Microbial Deracemization of α -Substituted Carboxylic Acids: Substrate Specificity and Mechanistic Investigation

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Experimental section

General methods

Starting materials and reagents were purchased from standard vendors and used without purification unless otherwise noted. Analytical thin-layer chromatography (TLC) was developed on E. Merck Silica Gel 60 F₂₅₆ plates (0.25 mm thickness). Preparative TLC was performed using E. Merck Silica Gel 60 F₂₅₆ plates (0.5 mm thickness). Column chromatography was performed on Katayama Chemical Co., Inc. Silica Gel 60 K070-WH (70-230 mesh). NMR spectra were obtained on JEOL EX-270, GX-400 spectrometers (¹H at 270, 400 MHz and ¹³C at 100 MHz) at ambient temperature. ¹H chemical shifts are referenced with CDCl₃ at 7.26 ppm or CD₃OD at 3.30 ppm or D₂O at 4.80 ppm and ¹³C chemical shifts with CDCl₃ at 77.0 ppm. IR spectra were measured as films for oils and as KBr discs for solids with a jasco FT / IR-410 instrument. Optical rotations were measured on a jasco DIP 360 polarimeter. Mass spectra were recorded on a Hitachi M-80B spectrometer. Melting points were measured by Yanaco MP-S3. OD value was measured on a Shimadzu UV-Visible Recording Spectrometer (UV-2100S). The strain, *Nocardia diaphanozonaria* JCM3208, used in this experiment is available from the Japan Collection of Microorganism: The Institute of Physical and Chemical Research (Riken), 2-1 Hirosawa, Wako 351-0106, Japan.

Ethyl 2-(4-bromophenoxy)propanoate (17d)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-bromophenoxy)propanoate (0.75 g, 93% yield) was obtained as a colorless oil starting from 4-bromophenol (0.51 g, 3.2 mmol); IR (film) 2986, 2938, 1753, 1589, 1488, 1376, 1282, 1240, 1198, 1134, 1073, 1051, 1005, 822, 640 cm⁻¹; ¹H NMR (CDCl₃) δ 7.36 (d, *J* = 8.8 Hz, 2H), 7.66 (d, *J* = 8.8 Hz, 2H), 4.69 (q, *J* = 6.8 Hz, 1H), 4.21 (q, *J* = 7.3 Hz, 2H), 1.61 (d, *J* = 6.8 Hz, 2H), 1.25 (t, *J* = 7.3 Hz, 3H).

Ethyl 2-(4-iodophenoxy)propanoate (17e)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-iodophenoxy)propanoate (0.52 g, 73% yield) was obtained as a colorless oil starting from 4-iodophenol (0.49 g, 2.2 mmol) ; IR (film) 2984, 2937, 1749, 1584, 1486, 1376, 1280, 1239, 1196, 1134, 1097, 1051, 821, 632 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.54 (m, 2H), 6.65 (m, 2H), 4.69 (q, $J = 6.8$ Hz, 1H), 4.21 (q, $J = 7.3$ Hz, 2H), 1.61 (d, $J = 6.8$ Hz, 2H), 1.25 (t, $J = 7.3$ Hz, 3H).

Ethyl 2-(4-trifluoromethylphenoxy)propanoate (17f)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-trifluoromethylphenoxy)propanoate (0.69 g, 85% yield) was obtained as a colorless oil starting from 4-trifluoromethylphenol (0.50 g, 3.1 mmol) ; IR (film) 2989, 2942, 1752, 1616, 1519, 1448, 1378, 1330, 1254, 1163, 1114, 1067, 838, 631 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.54 (d, $J = 8.3$ Hz, 2H), 6.93 (d, $J = 8.3$ Hz, 2H), 4.79 (q, $J = 6.8$ Hz, 1H), 4.22 (q, $J = 6.8$ Hz, 2H), 1.64 (d, $J = 6.8$ Hz, 2H), 1.25 (t, $J = 6.8$ Hz, 3H).

Ethyl 2-(4-methylphenoxy)propanoate (17g)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-methylphenoxy)propanoate (0.18 g, 26% yield) was obtained as a colorless oil starting from 4-methylphenol (0.45 g, 4.2 mmol) ; IR (film) 2986, 1754, 1614, 1511, 1287, 1237, 1195, 1135, 1053, 817, 742 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.06 (d, $J = 8.3$ Hz, 2H), 6.78 (td, $J = 2.9, 8.8$ Hz, 2H), 4.70 (q, $J = 6.8$ Hz, 1H), 4.21 (q, $J = 6.8$ Hz, 2H), 2.27 (s, 3H), 1.60 (d, $J = 6.8$ Hz, 3H), 1.25 (t, $J = 6.8$ Hz, 3H).

