

Supplementary Information

Biosynthesis of 5-hydroxymethyl-2-furancarboxylic acid from 5-hydroxymethylfurfural using new whole-cell biocatalysts

Shi-Kai Jiang^a, Heng Lei^a, Ji-Hang Shen^a, Xi Shen^a, Xiao-Jun Ji^b, Zhi-Gang Zhang^{a*}

^a School of Pharmaceutical Sciences, Nanjing Tech University, Nanjing 211800, P. R. China;

^b College of Biotechnology and Pharmaceutical Engineering, Nanjing Tech University,
Nanjing 211800, P. R. China

*Corresponding author

Dr. Zhi-Gang Zhang, Tel/Fax: +86 25 5813 9942; E-mail: zhangzg@njtech.edu.cn

1. **Figure S1.** The selective oxidation of 5-hydroxymethylfurfural (HMF) to 5-hydroxymethylfuroic acid (HMFA) by *Pseudochrobactrum* sp. B2L (**S-2**)
2. **Figure S2.** The selective oxidation of 5-hydroxymethylfurfural (HMF) to 5-hydroxymethylfuroic acid (HMFA) by *Lysinibacillus* sp. B2P (**S-3**)
3. **Figure S3.** Effect of substrate concentration on HMFA synthesis by *Pseudochrobactrum* sp. B2L and *Lysinibacillus* sp. B2P (**S-4**).
4. **Figure S4.** Effect of pH on the synthesis of HMFA from a high concentration of HMF substrate catalyzed by *Pseudochrobactrum* sp. B2L (A) and *Lysinibacillus* sp. B2P (**S-5**)

