Electronic Supplementary Material (ESI) for Reaction Chemistry & Engineering.

Supporting Information

Systematic metabolic engineering of *Klebsiella oxytoca* for production of 1,3-propanediol from glucose

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Table S1. Bacterial strains and plasmids used in this study.

Strain or plasmid	Description	Source
Strain		
Escherichia coli DH5α	*F⁻, φ80d lacZΔM15, Δ(lacZYA-argF)U169, recA1, endA1, hsdR17(rk⁻,mk⁻), phoA, supE44λ⁻, thi⁻¹, gyrA96, relA1*	Invitroge
E. coli S17-1 *λpir*	*TpR SmR recA*, *thi*, *pro*, *hsdR-M + RP4*: 2-*Tc:Mu: Km Tn7 λpir*.	[1]
PDL-0	*Klebsiella oxytoca*，wide type	[2]
PDL-*gpd*	PDL-0, harboring pBAD322cT-P_{tac}-*gpd*-P_{tac}-*gpp2*	This study
PDL-*gpsA*	PDL-0, harboring pBAD322cT-P_{tac}-*gpsA*-P_{tac}-*gpp2*	This study
PDL-01	PDL-0, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-02	PDL-01△*ldhA*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-03	PDL-02△*adhE*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-04	PDL-03△*pta*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-05	PDL-04△*poxB*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-06	PDL-05△*budA*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL-07	PDL-06△*frdA*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL1-1	PDL-07△*dhaD1*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL1-2	PDL1-1△*glpK*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL1-3	PDL1-2△*dhaD2*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study

PDL1-4	PDL1-3△*gldA*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL1-5	PDL1-4△*dhaD3*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL1-6	PDL1-4△*glpF*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL2-1	PDL1-5△*ptsG*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL2-2	PDL1-6△*ptsG*, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	This study
PDL2-3	PDL2-2, harboring pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*-P_{tac}-*glF*	This study
Plasmid		
$pKR6K_{Cm}$	Cm^r, gene replacement vector derived from plasmid pK18*mobsacB*, R6K origin, Mob^+ *sacB*, derived from PKR6K	[2]
pBAD322c	Cm^r, expressing vector, araBAD	[3]
pBAD322cT	Cm^r, promoter replacement vector derived from plasmid pBAD322c, P_{tac}	This study
pBAD322cT-P_{tac}-*gpd1*	pBAD322cT containing *gpd1* gene from *Saccharomyces cerevisiae* under the control of P_{tac} promoter	This study
pBAD322cT-P_{tac}-*gpd*	pBAD322cT containing *gpd* gene from *Pichia kudriavzevii* under the control of P_{tac} promoter	This study
pBAD322cT-Ptac-*gpsA*	pBAD322cT containing *gpd* gene from *Archaeoglobus fulgidus* under the control of P_{tac} promoter	This study
pBAD322cT-P_{tac}-*gpp2*	pBAD322cT containing *gpp2* gene from *Sugiyamaella lignohabitans* under the control of P_{tac} promoter	This study
pBAD322cT-P_{tac}-	pBAD322cT-P_{tac}-*gpsA* with the insertion of *gpp2* gene	This study

gpsA-P_{tac}-*gpp2*	under the control of P_{tac} promoter	
pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*	pBAD322cT-P_{tac}-*gpd1* with the insertion of *gpp2* gene under the control of P_{tac} promoter	This study
pBAD322cT-P_{tac}-*gpd*-P_{tac}-*gpp2*	pBAD322cT-P_{tac}-*gpd* with the insertion of *gpp2* gene under the control of P_{tac} promoter	This study
pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2*-P_{tac}-*glF*	pBAD322cT-P_{tac}-*gpd1*-P_{tac}-*gpp2* with the insertion of *glF* gene from *Zymomonas mobilis* under the control of P_{tac} promoter	This study

Cm^r: resistance to chloramphenicol

Table S2 Sequences of primers used in this study.

