SUPPORTING INFORMATION:

α,α'-Dihydroxyketone formation using aromatic and heteroaromatic aldehydes with evolved transketolase enzymes

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General methods

Unless otherwise noted, solvents and reagents were reagent grade from commercial suppliers (Sigma-Aldrich) and used without further purification. Dry CH₂Cl₂ was obtained using anhydrous alumina columns.¹ All moisture-sensitive reactions were performed under a nitrogen or argon atmosphere using oven-dried glassware. Reactions were monitored by TLC on Kieselgel 60 F₂₅₄ plates with detection by UV, potassium permanganate and phosphomolybdic acid (PMA) [PMA hydrate (12 g) and ethanol (250 mL)] stains. Flash column chromatography was carried out using silica gel (particle size 40-63 μm).¹ H NMR and ¹³C NMR spectra were recorded at the field indicated using Bruker AMX300 MHz, AMX400 Avance-500 MHz and Avance-600 MHz machines. Coupling constants are measured in Hertz (Hz) and unless otherwise specified, NMR spectra were recorded at 298 K. Mass spectra were recorded on a Thermo Finnegan MAT 900XP and Micro Mass Quattro LC electrospray mass spectrometers VG ZAB 2SE. Infrared spectra were recorded on a Shimadzu FTIR-8700 and Perkin Elmer Spectrum 100 FTIR spectrometer. Optical rotations were recorded on a Perkin Elmer model 343 polarimeter at 589 nm, quoted in deg cm² g⁻¹ and conc (c) in g/100 mL. Chiral HPLC analysis was performed on a Varian Prostar instrument equipped with Chiracel OD or Chiralpak AD chiral columns (Daicel; Chiral Technologies Europe, France) 25 cm × 0.46 cm.

Lithium hydroxypyruvate was synthesised as previously described.² 1,3-dihydroxy-1-phenylpropan-2-one 3a was prepared as previously described.³

Synthesis of racemic α,α'-dihydroxyketones. The corresponding aldehyde 2b–2f (1.00 mmol) was added to a solution of Li-I (110 mg, 1.00 mmol) and N-methylmorpholine (110 μL, 1.00 mmol) in water (20 mL) at pH 8 (adjusted with 10% HCl). The reaction was stirred for 24–48 h at rt and monitored by TLC analysis. Upon concentration in vacuo, the crude material was dry loaded and purified using flash silica chromatography.

Chiral HPLC analysis of 3a, 3d-f to determine ees. Compounds 3d and 3e were monobenzoylated (dibenzoylated compounds were not separable by chiral HPLC columns used) and the products analysed by chiral HPLC to determine ees.⁴ Ketodiols 3a and 3f were dibenzoylated for chiral HPLC analysis.⁴ HPLC analysis for 3a, 3d and 3e was carried out using a Chiralcel OD column, and for 3f on a Chiralpak AD column, and the hexane:2-propanol solvent system given.

1,3-Dihydroxy-1-phenylpropan-2-one ³ (3a). Racemic 3a was dibenzoylated and HPLC analysis of the product (82:18, 1.0 mL min⁻¹) gave retention times of 10.5 min (R-isomer) and 13.4 min (S-isomer).
1-Furan-2-yl-1,3-dihydroxopropan-2-one (3b). The reaction was carried out for 48 h and the product purified using flash silica chromatography (EtOAc:hexane, 1:1) to give 3b as a colorless oil (7 mg, 5%); ν\text{max}(neat)/cm^{-1} 3391, 1708; \textsuperscript{1}H NMR (300 MHz; CDCl\textsubscript{3}) \ δ 3.39 (2H, s, OH), 4.28 (1H, d, J 19.5, CH\textsubscript{HOH}), 4.43 (1H, d, J 19.5, CH\textsubscript{HOH}), 5.30 (1H, s, HOCH), 6.35–6.47 (2H, m), 7.41 (1H, m); \textsuperscript{13}C NMR (125 MHz; CDCl\textsubscript{3}) \ δ 66.1 (C\textsubscript{H2}), 71.0 (C\textsubscript{HOH}), 110.5, 111.0, 143.6, 149.6 (CCHOH), 206.9, (C=O); m/z (HRCI) found MH\textsuperscript{+} 157.04964. C\textsubscript{7}H\textsubscript{9}O\textsubscript{4} requires 157.05008.

