Supplementary information

A multi-enzyme cascade coupled with electrochemistry for efficient synthesis of L-lactate from carbon dioxide

Sanquan Liang, [a, b, 1] Zhiyao Peng, [a, 1] Xue Xia, [a] Kaixing Xiao, [a] Zhao Qin, [a] Xuemei Liu, [a] Fanzhen Lin, [a] Mengdie Wang, [a] Saimeng Jin, [*a] James H. Clark [c] and Dan Wang [*a]

[a] Department of Chemical Engineering, School of Chemistry and Chemical Engineering, Chongqing University, Chongqing 401331, China

[b] College of Food Science and Nutritional Engineering, China Agricultural University, Beijing 100083, China

[c] Circa Renewable Chemistry Institute, Green Chemistry Centre of Excellence, University of York, York YO10 5DD, UK

[*] Corresponding author, Saimeng Jin, E-mail: sj708@cqu.edu.cn; Dan Wang, dwang@cqu.edu.cn

[1] Contributed equally to this work.

Table S1. Strains and plasmids used in this study.

Strains	Relevant characteristics	Source
E. coli DH5α	Host for cloning plasmids	Lab stock
E. coli BL21(DE3)	Host for expression plasmids	Lab stock
ECLA	*E. coli DH5α* carrying plasmid pET28aA	This study
ECLP	*E. coli* DH5α carrying plasmid pET28aP	This study
ECLP-177	*E. coli* DH5α carrying plasmid pET28aP$_2$	This study
ECLL	*E. coli* DH5α carrying plasmid pET28aL	This study
Plasmids		
pET28a(+)	Expression vector, Kan^R	Lab stock
pET28aA	pET28a(+) carries a alcohol dehydrogenase (ADH) gene from *Kluyveromyces lactis*, Kan^R	This study
pET28aP	pET28a(+) carries a pyruvate decarboxylase (PDC) gene from *Zymomonas mobilis*, Kan^R	This study
pET28aP$_2$	pET28a(+) carries a mutated PDC gene (A25T/H414D/E473Q/I476Y) from *Zymomonas mobilis*, Kan^R	This study
pET28aL	pET28a(+) carries a L-lactate dehydrogenase (LDH) gene from *Theileria annulata*, Kan^R	This study

Table S2. List of primers used in this study.

Primers	Sequence of primer	Source
pET28aA-F	5'- AATGGGTCGCGGATCCATGTTCAGATTAGCCCG-3'	This study
pET28aA-R	5'- TGTCGACGGAGCTCGAATTTTATTTGTAAGTGTCAACGACG-3'	This study
pET28aP-F	5'-<u>GCAAATGGGTCGCGGATCC</u>ATGTCATATACTGTAGGAACATACC-3'	This study
pET28aP-R	5'-<u>GCTTGTCGACGGAGCTCGAATTC</u>TTACAGTAATTTGTTAACCGG-3'	This study
pET28aL-F	5'- AAATGGGTCGCGGATCCATGGCCCGTAACAACAA-3'	This study
pET28aL-R	5'- TTGTCGACGGAGCTCGAATTCTTATTTGATCAGCGCTTCC-3'	This study