

Supplementary materials

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gldA101      1 ATGACGAAAATCATTACCTCTCCAAGCAAGTTTATACAAGGCCCGATGAATTGTCCAGG
gldA-syn     1 .....C.....C.....C..CTC...A..C..C..G.....C..G.....C..

gldA101     61 CTTTCGGCGTATACGGAAAGCCTTGGCAAAAAGCATTATTATTGCGGATGATTTTGTC
gldA-syn     61 T.GAGT..C.....C...C..T.G.....G..G..C.....C..C..T..C..C..C..G

gldA101     121 ACCGGCCTTGTCGGCAAAACGGTTGAAGAAAGCTATGCCGGCAAAGAAACGGGGTATCAA
gldA-syn     121 .....T.G..G..T..G..T..G..G..GTC.....C..T..C..G

gldA101     181 ATGGCATTATTTCGGTGGTGTGAGTGTCTAAACCGGAAATCGAACGGCTTTGTGAAATGAGC
gldA-syn     181 .....C..G..T..C..C..A..CAG...G..T..G.....G..CT.G..C..G...TC.

gldA101     241 AAATCCGAGGAAGCCGATGTCGTTGTGCGAATCGGCGGCGGAAAAACATTGGATACCGCA
gldA-syn     241 .....A.....C..T..G..G..C.....T..C.....C..A..C..T..C

gldA101     301 AAAGCAGTCGGGTATTACAATAACATTCCGGTGATTGTGCGCCGACCATCGCTTCCACC
gldA-syn     301 ..G..C..G..C..C.....C.....C..G..C..C..T..T.....

gldA101     361 AATGCCCGACAAGCGCCCTGTCTGTTATTTACAAAGAGAACGGCGAGTTTGAAGAATAC
gldA-syn     361 .....C..CTC...T..AG...G..C..T..G.....A..C..G.....

gldA101     421 TTGATGCTGCCGCTGAACCCGACTTTTGTGATTATGGATACGAAAGTGATTGCCTCTGCC
gldA-syn     421 .....T...TT.....T..C.....G..C.....C..T..G.....C.....C..T

gldA101     481 CCTGCCCGCCTGCTCGTTTCCGGCATGGGAGATGCGCTTGCAACGTATTTTGAAGCGCGC
gldA-syn     481 .....T.AT.G.....C..C..CT.G..C..T..C.....C...

gldA101     541 GCCACTAAGCGGGCAAATAAAACGACGATGGCAGGCGGGCGTGTACGGAAGCGGCGATC
gldA-syn     541 .....C.....G..C..C.....T..T..C..C.....T..G..C..C...

gldA101     601 GCGCTTGCAAAACTTTGTTATGACACGCAAATTTTCGGAAGGTTTAAAAGCAAACTGGCA
gldA-syn     601 ..CT.G..C..GT.G..C..C.....C..G..CAGT..G..C..G..G..C..GT.A..C

gldA101     661 GCGGAAAAACATCTTGTACGGAAGCAGTGGAAAAAATCATTGAAGCGAATACGTATCTG
gldA-syn     661 ..C..G.....CT.G..G..C..G..C..T..G..G.....C..G..C..C..T..CT..

gldA101     721 AGCGGAATCGGTTCTGAAAGCGGCGCCTTGCTGCGGCACATGCGATCCATAATGGGCTT
gldA-syn     721 TC...T..T..C..C.....T.....T.A..C..C..T..C..C..T..C..C..CT.G

gldA101     781 ACCGTGCTCGAAGAAACCCATCATATGTACCACGGCGAAAAAGTGGCATTTCGGTACCCTC
gldA-syn     781 .....T.G..G..G.....C..C.....G..G.....T.....C..TT.G

gldA101     841 GCCCAGCTGATTTTGGAAAGATGCGCCGAAAGCGGAAATTGAAGAGGTGGTCTCCTTCTGC
gldA-syn     841 .....T...C.....G.....C..C..G..C..G.....G.....G.....T

gldA101     901 CTGAGTGTGCGGACTTCCCGTCACGCTCGGGGATTTGGGCGTGAAAGAAGTGAATGAGGAA
gldA-syn     901 T..TCC..G..CT.G..T..T..CT.G..C..C.....G...T...C.....

gldA101     961 AAGCTCCGAAAAGTGGCTGAACTTTTCTGTGCGGAAGGCGAAACGATTTATAACATGCCG
gldA-syn     961 ..AT.G..G..G.....C...T.G.....C..C..G.....C..C..C.....C

gldA101     1021 TTTGAAGTCACGCCTGACCTTGTGTACGCAGCAATCGTTACCGCTGATTCCGTCGGGCGG
gldA-syn     1021 .....G..G..C..C..TT.G.....C..C.....C.....G..C..C

gldA101     1081 TATTATAAGGAAAATGGGCATGA
gldA-syn     1081 .....C.....G.....C.A.

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Figure S1 Nucleotide sequence alignment of *gldA101* and the codon-optimized *gldA101* (i.e., *gldA101-syn*, synthesized by Genewiz Inc). Conserved nucleotide sequences in *gldA101-syn* are indicated as dotted lines.