1,3-Dihydroxy-1-(thiophen-2-yl)propan-2-one (3c). The reaction was carried out for 48 h and the product purified using flash silica chromatography (EtOAc:hexane, 1:1) to give 3c as a brown oil (8 mg, 5%); ν\text{max}(neat)/cm^{-1} 3391, 1708, 694; \textsuperscript{1}H NMR (500 MHz; CDCl\textsubscript{3}) \ δ 3.86 (1H, s, OH), 4.36 (1H, d, J 19.5, CH\textsubscript{HOH}), 4.42 (1H, d, J 19.5, CH\textsubscript{HOH}), 5.52 (1H, s, HOCH), 7.01–7.10 (2H, m), 7.35 (1H, m); \textsuperscript{13}C NMR (125 MHz; CDCl\textsubscript{3}) \ δ 65.2 (C\textsubscript{H2}), 73.1 (C\textsubscript{HO}), 126.6, 127.1, 127.5, 140.1, 207.8, (C=O); m/z (FTMS) found M\textsuperscript{+} 173.02695. C\textsubscript{7}H\textsubscript{9}O\textsubscript{3}S requires 173.02724.

1,3-Dihydroxy-4-phenylbutan-2-one (3d). The reaction was carried out for 48 h and the product purified using flash silica chromatography (EtOAc:hexane, 1:1) to give 3d as a white solid (27 mg, 15%); M.p. 122–130 ºC (EtOAc:hexane); ν\text{max}(neat)/cm^{-1} 3262, 2966–2883, 1720; \textsuperscript{1}H NMR (300 MHz; CDCl\textsubscript{3}) \ δ 2.85 (1H, dd, J 14.0 and 7.9, CH\textsubscript{HPh}) 3.08 (1H, dd, J 14.0 and 4.6, C\textsubscript{H}HPh), 4.28 (1H, d, J 18.0, CH\textsubscript{HOH}), 4.38 (1H, d, J 18.0, CH\textsubscript{HOH}), 4.47 (1H, m, C\textsubscript{H}OH), 7.18–7.34 (5H, m, Ar); \textsuperscript{13}C NMR (125 MHz; CDCl\textsubscript{3}) \ δ 40.4 (C\textsubscript{H2Ph}), 66.3 (C\textsubscript{H2}OH), 76.1 (C\textsubscript{HOH}), 127.4, 128.7, 129.3, 135.6, 211.2 (C=O); m/z (FTMS) found [2M+Na\textsuperscript{+}] 383.1454. C\textsubscript{20}H\textsubscript{24}O\textsubscript{6}Na requires 383.1471. Racemic 3d was monobenzoylated and HPLC analysis of the product (97:3, 0.8 mL min\textsuperscript{-1}) gave retention times of 45.2 min (3\textsuperscript{R}-isomer) and 57.0 min (3\textsuperscript{S}-isomer).