Ethyl 2-(4-methoxyphenoxy)propanoate (17h)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-methoxyphenoxy)propanoate (0.96 g, 88% yield) was obtained as a colorless oil starting from 4-methoxyphenol (0.60 g, 4.8 mmol) ; IR (film) 2987, 2938, 1752, 1507, 1465, 1376, 1230, 1135, 1099, 1037, 947, 825, 739 cm^{-1} ; ^1H NMR (CDCl_3) δ 6.81 (m, 4H), 4.64 (q, $J = 6.8$ Hz, 1H), 4.20 (q, $J = 7.3$ Hz, 2H), 3.76 (s, 3H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.25 (t, $J = 7.3$ Hz, 3H).

Ethyl 2-(4-acetylphenoxy)propanoate (17i)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-acetylphenoxy)propanoate (0.44 g, 42% yield) was obtained as a colorless oil starting from 4-acetylphenol (0.59 g, 4.4 mmol) ; IR (film) 2992, 2939, 1749, 1669, 1603, 1505, 1419, 1354, 1275, 1248, 1200, 1180, 1137, 1097, 1021, 955, 838, 591 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.91 (d, $J = 8.8$ Hz, 2H), 6.89 (d, $J = 8.8$ Hz, 2H), 4.82 (q, $J = 6.8$ Hz, 1H), 4.22 (q, $J = 7.3$ Hz, 2H), 2.55 (s, 3H), 1.64 (d, $J = 6.8$ Hz, 3H), 1.24 (t, $J = 7.3$ Hz, 3H).

Ethyl 2-(4-hydroxyphenoxy)propanoate (17j)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(4-benzyloxyphenoxy)propanoate (0.95 g, 91% yield) was obtained as a colorless oil starting from 4-benzyloxyphenol (0.70 g, 3.5 mmol) ; IR (film) 3063, 2985, 2871, 1748, 1592, 1506, 1455, 1377, 1276, 1216, 1134, 1098, 1050, 1017, 823, 742, 697 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.37 (m, 5H), 6.84 (m, 4H), 5.00 (s, 2H), 4.65 (q, $J = 6.8$ Hz, 1H), 4.21 (q, $J = 7.3$ Hz, 2H), 1.59 (d, $J = 6.8$ Hz, 3H), 1.25 (t, $J = 7.3$ Hz, 3H).

A mixture of this compound (0.50 g, 1.7 mmol) in ethanol (10 mL) containing 10% palladium on carbon (100 mg) was stirred vigorously in a H_2 atmosphere under balloon pressure at room temperature. After 5 h, the mixture was diluted with ethanol, filtered and concentrated *in vacuo* to give ethyl 2-(4-hydroxyphenoxy)propanoate (0.36 g, quant.) as a colorless oil; IR (film) 3421, 2985, 2359, 1732, 1508, 1455, 1376, 1214, 1135, 947, 827, 763 cm^{-1} ; ^1H NMR (CDCl_3) δ 6.75 (m, 5H), 4.63 (q, $J = 6.8$ Hz, 1H), 4.21 (q, $J = 7.3$ Hz, 2H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.25 (t, $J = 7.3$ Hz, 3H).

Ethyl 2-(2-naphthoxy)propanoate (18)

According to the same procedure as described above for the synthesis of **17b**, ethyl 2-(2-naphthoxy)propanoate (0.25 g, 32% yield) was obtained as a colorless oil starting from 2-naphthol (0.51

g, 3.5 mmol) ; IR (film) 3059, 2985, 2938, 1754, 1631, 1601, 1510, 1469, 1257, 1216, 1182, 1133, 1095, 1050, 971, 839, 749 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.76 (d, $J = 9.3$ Hz, 2H), 7.69 (d, $J = 8.3$ Hz, 1H), 7.43 (m, 1H), 7.35 (m, 1H), 7.20 (dd, $J = 2.4, 8.8$ Hz, 1H), 7.05 (d, $J = 2.4$ Hz, 1H), 4.89 (q, $J = 6.8$ Hz, 1H), 4.23 (m, 2H), 1.68 (d, $J = 6.8$ Hz, 3H), 1.25 (t, $J = 7.3$ Hz, 3H).

Methyl 2-(benzyloxy)propanoate (19)

DBU (0.15 mL, 1.0 mmol) was added dropwise over 5 min to a solution of benzyl alcohol (1 mL, 9.7 mmol) and trichloroacetonitrile (4.8 mL, 48 mmol) in dichloromethane (10 mL) cooled in an ice bath. The reaction mixture was allowed to warm to room temperature and was stirred for further 3 h. The solvent was concentrated *in vacuo*, and the residue was purified by silica gel column chromatography (hexane/EtOAc = 19/1) and distilled (150 $^{\circ}\text{C}$ / 1 mmHg) to give benzyl 3,3,3-trichloroacetoimide (1.6 g, 80% yield) as a colorless oil.

