

## SUPPLEMENTAL MATERIAL

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### A Genetic System for *Clostridium ljungdahlii*: A Chassis for Autotrophic Production of Biocommodities and a Model

#### Homoacetogen

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**Table S1: List of primers/oligos used in this study.**

Primers/Oligos	Sequences (5'-3')	Descriptions
pCL1polylinker F	AATTCCCGGGGATCCGTCGACCTGCA	Incorporate <i>Eco</i> R I- <i>Sma</i> I- <i>Bam</i> H I- <i>Sal</i> I- <i>Pst</i> I sites to pCL1
pCL1polylinder R	GGTCGACGGATCCCCGGG	Incorporate <i>Eco</i> R I- <i>Sma</i> I- <i>Bam</i> H I- <i>Sal</i> I- <i>Pst</i> I sites to pCL1
catP F	TCGAATTCCCCTTAAGCGCTC	Amplify the chloramphenicol resistance cassette ( <i>Eco</i> R I site underlined)
catP R	TCTAAGCTTAACACAAGGTCTTGTAC	Amplify the chloramphenicol resistance cassette ( <i>Hind</i> III site underlined)
fliA-1	TCTCTAGAATTGAAATAGACGTGGAC	Amplify <i>fliA</i> upstream region of homology ( <i>Xba</i> I site underlined)
fliA-2	TCGAATTCTCTTAAGTTCATCTATC	Amplify <i>fliA</i> upstream region of homology ( <i>Eco</i> R I site underlined)
fliA-3	TCTAAGCTTGTCAACTTCACAGCAGAGC	Amplify <i>fliA</i> downstream region of homology ( <i>Hind</i> III site underlined)
fliA-4	TCTCTCGAGGTCACTCTGCTTAAATCC	Amplify <i>fliA</i> downstream region of homology ( <i>Xho</i> I site underlined)
ermC F	TCGAATTCTAGATAAATCTCTCATATC	Amplify the <i>ermC</i> resistance cassette from pCL1 ( <i>Eco</i> R I site italicized)
ermC R	TCTAAGCTTGCCTCATATTATATAG	Amplify the <i>ermC</i> resistance cassette from pCL1 ( <i>Hind</i> III site italicized)
adhE1-1	TCTCTAGATGAAAGTTACAAACGTAGAAG	Amplify <i>adhE1</i> upstream region of homology ( <i>Xba</i> I site underlined)
adhE1-2	TCGAGCTCTTATTAAAGTTCTACCGATTCTACCTC	Amplify <i>adhE1</i> upstream region of homology ( <i>Sac</i> I site underlined; stop codons italicized)
adhE1-3	CAAAGCTTATACAGAACGGTAC	Amplify <i>adhE1</i> downstream region of homology (original <i>Hind</i> III site underlined)
adhE1-4	TTCTCGAGCTGCAATTCTAACCATACC	Amplify <i>adhE1</i> downstream region of homology ( <i>Xho</i> I site underlined)
adhE2-1	TCTCTAGACTATAGTTGATGCAGAACTTATG	Amplify <i>adhE2</i> upstream region of homology ( <i>Xba</i> I site underlined)
adhE2-2	TCGAATTCACTTGTCTTGAGTAGCTAG	Amplify <i>adhE2</i> upstream region of homology ( <i>Eco</i> R I site underlined)
adhE2-3	TCGGTCGACGAAGATATGCTAGAATAGCTGA	Amplify <i>adhE2</i> downstream region of homology ( <i>Sal</i> I site underlined)
adhE2-4	TTCTCGAGTAACATCTCATGTGCCTTATAA	Amplify <i>adhE2</i> downstream region of homology ( <i>Xho</i> I site underlined)
adhE1+E2-1	TACGTTCTAGAATTAGAAAG	Amplify <i>adhE1</i> upstream region of homology (original <i>Xba</i> I site underlined)
adhE1+E2-2	TGGAATTCCACTTGTCTTGAGTATATG	Amplify <i>adhE1</i> upstream region of homology ( <i>Eco</i> R I site underlined)
ermC-adhE-F	TATGAGCTAAAGAGGGTTATAATGAACGAG	Amplify <i>ermC</i> coding region from pCL1 ( <i>Sac</i> I site italicized)
ermC-adhE-R	TCTAAGCTTGCCTCATATTATATAG	Amplify <i>ermC</i> coding region from pCL1 ( <i>Hind</i> III site italicized)
adhE1-F	ACCATATGAAAGTTACAAACGTAGAAG	Amplify <i>adhE1</i> coding region ( <i>Nde</i> I site italicized)
adhE1-R	ATGGATCCAATTACTTTCTTCATCTTCTAC	Amplify <i>adhE1</i> coding region ( <i>Bam</i> H I site italicized)