1,3-Dihydroxy-4-phenyl-pentan-2-one (3e). The reaction was carried out for 48 h and the product purified using flash silica chromatography (EtOAc:hexane, 1:1) to give 3e as a white solid (25 mg, 13%); M.p. 120–130 ºC (EtOAc:hexane); ν\text{max}(neat)/cm^{-1} 3407, 2925–2855, 1720; \textsuperscript{1}H NMR (300 MHz; CDCl\textsubscript{3}) \ δ 1.27 (3H, d, J 7.1, C\textsubscript{H3}), 3.20 (1H, m, C\textsubscript{H}CH\textsubscript{3}), 4.19 (1H, d, J 19.8, CH\textsubscript{HOH}), 4.27 (1H, d, J 19.8, CH\textsubscript{HOH}), 4.33 (1H, d, J 4.5, C\textsubscript{H}OH), 7.21–7.37 (5H, m, Ar); \textsuperscript{13}C NMR (125 MHz; CDCl\textsubscript{3}) \ δ 14.3 (C\textsubscript{H3}), 43.6 (C\textsubscript{HCH\textsubscript{3}}), 127.4, 127.7, 129.0, 142.0, 211.5 (C=O); Isomer 1 \textsuperscript{1}H NMR (300 MHz; CDCl\textsubscript{3}) \ δ 1.27 (3H, d, J 7.1, CH\textsubscript{3}), 3.20 (1H, m, CH\textsubscript{CH\textsubscript{3}}), 4.19 (1H, d, J 19.8, CH\textsubscript{HOH}), 4.27 (1H, d, J 19.8, CH\textsubscript{HOH}), 4.33–4.40 (2H, m, CH\textsubscript{HOH} and CH\textsubscript{OH}), 7.21–7.37 (5H, m, Ar); m/z (FTMS) found MNa\textsuperscript{+} 217.0836. C\textsubscript{11}H\textsubscript{14}O\textsubscript{3}Na requires 217.0835. The 3e isomers were monobenzoylated and HPLC analysis of the product (97:3, 0.8 mL min\textsuperscript{-1}) gave retention times of approximately 20.0 min, 21.4 min, 24.3 min, and 26.2 min. Aldehyde 2\textsuperscript{R}–2e was synthesised as previously described.\textsuperscript{6} When used in the biomimetic reaction above, followed by monobenzoylation, HPLC analysis revealed that
the peaks at retention times of 20.0 min and 26.2 min (peaks 1 and 4) were enhanced significantly indicating these corresponded to 3e-(3RS,4R).

1,3-Dihydroxy-1-(3-hydroxyphenyl)propan-2-one (3f). The reaction was carried out for 48 h and the product purified using flash silica chromatography (EtOAc:hexane, 1:1) to give 3f as a colorless oil (10 mg, 4%). $\nu_{\text{max}}$ (neat)/cm$^{-1}$ 3407, 2925–2855, 1720; $^1$H NMR (600 MHz; CD$_3$OD) $\delta$ 4.32 (1H, d, $J$ 19.0, CH$_2$OH), 4.37 (1H, d, $J$ 19.0, CH$_2$OH), 5.18 (1H, s, CH$_2$OH), 6.73 (1H, dd, $J$ 8.1 and 1.8, 4'-H), 6.84 (1H, br s, 2'-H), 6.87 (1H, d, $J$ 7.7, 6'-H), 7.18 (1H, app t, $J$ 7.9, 5'-H); $^{13}$C NMR (125 MHz; CD$_3$OD) $\delta$ 65.0 (CH$_2$OH), 77.1 (CHOH), 114.5, 119.9, 123.0, 131.0, 137.7, 156.5, 211.0 (C=O); $m/z$ (FTMS) found MH$^+$ 183.06632. C$_9$H$_{11}$O$_4$ requires 183.06730. Racemic 3f was dibenzoylated and HPLC analysis of the product (80:20, 1.0 mL min$^{-1}$) gave retention times of 12.9 min (3S-isomer) and 17.1 min (3R-isomer).

**Mutant Change F434A.** Site-directed mutagenesis for the F434A TK mutant was performed using the Quikchange kit (Stratagene, Netherlands), forward (5'-CGTACACACCTCCACCACGGCTGTTCGTCG-3') and reverse (5'-CCACGAAACATCGGCGGAGGTGTCGTTG-3') primers, and plasmid template pQR706 which harbours the *E. coli* tkta gene complete with its natural promoter. The resulting pQR780 plasmid was transformed into supercompetent XL1-Blue (Stratagene) *E. coli* cells, and subsequently re-transformed into JM107 *E. coli* cells for activity measurements.