Trifluoromethanesulfonic acid (catalytic amounts) was added to a stirred solution of methyl lactate (0.2 g, 1.96 mmol) and benzyl 3,3,3-trichloroacetoimide (1.0 g, 3.96 mmol) in cyclohexane-dichloromethane (2: 1; 2.5 mL) and the reaction mixture was stirred for 40 h at room temperature. The reaction mixture was filtered through a pad of celite and extracted with EtOAc. The organic layer was washed with saturated sodium hydrogen carbonate solution and brine, dried over anhydrous sodium sulfate and concentrated *in vacuo*. The residue was purified by silica gel column chromatography (hexane/EtOAc = 29/1) to give methyl 2-(benzyloxy)propanoate (0.27 g, 72% yield) as a colorless oil; IR (film) 3031, 2952, 2873, 1748, 1496, 1454, 1274, 1207, 1143, 1065, 1026, 740, 699 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.20 (m, 5H), 4.59 (d, $J = 11.7$ Hz, 1H), 4.35 (d, $J = 11.7$ Hz, 1H), 3.97 (q, $J = 6.8$ Hz, 1H), 3.66 (s, 3H), 1.34 (t, $J = 6.8$ Hz, 3H).

Ethyl 2-(phenylthio)propanoate (20a)

Sodium hydride (60% in paraffin, 0.47 g, 11.8 mmol) was stirred for 5 min in hexane (5 mL) under Ar, then the solvent was removed by a syringe followed by evaporation with a vacuum pump. After

repeating the same operation for three times, the residue was suspended in tetrahydrofuran (5 mL) and the mixture was cooled to 0 °C. To this mixture, a solution of thiophenol (1.0 g, 9.7 mmol) in tetrahydrofuran (10 mL) was added dropwise over 10 min, and the mixture was stirred for 5 min at the same temperature. Then the mixture was allowed to warm to room temperature, and stirred for additional 15 min. A solution of ethyl 2-bromopropanoate (3.5 g, 19.3 mmol) in tetrahydrofurane (3 mL) was added and the mixture was stirred for 6 h. The reaction mixture was acidified by 2 *N* hydrochloric acid and extracted with EtOAc. The organic layer was washed with saturated sodium hydrogen carbonate solution and brine, dried over anhydrous sodium sulfate and concentrated *in vacuo*. The residue was purified by silica gel column chromatography (hexane/EtOAc = 19/1) to give ethyl 2-phenylthiopropoanoate (1.9 g, 99% yield) as a colorless oil; IR (film) 3060, 2980, 2933, 1731, 1583, 1441, 1322, 1256, 1160, 1065, 859, 748, 692 cm⁻¹; ¹H NMR (CDCl₃) δ 7.39 (m, 2H), 7.22 (m, 2H), 4.04 (q, *J* = 7.1 Hz, 2H), 3.72 (q, *J* = 7.1 Hz, 1H), 1.41 (d, *J* = 7.1 Hz, 3H), 1.10 (t, *J* = 7.1 Hz, 3H).

Methyl 2-methyl-3-phenylpropanoate (21)

According to the same procedure as described above for the synthesis of **2h**, methyl 2-methyl-3-phenylpropanoate (0.31 g, 47% yield) was obtained as a colorless oil starting from methyl 3-phenylpropanoate (0.60 g, 3.7 mmol); IR (film) 3028, 2975, 2951, 1739, 1496, 1455, 1167, 986, 832, 745, 701 cm⁻¹; ¹H NMR (CDCl₃) δ 7.29 (m, 2H), 7.19 (m, 2H), 3.64 (s, 3H), 3.03 (q, *J* = 6.8 Hz, 1H), 2.70 (m, 2H), 1.15 (d, *J* = 6.8 Hz, 2H).

2-(4-Bromophenoxy)propanoic acid (10d)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-bromophenoxy)propanoic acid (0.25 g, 90% yield) was obtained as a colorless solid starting from **17d** (0.31 g, 1.1 mmol); IR (KBr disc) 2995, 2542, 1714, 1586, 1486, 1283, 1224, 1138, 923, 823, 642, 504 cm⁻¹.

2-(4-Iodophenoxy)propanoic acid (10e)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-iodophenoxy)propanoic acid (0.23 g, 94% yield) was obtained as a colorless solid starting from **17e** (0.26 g, 0.82 mmol); IR (KBr disc) 2983, 2646, 1702, 1584, 1486, 1281, 1235, 1182, 1134, 1094, 907, 817, 632, 504 cm^{-1} .

2-(4-Trifluoromethylphenoxy)propanoic acid (10f)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-trifluoromethylphenoxy)propanoic acid (0.22 g, 99% yield) was obtained as a colorless solid starting from **17f** (0.25 g, 0.96 mmol); IR (KBr disc) 2995, 2649, 1719, 1618, 1521, 1335, 1236, 1113, 1068, 838, 638 cm^{-1} .

