Experimental

General. Methyl *o*-chlorobenzoylformate (**2**) was synthesized according to the literature.[1] Methyl benzoylformate was purchased. *E. coli* BL21(DE3) cells harboring pESCR and pABGD were grown as reported previously.[2] $NADP^+$ was purchased from Oriental Yeast. Silica gel and basic alumina column chromatography was performed using Fuji Silysia BW-127 ZH (100–270 mesh) and Merck aluminum oxide 90 active basic (0.063–0.200 mm), respectively. Thin layer chromatography (TLC) was performed on Merck silica gel 60 F_{254}.

Typical procedure for the whole-cell asymmetric reduction of 2 to give methyl (*R*)-*o*-chloromandelate (1). To a mixture of glucose (3.60 g, 20.0 mmol), $NADP^+$ (10 mg, 12 μmol), and *E. coli* BL21(DE3) cells harboring pESCR and pABGD (2.0 g) in 0.1 M phosphate buffer (pH 7.0, 10 mL) was added methyl *o*-chlorobenzoylformate (**2**) (1.98 g, 10.0 mmol). The mixture was stirred in a water bath at 20 ˚C for 24 h, during which 2 N NaOH was added to neutralize the solution acidified by the progress of the reaction. Solid NaCl (5.5 g) was added, and the product was extracted with EtOAc (25 mL, 3 times). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated under reduced pressure. Purification by silica gel column chromatography (hexane:EtOAc (10:1)) gave methyl (*R*)-*o*-chloromandelate ((*R*)-**1**) as a colorless oil (1.78 g, 89%): $[\alpha]^{19}_D$ = –178.3 (*c* 1.3, $CHCl_3$), >99% ee, (*R*) (lit.[3] $[\alpha]^{22}_D$ = –146.3 (*c* 1.05, $CHCl_3$), 80.8% ee, (*R*)); HPLC: Chiralpak AD-H (Daicel Chemical Industries), hexane/*i*-PrOH (9:1), flow rate 0.5 mL/min, detection 254 nm, (*S*) 20.3 min, (*R*) 22.7 min; ^{1}H NMR ($CDCl_3$, 600 MHz) δ3.56 (d, *J* = 5.4 Hz, 1H), 3.78 (s, 3H), 5.57 (d, *J* = 5.4 Hz, 1H), 7.28–7.29 (m, 2H), 7.39–7.40 (m, 2H); ^{13}C NMR ($CDCl_3$, 150 MHz) δ53.2, 70.3, 127.2, 128.8, 129.8, 130.0, 133.5, 135.9, 173.7; IR (film) 3454, 1744, 1441, 1223 cm^{-1}.

Methyl (*R*)-mandelate (4). The whole-cell reduction of methyl benzoylformate was performed at 30 ºC on a 3.0 mmol scale as described above: Isolated yield 72%; $[\alpha]^{23}_D$ = –130.8 (*c* 1.0, MeOH), 96% ee, (*R*) (lit.[4] $[\alpha]^{20}_D$ = –124.8 (*c* 0.8, MeOH), 86% ee, (*R*)); GC: CP-cyclodextrin-β-2,3,6-M-19 (Chrompack, ϕ0.25 mm × 25 m), Inj. 300 ˚C, Col. 100 ˚C, Det. 250 ˚C, (*R*) 77.8 min, (*S*) 83.6 min; ^{1}H NMR (600 MHz, $CDCl_3$) δ3.42 (d, *J* = 5.4 Hz, 1H), 3.77 (s, 3H), 5.18 (d, *J* = 5.4 Hz, 1H), 7.34–7.43 (m, 5H); ^{13}C NMR (150 MHz, $CDCl_3$) δ53.1, 72.9, 126.6, 128.5, 128.6, 138.2, 174.1; IR (KBr) 3442, 3032, 2955, 1740, 1209, 739 cm^{-1}.

Ethyl (*R*)-*o*-chloromandelate (5). A solution of methyl *o*-chlorobenzoylformate (**2**) (3.01 g, 15.2 mmol) and H_2SO_4 (0.6 mL) in EtOH (380 mL) was heated at reflux for 4 d. After removal of EtOH by rotary evaporation, brine was added, and the product was extracted with EtOAc. The organic layer was dried over $MgSO_4$ and concentrated. The product was purified by basic alumina column chromatography (hexane/EtOAc (10:1)–(0:1)) to afford ethyl *o*-chlorobenzoylformate[5] as a pale yellow oil (1.42 g, 44%): ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.41 (t, J = 7.5 Hz, 3H), 4.43 (q, J = 7.5 Hz, 2H), 7.40–7.46 (m, 2H), 7.53 (ddd, J = 8.1, 7.8, 1.8 Hz, 1H), 7.77 (dd, J = 7.8, 1.8 Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 13.9, 62.8, 127.2, 130.5, 131.6, 133.3, 133.9, 134.3, 163.1, 186.6; IR (film) 2988, 1732, 1699, 1589, 1198, 1065, 1018, 760 cm^{-1}. The whole-cell reduction of ethyl *o*-chlorobenzoylformate was performed at 30 ºC on a 3.0 mmol scale as described above: Isolated yield 93%; $[\alpha]^{22}_D$ = –149.0 (*c* 1.0, $CHCl_3$), 99% ee, (*R*) (lit.[6] $[\alpha]^{25}_D$ = –45.0 (*c* 4.0, $CHCl_3$), 62% ee, (*R*)); HPLC: Chiralcel OD-H, hexane/*i*-PrOH (20:1), flow rate 1.0 mL/min, detection 254 nm, (*S*) 12.3 min, (*R*) 14.6 min; ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.22 (t, J = 6.9 Hz, 3H), 3.56 (d, J = 5.1 Hz, 1H), 4.18–4.30 (m, 2H), 5.55 (d, J = 5.1 Hz, 1H), 7.27–7.30 (m, 2H), 7.38–7.41 (m, 2H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 14.0, 62.5, 70.4, 127.1, 128.8, 129.7, 129.9, 133.6, 136.1, 173.2; IR (film) 3468, 2984, 1732, 1217, 1090, 756 cm^{-1}.

References

1 M. Ma, C. Li, L. Peng, F. Xie, X. Zhang and J. Wang, *Tetrahedron Lett*., 2005, **46**, 3927–3929.

2 T. Ema, H. Yagasaki, N. Okita, M. Takeda and T. Sakai, *Tetrahedron*, 2006, **62**, 6143–6149.

3 H. Ohta, Jpn. Kokai Tokkyo Koho, JP06165695A2, 1994; *Chem. Abstr*. 1994, **121**, 177884.

4 W. Yang, J.-H. Xu, Y. Xie, Y. Xu, G. Zhao and G.-Q. Lin, *Tetrahedron: Asymmetry*, 2006, **17**, 1769–1774.

5 Y. Sun, X. Wan, J. Wang, Q. Meng, H. Zhang, L. Jiang and Z. Zhang, *Org. Lett*., 2005, **7**, 5425–5427.

6 L. Tang and L. Deng, *J. Am. Chem. Soc.,* 2002, **124**, 2870–2871.