**TK conversions.** The TK cell free lysates were prepared using an identical procedure to that previously described.$^4$ The biotransformations were conducted at 50 mM reaction concentrations. ThDP (22 mg, 48 $\mu$mol) and MgCl$_2$.6H$_2$O (39 mg, 180 $\mu$mol) were dissolved in H$_2$O (10 mL) and the pH adjusted to 7 using 0.1 M NaOH. To this stirred solution, at 25 °C, was added TK clarified lysate (2 mL, containing approximately 0.3 mg mL$^{-1}$ of TK) and the mixture stirred for 20 min. In another flask, Li-1 (110 mg, 1.00 mmol) and the aldehyde (1.00 mmol) were dissolved in H$_2$O (8 mL) and the pH adjusted to 7 with 0.1 M NaOH. Following the 20 min enzyme/cofactor pre-incubation, the Li-1/aldehyde mixture (2a-2f) was added to the enzyme solution and the mixture stirred at rt for 24 h. During this time, the pH was maintained at 7.0 by addition of 1 M HCl using a pH stat (Stat Titrino, Metrohm) and the reactions were followed by TLC analysis. Silica was added and the reaction mixture concentrated to dryness, dry loaded onto a flash silica gel column, and purified. The ees were determined from the mono/dibenzoylated procedure given above.

**TK formation of 1,3-dihydroxy-1-phenylpropan-2-one (3a).** WT-TK gave no 3a; D469E-TK gave 3a (3 mg, 2%) as a racemate and 5% of 4; D469T-TK gave 3a (3 mg, 2%) in 70% ee (3R-isomer) and 5% of 4; D469K-TK gave 3a (3 mg, 2%) in 82% ee (3R-isomer) and 5% of 4, and F434A-TK generated 3a (16 mg, 10%) in 82% ee (3R-isomer) as a colorless oil; [$\alpha$]$^2$D = +74.0 ($c$
Data for **1,3,4,5-Tetrahydroxy-3-phenylpentan-2-one (4, R = Ph)**. The biotransformation side product (4) was generated using the D469-TK mutants as a mixture of isomers (11 mg, 5%) as an oil. \( \nu_{\text{max}}(\text{neat})/\text{cm}^{-1} \) 3391, 1684; \(^1\text{H NMR}\) (500 MHz; CD\(_2\)OD) \( \delta \) 3.05 (0.6H, d, \( J \) 11.1, CH\(_{\text{H}}\)OH), 3.73–3.78 (1.4H, m, CH\(_{\text{H}}\)OHs and CH\(_{\text{H}}\)OHs), 3.96 (0.4H, d, \( J \) 11.1, CH\(_{\text{H}}\)OH), 4.34 (0.4H, d, \( J \) 11.1, CH\(_{\text{H}}\)OH), 4.60 (0.6H, d, \( J \) 19.4, CH\(_{\text{H}}\)OH), 4.65 (0.6H, d, \( J \) 19.4, CH\(_{\text{H}}\)OH), 4.81 (1H, m, CH\(_{\text{H}}\)OH), 7.24–7.32 (3H, m, Ph), 7.40 (2H, dd, \( J \) 8.0 and 1.2, Ph); \(^{13}\text{C NMR}\) (150 MHz; CD\(_3\)OD) \( \delta \) 67.0 and 67.4 (CH\(_2\)s), 68.8 and 69.1 (CH\(_2\)s), 76.6 and 76.7, 128.8–129.0 (signals superimposed), 141.2 and 141.3, 214.6 and 215.4 (C=O); \( m/z \) (HRCl) found MH\(^+\) 227.09110. C\(_{11}\)H\(_{15}\)O\(_5\) requires 227.09195.

The absolute stereochemistry of 3a generated using F434A-TK was determined using the Mosher’s derivatisation method (both Mosher’s esters formed). \(^{8}\)