2-(4-Methylphenoxy)propanoic acid (10g)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-methylphenoxy)propanoic acid (0.13 g, 94% yield) was obtained as a colorless solid starting from **17g** (0.16 g, 0.76 mmol); IR (KBr disc) 2997, 1715, 1613, 1509, 1453, 1291, 1225, 1138, 1097, 1044, 920, 807, 749, 682, 510 cm^{-1} .

2-(4-Methoxyphenoxy)propanoic acid (10h)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-methoxyphenoxy)propanoic acid (0.22 g, 99% yield) was obtained as a colorless solid starting from **17h** (0.26 g, 1.1 mmol); IR (KBr disc) 2950, 2643, 1717, 1507, 1450, 1290, 1220, 1140, 1044, 921, 821, 739, 682, 525 cm^{-1} .

2-(4-Acetylphenoxy)propanoic acid (10i)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-acetylphenoxy)propanoic acid (0.22 g, quant.) was obtained as a colorless solid starting from **17i** (0.25 g, 1.1 mmol); IR (KBr disc) 2993, 2611, 1754, 1651, 1595, 1512, 1368, 1317, 1263, 1212, 1175, 1098, 964, 836, 739, 640, 590 cm^{-1} .

2-(4-Hydroxyphenoxy)propanoic acid (10j)

According to the same procedure as described above for the synthesis of **10b**, 2-(4-hydroxyphenoxy)propanoic acid (0.21 g, 95% yield) was obtained as a colorless solid starting from **17j** (0.25 g, 1.2 mmol); IR (KBr disc) 3266, 2989, 2606, 1708, 1510, 1454, 1371, 1232, 1198, 1135, 822, 762, 672, 507 cm^{-1} .

2-(2-Naphthoxy)propanoic acid (12)

According to the same procedure as described above for the synthesis of **10b**, 2-(2-naphthoxy)propanoic acid (0.13 g, 91% yield) was obtained as a colorless solid starting from **18** (0.16 g, 0.64 mmol); IR (KBr disc) 2994, 2642, 1704, 1629, 1599, 1509, 1466, 1390, 1256, 1215, 1182, 1136, 953, 837, 747, 624, 471 cm^{-1} .

2-(Benzyloxy)propanoic acid (13)

According to the same procedure as described above for the synthesis of **10b**, 2-(benzyloxy)propanoic acid (0.19 g, quant.) was obtained as a colorless oil starting from **19** (0.20 g, 1.1 mmol); IR (film) 3032, 2634, 1718, 1455, 1210, 1118, 1063, 1019, 737, 699 cm^{-1} .

2-(phenylthio)propanoic acid (14a)

According to the same procedure as described above for the synthesis of **10b**, 2-phenylthiopropionic acid (0.49 g, quant.) was obtained as a colorless oil starting from **20a** (0.50 g, 2.4 mmol); IR (film) 2980, 2625, 1717, 1583, 1440, 1284, 1239, 1186, 1025, 748, 691 cm^{-1} .

2-Methyl-3-phenylpropanoic acid (15)

According to the same procedure as described above for the synthesis of **1h**, 2-methyl-3-phenylpropanoic acid (0.15 g, quant.) was obtained as a colorless oil starting from **21** (0.16 g, 0.9 mmol); IR (film) 2977, 2664, 1705, 1455, 1292, 1240, 939, 911, 744, 699 cm^{-1} .

Methyl (R)-(-)-2-phenylpropanoate ((R)-2a)

Incubation time 48 h; yield 81% yield (colorless oil): IR (film) 2981, 1738, 1602, 1454, 1208, 1167, 1067, 699, 513 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.25 (m, 5H), 3.68 (q, J = 7.1 Hz, 1H), 3.61 (s, 3H), 1.46 (d, J = 7.1 Hz, 3H); ^{13}C NMR (CDCl_3) δ 174.8 (CO), 140.4 (C), 128.5 (CH), 127.3 (CH $\times$ 2), 127.0 (CH $\times$ 2), 52.0 (CH), 45.4 (CH_3), 18.6 (CH_3); HRMS exact mass calcd for $\text{C}_{10}\text{H}_{12}\text{O}_2$ requires m/z 164.0837, found m/z 164.0799. Ee 69% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*S*)-(minor) t_R = 17.3 min, (*R*)-(major) t_R = 19.2), $[\alpha]_D^{24}$ -71.3 (c 1.03, EtOH), lit¹⁶, *R* form, 98% ee, $[\alpha]_D$ -96 (c 1.1, EtOH).