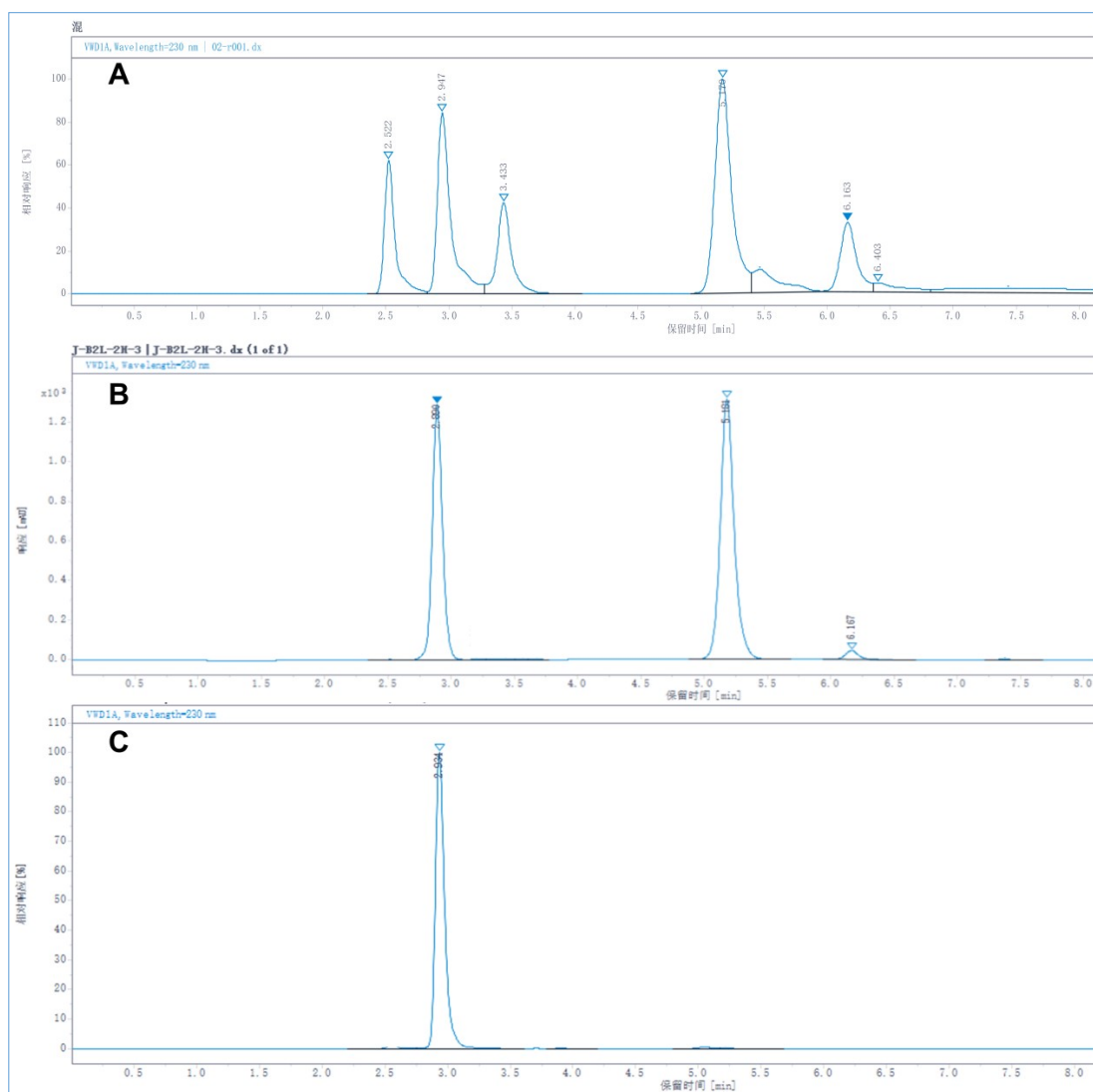


Figure S1. The selective oxidation of 5-hydroxymethylfurfural (HMF) to 5-hydroxymethylfuroic acid (HMFA) by *Pseudochrobactrum sp.* B2L. Retention time (RT) of commercially available standard substrates: HMF (6.163 min), 2,5-bis(hydroxymethyl)furan (BHMF)(5.170 min), 5-formyl-2-furancarboxylic acid (FFCA)(3.433 min), HMFA(2.947 min), 2,5-Furandicarboxylic acid (FDCA)(2.522 min)(A); the product of HMF oxidation after 2 h (B); the product of HMF oxidation after 24 h (C). Reaction conditions: 100 mM of HMF, 0.10 g/mL microbial cells, 100 mM of phosphate buffer (pH 7.4), 800 rpm, and 30 °C for 24 h.