**(2S,3'R)-3,3,3-Trifluoro-2-methoxy-2-phenyl propionic acid 3'-hydroxy-2'-oxo-3'-phenylpropyl ester.** The reaction was carried out under anhydrous conditions. To a stirred solution of F434A-TK generated 3a (0.008 g, 0.05 mmol) in CH\(_2\)Cl\(_2\) (1 mL) was added triethylamine (34 \( \mu \)L, 0.25 mmol) and (R)-MTPA chloride (10 \( \mu \)L, 0.04 mmol) the reaction was stirred for 12 h at rt. The product was dry loaded onto silica gel and purified using flash chromatography (EtOAc:hexane, 1:4) to afford the titled compound as a colourless oil (0.016 g, 84%). \( \alpha \)\(_{25}^\text{D} \) = +30.0 (c 0.5, CHCl\(_3\)); \( \nu_{\text{max}}(\text{neat})/\text{cm}^{-1} \) 3420, 2930, 2855, 1734; \(^1\text{H NMR}\) (300 MHz; CDCl\(_3\)) \( \delta \) 3.61 (3H, s, OC\(_{\text{H}}\)\(_3\)), 4.69 (0.93H, d, \( J \) 17.0, CHHO (2\( R \),3'\( R \))), 4.84 (0.07H, d, \( J \) 17.0, CHHO (2\( S \),3'\( R \))), 5.02 (0.93H, d, \( J \) 17.0, CHHO (2\( R \),3'\( R \))), 5.25 (1H, s, CH\(_{\text{H}}\)OH), 7.25–7.60 (8H, m, Ar), 7.59 (2H, m, Ar); \(^{13}\text{C NMR}\) (150 MHz; CDCl\(_3\)) \( \delta \) 55.9, 55.9 (O\(_{\text{C}}\)H\(_3\)), 66.5, 78.1 (CH\(_{\text{H}}\)OH), 127.4, 127.5, 128.6, 129.5, 129.6, 130.0, 131.7, 136.8, 166.2 (C=O ester), 201.6 (C=O, ketone); \(^{19}\text{F NMR}\) (282 MHz; CDCl\(_3\)) \( \delta \) -72.2; \( m/z \) (HRCl) found MH\(^+\) 383.11063.

****(2R,3'R)-3,3,3-Trifluoro-2-methoxy-2-phenyl propionic acid 3'-hydroxy-2'-oxo-3'-phenylpropyl ester.** The reaction was carried out under anhydrous conditions. To a stirred solution of F434A-TK generated 3a (0.008 g, 0.05 mmol) in CH\(_2\)Cl\(_2\) (1 mL) was added triethylamine (34 \( \mu \)L, 0.25 mmol) and (R)-MTPA chloride (10 \( \mu \)L, 0.04 mmol) the reaction was stirred for 12 h at rt. The product was dry loaded onto silica gel and purified using flash chromatography (EtOAc:hexane, 1:4) to afford the titled compound as a colourless oil (0.016 g, 84%). \( \alpha \)\(_{25}^\text{D} \) = -12.0 (c 0.25, CHCl\(_3\)); \( \nu_{\text{max}}(\text{neat})/\text{cm}^{-1} \) 3420, 2930, 2855, 1734; \(^1\text{H NMR}\) (300 MHz; CDCl\(_3\)) \( \delta \) 3.61 (3H, s, OC\(_{\text{H}}\)\(_3\)), 4.70 (0.17H, d, \( J \) 17.0, CHHO (2\( R \),3'\( R \))), 4.84 (0.83H, d, \( J \) 17.0, CHHO (2\( S \),3'\( R \))), 4.92 (0.83H, d, \( J \) 17.0, CHHO (2\( S \),3'\( R \))), 5.02 (0.17H, d, \( J \) 17.0, CHHO (2\( R \),3'\( R \))), 5.26 (1H, s, CH\(_{\text{H}}\)OH), 7.32–7.43 (8H, m, Ar), 7.58 (2H, m, Ar); \(^{13}\text{C NMR}\) (150 MHz; CDCl\(_3\)) \( \delta \) 55.9, 55.9 (O\(_{\text{C}}\)H\(_3\)), 66.5, 78.1 (CH\(_{\text{H}}\)OH), 127.4, 127.5, 128.6, 129.5, 129.6, 130.0, 131.7, 136.8, 166.2 (C=O ester), 201.6 (C=O, ketone); \(^{19}\text{F NMR}\) (282 MHz; CDCl\(_3\)) \( \delta \) -72.2; \( m/z \) (HRCl) found MH\(^+\) 383.11063.
(OCH₃), 66.5, 78.1 (CHOH), 127.4, 127.5, 128.6, 129.5, 129.6, 130.0, 131.8, 136.7, 166.3 (C=O ester), 201.7 (C=O, ketone); ¹⁹F NMR (282 MHz; CDCl₃) δ –72.2; m/z (HRCl) found MH⁺ 383.11154. C₁₉H₁₈F₃O₅ requires 383.11063.