Methyl (R)-(-)-2-(4-fluorophenyl)propanoate ((R)-2h)

Incubation time 16 h; yield 58% yield (colorless oil): IR (film) 2983, 2953, 1739, 1604, 1510, 1456, 1436, 1224, 1170, 1070, 838, 732, 551 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.19 (dt, J = 2.0, 8.8 Hz, 2H), 6.94 (tt, J = 2.0, 8.8 Hz, 2H), 3.64 (q, J = 6.8 Hz, 1H), 3.59 (s, 3H), 1.41 (d, J = 6.8 Hz, 3H); ^{13}C NMR (CDCl_3) δ 171.8 (CO), 160.7 (C), 130.8 (CH), 130.7 (CH), 129.6 (C), 115.5 (CH), 115.3 (CH $\times$ 2), 65.9 (CH), 52.1 (CH_3), 40.3 (CH_3); HRMS exact mass calcd for $\text{C}_{10}\text{H}_{11}\text{FO}_2$ requires m/z 182.0743, found m/z 182.0744. Ee 6% (Daicel Chiralcel OJ column (50/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 17.6 min, (*S*)-(minor) t_R = 18.8), $[\alpha]_D^{26}$ -5.6 (c 1.00, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (R)-(-)- α -fluorophenylacetate ((R)-7a)

Incubation time 48 h; yield 74% yield (colorless oil): Ee 55% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 22.1 min, (*S*)-(minor) t_R = 24.4). The configuration was revealed to be *R* judging from the literature data¹⁷.

Methyl (*R*)-(-)-mandelate ((*R*)-9a)

Incubation time 48 h; yield 45% yield (colorless oil): IR (film) 3445, 3033, 2952, 1739, 1435, 1264, 1204, 1188, 1096, 1068, 984, 894, 735, 696, 508 cm⁻¹; ¹H NMR (CDCl₃) δ 7.38 (m, 5H), 5.18 (s, 1H), 3.76 (s, 3H), 3.43 (br s, 1H). Ee >99% (No *S*-enantiomer could be detected.) (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 32.6 min, (*S*)-(minor) t_R = 37.1), $[\alpha]^{18}_D$ -113.6 (c 1.42, acetone), lit¹⁸, *R* form, -115.4 (c 1.0, acetone).

Methyl (*R*)-(-)-(4-chloro)mandelate ((*R*)-9b)

Incubation time 48 h; yield 43% yield (colorless oil): IR (film) 3327, 2951, 1738, 1489, 1438, 1489, 1438, 1412, 1260, 1217, 1092, 1006, 834, 775, 641, 546 cm⁻¹; ¹H NMR (CDCl₃) δ 7.35 (m, 5H), 5.16 (s, 1H), 3.77 (s, 3H), 3.46 (br s, 1H). Ee >99% (No *S*-enantiomer could be detected.) (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 28.4 min, (*S*)-(minor) t_R = 30.5), $[\alpha]^{21}_D$ -96.3 (c 0.38, acetone), lit¹⁸, *R* form, -96.7 (c 1.0, acetone).

Methyl (*R*)-(-)-(4-methoxy)mandelate ((*R*)-9c)

Incubation time 48 h; yield 38% yield (colorless oil): IR (film) 3403, 2958, 2842, 1739, 1675, 1599, 1513, 1461, 1307, 1259, 1166, 1081, 1028, 833, 618 cm⁻¹; ¹H NMR (CDCl₃) δ 6.97 (m, 2H), 6.90 (m, 2H), 5.13 (s, 1H), 3.80 (s, 3H), 3.75 (s, 3H), 3.39 (br s, 1H). Ee >99% (No *S*-enantiomer could be detected.) (Daicel Chiralcel OD column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*S*)-(minor) t_R = 24.8, (*R*)-(major) t_R = 44.2 min), $[\alpha]^{21}_D$ -102.6 (c 0.19, acetone), lit¹⁸, *R* form, -110.2 (c 1.0, acetone).

Methyl (*R*)-(+)-2-phenoxypropanoate ((*R*)-11a)

Incubation time 24 to 84 h; yield 66 to 86% yield (colorless oil): IR (film) 2992, 2953, 1758, 1589, 1494, 1449, 1283, 1240, 1211, 1135, 1099, 1053, 978, 754, 691 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.28 (m, 2H), 6.98 (m, 1H), 6.88 (m, 2H), 4.77 (q, $J = 6.8$ Hz, 1H), 3.76 (s, 3H), 1.62 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.6 (CO), 157.4 (C), 129.5 ($\text{CH} \times 2$), 121.6 (C), 115.0 ($\text{CH} \times 2$), 72.5 (CH), 52.3 (CH_3), 18.7 (CH_3); HRMS exact mass calcd for $\text{C}_{10}\text{H}_{12}\text{O}_3$ requires m/z 180.0786, found m/z 180.0780. Ee 75% (entry 4, incubation time 48 h), 97% ee (entry 6, incubation time 72 h) (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) $t_R = 22.6$ min, (*S*)-(minor) $t_R = 32.1$), $[\alpha]_D^{21} +41.0$ (c 0.71, EtOH) (entry 2, incubation time 48 h), $[\alpha]_D^{26} +41.2$ (c 0.58, CHCl_3) (entry 4, incubation time 72 h), lit¹⁵, *R* form, 97% ee, $[\alpha]_D +41.2$ (c 0.76, CHCl_3).