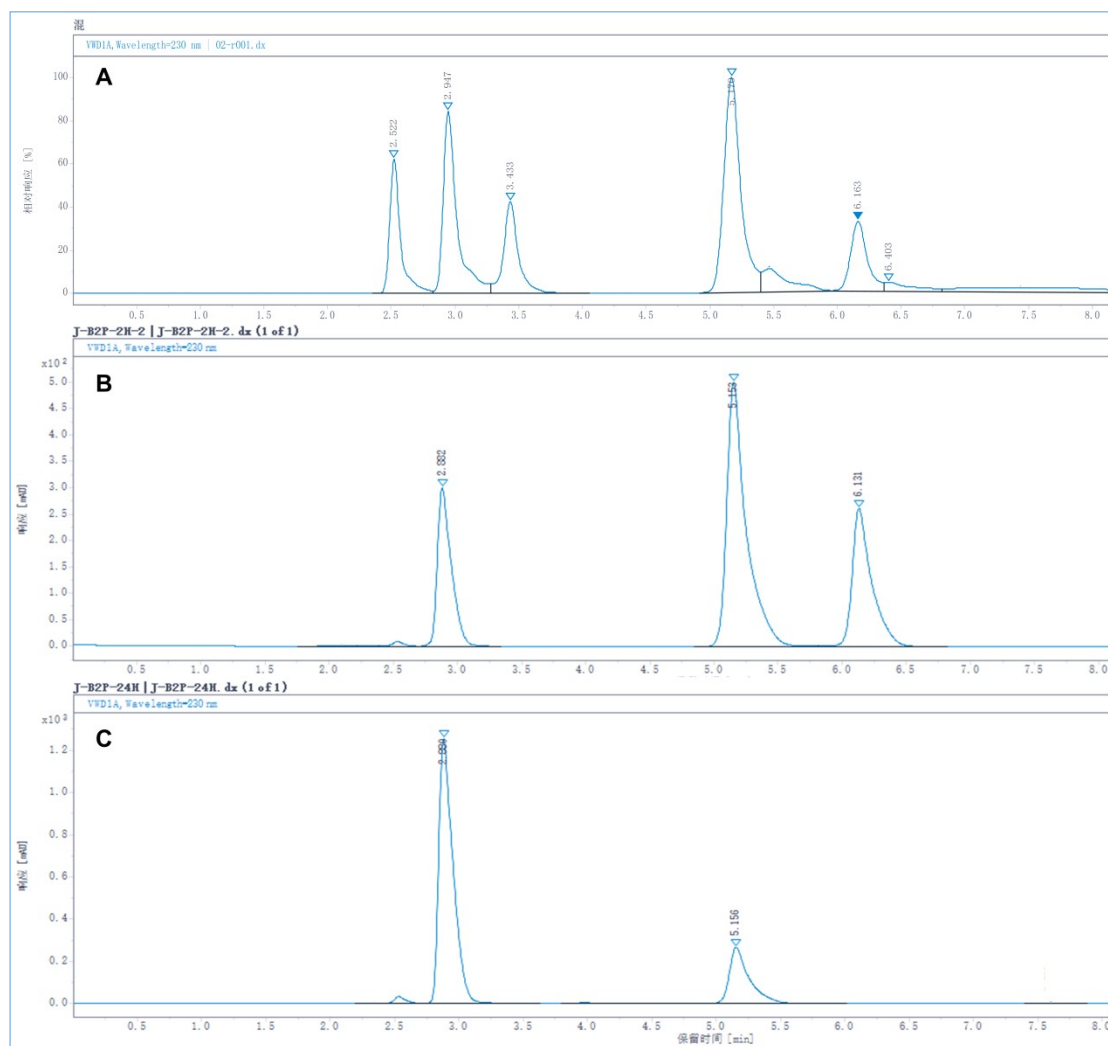


Figure S2. The selective oxidation of 5-hydroxymethylfurfural (HMF) to 5-hydroxymethylfuroic acid (HMFA) by *Lysinibacillus* sp. B2P. Retention time (RT) of commercially available standard substrates: HMF (6.163 min), 2,5-bis(hydroxymethyl)furan (BHMF)(5.170 min), 5-formyl-2-furancarboxylic acid (FFCA)(3.433 min), HMFA(2.947 min), 2,5-Furandicarboxylic acid (FDCA)(2.522 min)(A); the product of HMF oxidation after 2 h (B); the product of HMF oxidation after 24 h (C). Reaction conditions: 100 mM of HMF, 0.10 g/mL microbial cells, 100 mM of phosphate buffer (pH 7.4), 800 rpm, and 30 °C for 24 h.

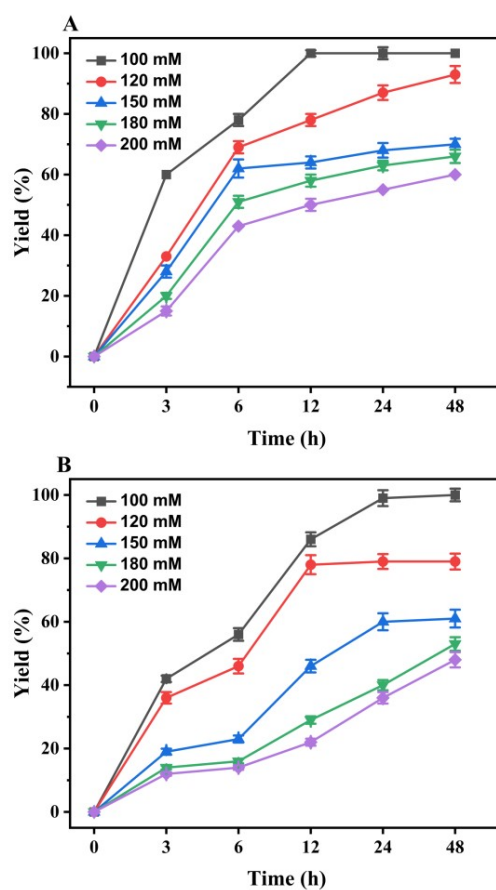


Figure S3. Effect of substrate concentration on HMFCA synthesis by *Pseudochrobactrum* sp. B2L (A) and *Lysinibacillus* sp. B2P (B). Reaction conditions: 0.2 g/mL microbial cells, 5 mL phosphate buffer (100 mM, pH 7.4), 800 rpm, and 20 °C / 25 °C.

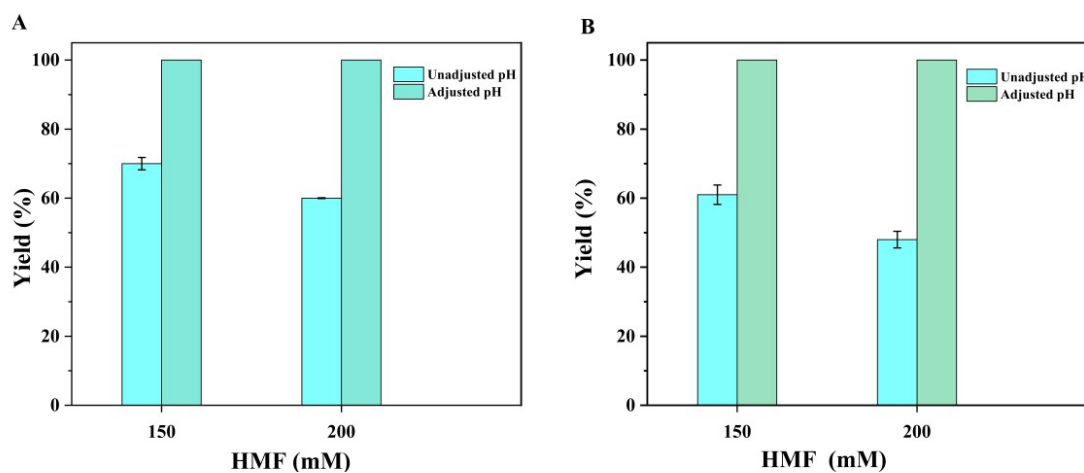


Figure S4. Effect of pH on the synthesis of HMFCa from a high concentration of HMF substrate catalyzed by *Pseudochrobactrum* sp. B2L (A) and *Lysinibacillus* sp. B2P (B). Reaction conditions: 0.2 g/mL microbial cells, 5 mL phosphate buffer (100 mM, pH 7.4), 800 rpm, and 20 °C / 25 °C for 48 h. The pH was adjusted to around 7.0 every 3 h for the first 12 h, then every 12 h for the next 36 h.