>	E264 <i>phbB2</i>	T23 transposon mutant in BTH_I2256 (<i>phbB</i>) encoding a acetylacetyl-CoA reductase	tnbt1_er100406p01q116
<i>B. thailandensis</i>	E264 <i>phbB3</i>	T23 transposon mutant in BTH_I2256 (<i>phbB</i>) encoding a acetylacetyl-CoA reductase	tnbt1_er100419p05q185

Supplementary Figure Legends

Figure S1. PHA synthesised within the cell was stained using Sudan Black. A. *E. coli* JM109 (PHA -). B. *P. aeruginosa* PAO1 (PHA +). C. *B. thailandensis* E264 WT. D. *B. thailandensis* E264 $\Delta phbA1$. E. *B. thailandensis* E264 $\Delta phbB1$. F. *B. thailandensis* E264 $\Delta phbC1$

Figure S2. Growth analysis of *B. thailandensis phb* transposon mutants, *B. thailandensis* E264 WT and *B. thailandensis rhIA1/A2* throughout a 264 h fermentation period. Mean values with error bars representing standard deviation from the mean (n=3).

Figure S3. Glycerol concentrations of *B. thailandensis phb* transposon mutants, *B. thailandensis* E264 WT and *B. thailandensis rhIA1/A2* throughout a 264 h fermentation period. Values were obtained using GC-MS. Mean values with error bars representing standard deviation from the mean (n=3).

Figure S4. RL production in *B. thailandensis* transposon mutants after 264 h fermentation in shake flasks. Mean weights of crude RL extracts from three replicate cultures with error bars representing SD in weight from the mean. Data analysed using a one-way ANOVA with *post hoc*. Tukey's multiple comparisons tests, (* $p < 0.05$).