Methyl (*R*)-(+)-2-(4-fluorophenoxy)propanoate ((*R*)-11b)

Incubation time 48 h; yield 64% yield (colorless oil): IR (film) 2994, 2955, 1758, 1505, 1448, 1283, 1216, 1135, 1098, 1053, 979, 829, 749 cm^{-1} ; ^1H NMR (CDCl_3) δ 6.95 (m, 2H), 6.82 (m, 2H), 4.69 (q, $J = 6.8$ Hz, 1H), 3.75 (s, 3H), 1.60 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.4 (CO), 156.4 (C), 153.5 (C), 116.4 (CH), 116.3 (CH), 116.0 (CH), 115.8 (CH), 73.4 (CH), 52.3 (CH_3), 18.6 (CH_3); HRMS exact mass calcd for $\text{C}_{10}\text{H}_{11}\text{FO}_3$ requires m/z 198.0691, found m/z 198.0697. Ee 96% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) $t_R = 18.6$ min, (*S*)-(minor) $t_R = 29.7$), $[\alpha]_D^{26} +52.0$ (c 1.02, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(4-chlorophenoxy)propanoate ((*R*)-11c)

Incubation time 48 h; yield 95% yield (colorless oil): IR (film) 2954, 1758, 1596, 1492, 1283, 1240, 1135, 1090, 979, 825, 667 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.25 (dd, $J = 3.4, 9.9$ Hz, 2H), 6.83 (dd, $J = 3.4, 9.9$ Hz, 2H), 5.50 (q, $J = 6.8$ Hz, 1H), 3.78 (s, 3H), 1.64 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.2 (CO), 156.0 (C), 129.4 ($\text{CH} \times 2$), 126.5 (C), 116.3 ($\text{CH} \times 2$), 72.9 (CH), 52.4 (CH_3), 18.6 (CH_3); HRMS

exact mass calcd for $C_{10}H_{11}ClO_3$ requires m/z 214.0396, found m/z 214.0411. Ee 97% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 18.5 min, (*S*)-(minor) t_R = 25.0), $[\alpha]_D^{20}$ +44.7 (c 0.94, EtOH), lit^{4b}, *S* form, 96% ee, -41.1 (c 50, EtOH).

Methyl (*R*)-(+)-2-(4-bromophenoxy)propanoate ((*R*)-11d)

Incubation time 48 h; yield 83% yield (colorless oil): IR (film) 2992, 2953, 1758, 1590, 1488, 1282, 1239, 1134, 1099, 1074, 1052, 1005, 823, 640 cm^{-1} ; 1H NMR ($CDCl_3$) δ 7.36 (dt, J = 3.4, 9.6 Hz, 2H), 6.75 (dt, J = 3.4, 9.6 Hz, 2H), 4.72 (q, J = 6.8 Hz, 1H), 3.75 (s, 3H), 1.61 (d, J = 6.8 Hz, 3H); ^{13}C NMR ($CDCl_3$) δ 172.1 (CO), 156.7 (C), 132.3 (CH $\times$ 2), 116.8 (CH $\times$ 2), 113.8 (C), 72.8 (CH), 52.4 (CH₃), 18.5 (CH₃); HRMS exact mass calcd for $C_{10}H_{11}BrO_3$ requires m/z 257.9890, found m/z 257.9861. Ee 99% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 19.8 min, (*S*)-(minor) t_R = 23.3), $[\alpha]_D^{26}$ +47.5 (c 1.00, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(4-iodophenoxy)propanoate ((*R*)-11e)

Incubation time 48 h; yield 76% yield (colorless oil): IR (film) 2990, 2952, 1757, 1584, 1487, 1447, 1280, 1238, 1134, 1098, 1052, 821, 632 cm^{-1} ; 1H NMR ($CDCl_3$) δ 7.54 (dt, J = 3.4, 9.8 Hz, 2H), 6.64 (dt, J = 3.4, 9.8 Hz, 2H), 4.72 (q, J = 6.8 Hz, 1H), 3.75 (s, 3H), 1.61 (d, J = 6.8 Hz, 3H); ^{13}C NMR ($CDCl_3$) δ 172.1 (CO), 157.3 (C), 138.3 (CH $\times$ 2), 117.3 (CH $\times$ 2), 83.9 (C), 72.6 (CH), 52.4 (CH₃), 18.5 (CH₃); HRMS exact mass calcd for $C_{10}H_{11}IO_3$ requires m/z 305.9753, found m/z 305.9703. Ee 97% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) t_R = 23.1 min, (*S*)-(minor) t_R = 25.9), $[\alpha]_D^{26}$ +43.4 (c 1.01, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(4-trifluoromethylphenoxy)propanoate ((*R*)-11f)