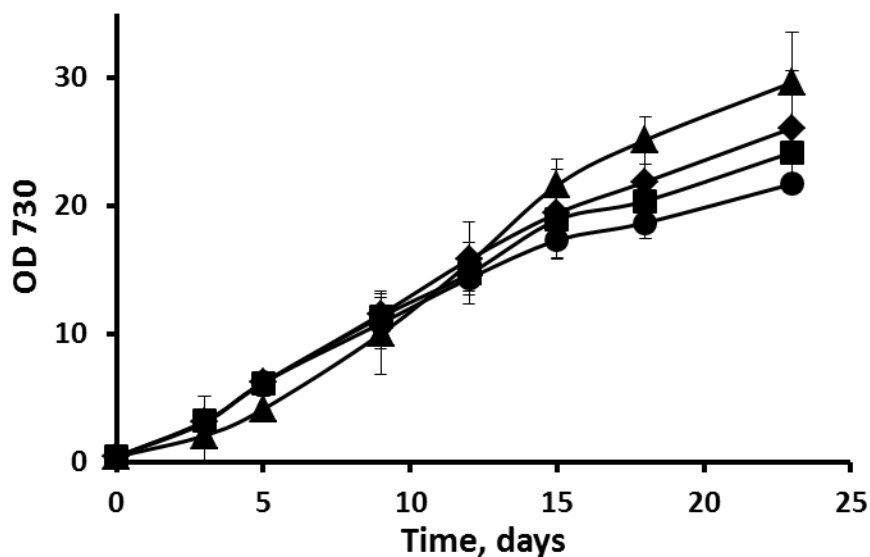


Figure S2 Autotrophic growth curve for *Synechocystis* 6803 strains shows similar growth of the engineered D-lactate producing strains as compared to the wild type strain. Diamond: Wild type. Square: AV08. Triangle: AV10. Circle: AV11.

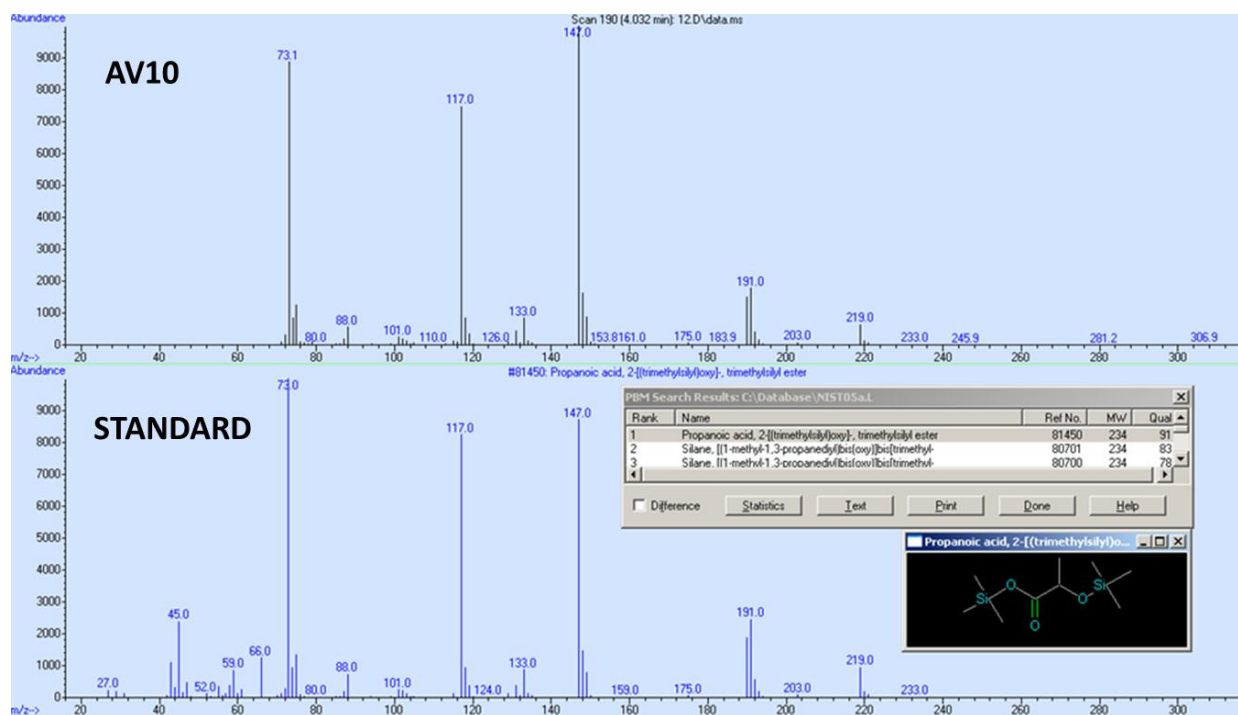


Figure S3 Mass spectra obtained via GC-MS confirm the presence of lactate in the cell culture supernatant of AV10 strain. D/L lactate enzyme kit (R-Biopharm) was used to further confirm that the product is an optically pure D-lactate.