TK formation of 1-furan-2-yl-1,3-dihydroxypropan-2-one (3b). WT-TK gave no 3b; D469E-TK gave 3b (8 mg, 5%) and 5 (R = furyl) in <2%; D469T-TK gave 3b (5 mg, 3%) and 5 (R = furyl) in <2%; D469K-TK gave 3b (5 mg, 3%) and 5 (R = furyl) in <2%, and F434A-TK generated 3b (2 mg, 1%) as a colorless oil and 5 (R = furyl) in <2%. [α]₂₀° of 3b = +54.0 (c 0.4, CHCl₃).

Data for 1-Furan-2-yl-2,3-dihydroxy-propan-1-one (5, R = furyl). The biotransformation side product (5) was generated using the D469 and F434- TK mutants (3 mg, 2%) as an oil. νmax(neat)/cm⁻¹ 3391, 1708; ¹H NMR (300 MHz; CDCl₃) δ 3.93 (1H, dd, J 11.8 and 4.5, CHHOH), 4.06 (1H, dd, J 11.8 and 3.3, CHHOH), 4.91 (1H, m, CHHOH), 6.60 (1H, m, Ar), 7.37 (1H, m, Ar), 7.64 (1H, m, Ar); ¹³C NMR (150 MHz; CD₃OD) δ 64.9 (CH₂), 74.8 (CHO), 112.8, 119.5, 147.4, 150.3, 187.7 (C=O); m/z (HRCl) found M⁺ 157.04964. C₇H₉O₄ requires 157.05008.

TK formation of 1,3-dihydroxy-1-(thiophen-2-yl)propan-2-one (3c). WT-TK gave no 3c; D469E-TK gave 3c (4 mg, 2%) and 5 (R = thieryl) in <2%; D469T-TK gave 3c (5 mg, 3%) and 5 (R = thieryl) in <2%; D469K-TK gave 3c (4 mg, 2%) and 5 (R = thieryl) in <2%, and F434A-TK gave 3c (2 mg, 1%) as an oil and 5 (R = furyl) in <2%. [α]₂₀° of 3c = +26.0 (c 0.15, CHCl₃).

Data for 2,3-Dihydroxy-1-thiophen-2-yl-propan-1-one (5, R = thieryl). The biotransformation side product (5) was generated using the D469 and F434- TK mutants (3 mg, 2%) as an oil. νmax(neat)/cm⁻¹ 3391, 1708; ¹H NMR (300 MHz; CDCl₃) δ 3.86 (1H, dd, J 11.7 and 5.1, CHHOH), 4.05 (1H, dd, J 11.7 and 3.3, CHHOH), 4.97 (1H, m, CHHOH), 7.20 (1H, app t, J 4.5, Ar), 7.76 (1H, d, J 5.0, Ar), 7.82 (1H, d, J 4.0, Ar); ¹³C NMR (150 MHz; CD₃OD) δ 66.2 (CH₂), 75.3 (CHO), 128.6, 133.5, 135.5, 139.7, 191.8 (C=O); m/z (HRCl) found M⁺ 173.02695. C₇H₉O₃S requires 173.02724.

TK formation of 1,3-dihydroxy-4-phenylbutane-2-one (3d). WT-TK gave 3d (9 mg, 5%) in 93% ee (3S-isomer); D469E-TK gave 3d (77 mg, 43%) in 90% ee (3S-isomer); D469T-TK gave 3d (90 mg, 50%) in 96% ee (3S-isomer); D469K-TK gave 3d (81 mg, 45%) in 95% ee (3S-isomer), and F434A-TK generated 3d (86 mg, 48%) in 97% ee (3S-isomer) as a white solid. M.p. 122-130 °C; [α]₂₀° = +30.0 (c 0.5, CHCl₃).