Incubation time 48 h; yield 64% yield (colorless oil): IR (film) 2996, 2957, 1759, 1615, 1519, 1450, 1332, 1254, 1214, 1164, 1121, 1068, 838, 632 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.53 (d, J = 8.8 Hz, 2H), 6.92 (d, J = 8.8 Hz, 2H), 4.81 (q, J = 6.8 Hz, 1H), 3.76 (s, 3H), 1.65 (d, J = 6.8 Hz, 3H); ^{13}C NMR (CDCl_3) δ 171.9 (CO), 159.8 (C), 127.0 ($\text{CH} \times 2$), 126.9 (CF_3), 125.5 (C), 114.8 ($\text{CH} \times 2$), 72.5 (CH), 52.5 (CH_3), 18.5 (CH_3); HRMS exact mass calcd for $\text{C}_{11}\text{H}_{11}\text{F}_3\text{O}_3$ requires m/z 248.0659, found m/z 248.0639. Ee 99% (Daicel Chiralcel OJ column (120/1 hexane/2-propanol; 0.5 mL/min; 210 nm): (*R*)-(major) t_R = 23.4 min, (*S*)-(minor) t_R = 34.8), $[\alpha]_D^{26}$ +43.3 (c 1.05, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(4-methylphenoxy)propanoate ((*R*)-11g)

Incubation time 48 h; yield 69% yield (colorless oil): IR (film) 3030, 2993, 2952, 2869, 1759, 1615, 1509, 1449, 1284, 1238, 1200, 1177, 1134, 1098, 979, 820 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.06 (d, J = 8.3 Hz, 2H), 6.77 (d, J = 8.3 Hz, 2H), 4.72 (q, J = 6.8 Hz, 1H), 3.75 (s, 3H), 2.28 (s, 3H), 1.60 (d, J = 6.8 Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.8 (CO), 155.3 (C), 130.9 (CH), 129.9 ($\text{CH} \times 2$), 114.9 ($\text{C} \times 2$), 72.7 (CH), 52.3 (CH_3), 20.5 (CH_3), 18.7 (CH_3). Ee 90.1% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 1.0 mL/min; 254 nm): (*R*)-(major) t_R = 11.5 min, (*S*)-(minor) t_R = 20.1), $[\alpha]_D^{26}$ +53.3 (c 1.11, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(4-methoxyphenoxy)propanoate ((*R*)-11h)

Incubation time 48 h; yield 83% yield (colorless oil): IR (film) 2996, 2952, 2905, 2837, 1758, 1507, 1457, 1283, 1230, 1202, 1135, 1101, 1037, 826 cm^{-1} ; ^1H NMR (CDCl_3) δ 6.75 (m, 4H), 4.60 (q, J = 6.8 Hz, 1H), 3.69 (s, 3H), 3.68 (s, 3H), 1.52 (d, J = 6.8 Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.8 (CO), 154.3 (C), 151.5 (C), 116.4 ($\text{CH} \times 2$), 114.6 ($\text{CH} \times 2$), 73.6 (CH), 55.7 (CH_3), 52.3 (CH_3), 18.7 (CH_3). Ee 43.6% (Daicel Chiralcel OD column (9/1 hexane/2-propanol; 1.0 mL/min; 210 nm): (*S*)-(minor) t_R = 7.15, (*R*)-

(major) $t_R = 9.92$ min), $[\alpha]_D^{26} +34.0$ (c 1.07, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-(2-naphthoxy)propanoate ((*R*)-22)

Incubation time 48 h; yield 78% yield (colorless oil): IR (film) 3054, 2993, 2949, 2359, 1733, 1633, 1507, 1391, 1259, 1213, 1182, 1132, 1095, 1050, 980, 839, 752 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.68 (d, $J = 8.8$ Hz, 2H), 7.62 (d, $J = 8.3$ Hz, 1H), 7.36 (dt, $J = 1.5, 7.4$ Hz, 1H), 7.27 (dt, $J = 1.5, 7.4$ Hz, 1H), 7.12 (dd, $J = 2.4, 9.3$ Hz, 1H), 6.97 (d, $J = 2.4$ Hz, 1H), 4.85 (q, $J = 6.8$ Hz, 1H), 3.69 (s, 3H), 1.61 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.6 (CO), 155.3 (C), 134.2 (CH), 129.6 (CH), 129.2 (CH), 127.5 (CH), 126.8 (CH), 126.4 (CH), 123.9 (CH), 118.8 (C), 107.5 (C), 72.6 (CH), 52.4 (CH_3), 18.7 (CH_3). Ee 18.8% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 1.0 mL/min; 254 nm): (*R*)-(major) $t_R = 16.8$ min, (*S*)-(minor) $t_R = 19.3$), $[\alpha]_D^{20} +19.4$ (c 1.38, EtOH). The configuration was tentatively estimated comparing the sign of the optical rotation with those of others^{2,4a}.