Primer[a]	Sequence (5′-3′)[b]
gpsA-F(*Eco*RI)	AATTGAATTCATGAACGCGCTTAACGCTG
gpsA-R(*Pst*I)	TTAACTGCAGTTAGTTGCTACGCTCGTCCTTG
gpd1-F(*Eco*RI)	AATTGAATTCATGAGTGCCGCCGCCGATCG
gpd1-R(*Pst*I)	TTAACTGCAGTCAATCTTCATGCAGATCCAGTTC
gpd-F(*Eco*RI)	AATTGAATTCATGGTTAGCCCGGCCGAA
gpd-R(*Pst*I)	TTAACTGCAGTTAATCTTCCACCGGTTCCAGGCA
gpp2-F(*Eco*RI)	AATTGAATTCATGAGCTCTGCCAGTACTCCG
gpp2-R(*Pst*I)	TTAACTGCAGTTAGGCCACGATGTTGCTAAAGGT
Δ*frdA*-FU(*Eco*RI)	AATTGAATTCCAGTCCCTGAGAATGACCAG
Δ*frdA*-RU	TCTTGTTGACCAGAACGACATTCCTCCAGTTTTTGTTTA
Δ*frdA*-FD	ACTGGAGGAATGTCGTTCTGGTCAACAAGAACGGCTAC
Δ*frdA*-RD(*Bam*HI)	ATGCGGATCCGCCCGCTTCGGTTGCGC
Δ*glpK*-FU(*Eco*RI)	AATTGAATTCCGATGATGAAGATCCTGTGATCG
Δ*glpK*-RU	ATTCCCTGGTGGGCGTAGGTAGTCTCCTGAGTCACAC
Δ*glpK*-FD	TCAGGAGACTACCTACGCCCACCAGGGAATCGAACGC
Δ*glpK*-RD(*Bam*HI)	ATGCGGATCCCGCGGTTAATTAAAACCACCGGAA
Δ*dhaD1*-FU(*Eco*RI)	AATTGAATTCACCGCCTGACGATATGCGAT
Δ*dhaD1*-RU	CACCAAAGGAAAGCTATGATGAGCCCCGGTGTTTCTC
Δ*dhaD1*-FD	GTGAGGGTGTAGAGAAGCTTTCCTTTGGTGATAACCC
Δ*dhaD1*-RD(*Bam*HI)	ATGCGGATCCGGAAATGGAACCGAACAGACGCC
Δ*dhaD2*-FU(*Eco*RI)	AATTGAATTCCCACCTGGTCAGTGAGCCCG
Δ*dhaD2*-RU	GCACCGGTAACGGCAATCTCTACACCCTCACATTGAA
Δ*dhaD2*-FD	GTGAGGGTGTAGAGATTGCCGTTACCGGTGCCGG
Δ*dhaD2*-RD(*Bam*HI)	ATGCGGATCCGAGCCAAATAAACGCCCA
Δ*dhaD3*-FU(*Eco*RI)	AATTGAATTCATCCCCAGCTGTTTGGCAT
Δ*dhaD3*-RU	GAATAAAACGGGGGACAATGCTGTCCTTGTGCTGTTTC
Δ*dhaD3*-FD	ACAAGGACAGCATTGTCCCCCGTTTTATTCGCCTC

Δ*dhaD3*-RD(*Bam*HI)	ATGCGGATCCGACACGATAAACAGGTCGCTCAG
Δ*glpF*-FU(*Eco*RI)	AATTGAATTCGCGCGCTCTGTACTTCCTG
Δ*glpF*-RU	ACCGGATGATGGGAAAATCGAATCCTGAAGCGTAGTG
Δ*glpF*-FD	CTTCAGGATTCGATTTTCCCATCATCCGGTAGGTC
Δ*glpF*-RD(*Bam*HI)	ATGCGGATCCGACGTTCCTGATTGTGGTGGT
Δ*ptsG*-FU(*Eco*RI)	AATTGAATTCGCACAACAGACGACTGAAAATTT
Δ*ptsG*-RU	TGGCGTCTTCACGACTTCACCATGATGCCGTAAGC
Δ*ptsG*-FD	CGGCATCATGGTGAAGTCGTGAAGACGCCACCG
Δ*ptsG*-RD(*Bam*HI)	ATGCGGATCCCGGTGGCGTCTTCACGAC
glF -F(P_{tac})	CCAAGAGATATACAT ATGAGCAGCGAAAGCAGCCA
glF -R(*Hin*dIII)	TTTCGCCAAACAGCCAAGCTTTTATTTCTGCGAGCGCCACAT
P_{tac}-F(HindIII)	AACATCGTGGCCTAAAAGCTTCTGTGGTATGGCTGTGCAGGTC
P_{tac}-R(glF)	GCTTTCGCTGCTCATATGTATATCTCTTGGTGAAATTGTT

[a]-F indicates that this is a sense primer; -R indicates that this is an antisense primer.

[b]Restriction sites are underlined.

References

1. Y. Wang, F. Tao and P. Xu, *J. Biol. Chem.*, 2014, **289**, 6080–6090.
2. B. Xin, F. Tao, Y. Wang, H. Liu, C. Ma and P. Xu, *Metab. Eng.*, 2017, **41**, 102–114.
3. J. E. Cronan, *Plasmid*, 2006, **55**, 152–157.