The absolute stereochemistry of 3d generated using D469E-TK was determined using the Mosher’s derivatisation method.¹⁸ (2S,3'S)-3,3,3-Trifluoro-2-methoxy-2-phenyl propionic acid 3'-hydroxy-2'-oxo-4'-phenylbutyl ester. The reaction was carried out under anhydrous conditions. To a stirred solution of synthesized...
D469E **3d** (0.01 g, 0.04 mmol) in CH₂Cl₂ (1 mL) was added triethylamine (34 μL, 0.25 mmol) and (R)-MTPA chloride (10 μL, 0.04 mmol) and the reaction was stirred for 12 h at rt. The product was dry loaded onto silica gel and purified using flash chromatography (EtOAc:hexane, 1:4) to afford the titled compound as a colourless oil (0.015 g, 85%). \( [\alpha]^{25}_D = -30.0 \) (c 0.3, CHCl₃); \( \nu_{\text{max}}(\text{neat})/\text{cm}^{-1} \) 3600, 2932, 1759, 1737; \(^1\)H NMR (300 MHz; CDCl₃) \( \delta \) 2.59 (1H, d, \( J \) 4.0 Hz, O₃H) 2.91 (1H, dd, \( J \) 14.1 and 8.6, PhCH₂H) 3.17 (1H, dd, \( J \) 14.1 and 4.4, PhCH₂H) 3.66 (1H, s, OC₃H₃), 4.50 (1H, m, CH₃OH) 4.95 (0.95H, d, \( J \) 17.4, CH₃HO (2S,3'S)), 5.03 (0.05H, d, \( J \) 17.4, CH₃HO (2R,3'S)), 5.09 (0.05H, d, \( J \) 17.4, CH₃HO (2R,3'S)), 7.21–7.42 (5H, m), 7.43 (3H, m), 7.63 (2H, m); \(^1^3\)C NMR (125 MHz; CDCl₃) \( \delta \) 40.2 (CH₂Ar), 55.8 (OC₃H₃), 67.8 (CH₂O), 76.7 (CHOH), 127.5, 127.6, 128.5, 129.0, 129.5, 129.9, 131.9, 135.6, 166.3 (C=O ester), 203.5 (C=O, ketone); \(^1^9\)F NMR (282 MHz; CDCl₃) \( \delta \) -72.2; \( m/z \) (-HRES) found [M-H] 395.1119. C₂₀H₁₉F₃O₅ requires 395.1106.

**TK formation of 1,3-dihydroxy-4-phenylbutane-2-one (3e).** Using racemic 2e WT-TK gave 3e (68 mg, 35%) with 88% (3S,4R) and 12% (3S,4S); D469E-TK gave 3e (59 mg, 30%) with 95% (3S,4R) and 5% (3S,4S); D469T-TK gave 3e (78 mg, 40%) with 95% (3S,4R) and 5% (3S,4S); D469K-TK gave 3e (73 mg, 38%) with 95% (3S,4R) and 5% (3S,4S), and F434A-TK gave 3e (68 mg, 35%) with 85% (3S,4R) and 15% (3S,4S) as a white solid. M.p. 120-130 °C; \( [\alpha]^{25}_D = +30.9 \) (c 0.22, CHCl₃). Major isomer by \(^1^H\) NMR is isomer 1 from the biomimetic reaction. (2R)-2e was readily accepted by D469T to give one product in 40% yield, the (3S,4R)-isomer (with a retention time of approximately 26.2 min (peak 4) using the same HPLC condition as for the biomimetic reaction.