Methyl (*R*)-(+)-2-benzyloxypropanoate ((*R*)-19)

Incubation time 48 h; yield 77% yield (colorless oil): IR (film) 3031, 2952, 2873, 1748, 1496, 1454, 1274, 1207, 1143, 1065, 1026, 740, 699 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.20 (m, 5H), 4.59 (d, $J = 11.7$ Hz, 1H), 4.35 (d, $J = 11.7$ Hz, 1H), 3.97 (q, $J = 6.8$ Hz, 1H), 3.66 (s, 3H), 1.34 (t, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 173.6 (CO), 137.4 (CH), 128.3 ($\text{CH} \times 2$), 127.9 (CH), 127.8 (CH), 73.9 ($\text{CH}_2 \times 2$), 72.0 (CH), 52.0 (CH_3), 18.8 (CH_3). Ee 21.3% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) $t_R = 29.3$ min, (*S*)-(minor) $t_R = 34.6$), $[\alpha]_D^{21} +18.1$ (c 1.50, CHCl_3), lit¹⁹, *R* form, +151 (c 1.5, CHCl_3).

Incubation of 2-(phenylthio)propanoic acid (14a)

Incubation of **11a** with the bacterium according to the general procedure for 48 h and the corresponding methyl esters of the products were obtained as oily residue. The yield and the spectral

data of the esters are as follows; Methyl (*R*)-(+)-2-(phenylthio)propanoate ((*R*)-23a): Yield 49% (colorless oil): IR (film) 3059, 2989, 2952, 1733, 1437, 1330, 1260, 1227, 1191, 1161, 1067, 854, 749, 691 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.38 (m, 2H), 7.23 (m, 2H), 3.72 (q, $J = 6.8$ Hz, 1H), 3.60 (s, 3H), 1.41 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (CDCl_3) δ 172.9 (CO), 136.6 (C), 133.0 ($\text{CH} \times 2$), 128.8 ($\text{CH} \times 2$), 128.0 (C), 52.3 (CH), 45.2 (CH_3), 17.5 (CH_3). Ee 93.0% (Daicel Chiralcel OJ column (9/1 hexane/2-propanol; 0.5 mL/min; 254 nm): (*R*)-(major) $t_R = 30.1$ min, (*S*)-(minor) $t_R = 39.7$), $[\alpha]^{23}_D +145.7$ (c 1.02, EtOH), $[\alpha]^{22}_D +141.2$ (c 0.68, acetone), lit²⁰, *R* form, $+65.5$ (c 1.0, acetone); Methyl 2-(phenylsulfinyl)propanoate (23b): Yield 47% (colorless oil): IR (film) 3058, 2982, 2939, 2359, 1731, 1318, 1253, 1206, 1167, 1050, 858, 693 cm^{-1} ; ^1H NMR (CDCl_3) δ 7.59 (m, 5H), 3.84 (q, $J = 7.1$ Hz, 0.5H, diastereomer), 3.51 (q, $J = 7.1$ Hz, 0.5H, diastereomer), 3.67 (s, 1.5H, diastereomer), 3.66 (s, 1.5H, diastereomer), 1.47 (d, $J = 7.1$ Hz, 1.5H, diastereomer), 1.32 (d, $J = 7.1$ Hz, 1.5H, diastereomer).

Incubation of 2-methyl-3-phenylpropanoic acid (**15**)

Incubation of **15** with the bacterium according to the general procedure for 48 h and the product was obtained as a colorless crystal. The yield and the spectral data are as follows; Benzoic acid (**16**): Yield 78% (colorless crystal): IR (film) 2846, 2674, 2563, 1686, 1454, 1425, 1327, 1292, 1186, 934, 809, 708, 667 cm^{-1} ; ^1H NMR (CDCl_3) δ 8.13 (dt, $J = 1.5, 8.5$ Hz, 2H), 7.63 (dt, $J = 1.5, 8.5$ Hz, 1H), 7.48 (dt, $J = 1.5, 8.5$ Hz, 2H). The product was analyzed by HPLC with a Shenshu Pak PEGASIL Silica 60-5 (98/2/0.05 hexane/2-propanol/TFA; 1.0 mL/min; 254 nm): $t_R = 5.84$ min.