The absolute stereochemistry of 3e at C-3 generated using D469E-TK was determined using the Mosher’s derivatisation method.\(^8\)

**\(2S,3'S,4'S\)-3'-hydroxy-2'-oxo-4'-phenylpentyl-3,3,3-trifluoro-2-methoxy-2-phenylpropanoate.** The reaction was carried out under anhydrous conditions. To a stirred solution of synthesized D469E 3e (0.012 g, 0.06 mmol) in CH₂Cl₂ (1 mL) was added triethylamine (34 μL, 0.25 mmol) and (R)-MTPA chloride (10 μL, 0.04 mmol) the reaction was stirred for 12 h at rt. The product was dry loaded onto silica gel and purified using flash chromatography (hexane:EtOAc, 4:1) to afford the titled compound as a colourless oil (0.018 g, 75%), \( [\alpha]^{25}_D = -37.2 \) (c 0.25, CHCl₃); \( \nu_{\text{max}}(\text{neat})/\text{cm}^{-1} \) 3471, 2954, 1759, 1735, 1170; \(^1\)H NMR (300 MHz; CDCl₃) \( \delta \) 1.32 (3H, d, \( J \) 7.1, CHC₃H₃), 1.59 (1H, br, O₃H), 3.27 (1H, m, CHCH₃), 3.65 (1H, s, OCH₃), 4.40 (1H, d, \( J \) 3.9, CHOH) 4.87 (1H, d, \( J \) 17.0, CHOHO (2S,3'S)), 4.96 (trace 2S,3'R)-isomer), 5.04 (1H, d, \( J \) 17.0, CHOHO (2S,3'S)), 7.21–7.42 (5H, m), 7.43 (3H, m, Ph), 7.63 (2H, m, Ph); \(^1^3\)C NMR (125 MHz; CDCl₃) \( \delta \) 13.8, 43.2, 55.8 (OCH₃), 68.5 (CH₂), 80.1 (CHOH), 127.4,
127.8, 128.5, 129.0, 129.9, 131.9, 142.0, 166.2 (C=O ester), 203.7 (C=O, ketone); $^{19}$F NMR (282 MHz; CDCl$_3$) $\delta$ –72.3; m/z (HRMS) found MH$^+$ 433.1234. C$_{21}$H$_{21}$F$_3$O$_5$ requires 433.1239.

TK formation of 1,3-dihydroxy-1-(3-hydroxyphenyl)propan-2-one (3f). WT-TK and D469E-TK gave no 3f; D469T-TK gave 3f (7 mg, 4%) as a racemate, and F434A-TK gave 3f (11 mg, 6%) in 53% ee (3R-isomer) as a colorless oil. [$\alpha$]$_{25}^D$ = +99.3 (c 0.08, MeOH).

The absolute stereochemistry of 3f using F434A-TK was determined using the Mosher’s derivatisation method. (2R,3'R)-3-(1-Hydroxy-2-oxo-3-[((2R)-3,3,3-trifluoro-2-methoxy-2-phenylpropanoyl)oxy]-propyl]phenyl-3,3,3-trifluoro-2-methoxy-2-phenylpropanoate. To a stirred solution of 1,3-dihydroxy-1-(3-hydroxyphenyl)propan-2-one (3 mg, 0.02 mmol) in CH$_2$Cl$_2$ (2 mL) was added 2,4,6-collidine (10 $\mu$L, 0.08 mmol) and (S)-MTPA chloride (10 $\mu$L, 0.04 mmol). The reaction was stirred for 36 h at rt. The product was dry loaded onto silica gel and purified using flash silica chromatography (EtOAc:hexane, 3:7) to afford the titled compound as a colourless oil (1 mg, 8%). $R_f$ 0.14 (EtOAc:hexane, 3:7). $\nu_{\max}$(neat)/cm$^{-1}$ 3420, 2930, 2855, 1734; 1H NMR (600 MHz; CDCl$_3$) $\delta$ 3.62 (6H, s, OCH$_3$), 4.74 (0.4H, d, $J$ 16.9, CHHO (2R,3'R,2''R)), 4.90 (0.2H, m, CHHO (2R,3'S,2''R)), 5.02 (0.4H, d, $J$ 16.9, CHHO (2R,3'R,2''R)), 5.20 (1H, d, $J$ 3.8, CHOH), 7.25–7.60 (14H, m, Ar); $^{13}$C NMR (150 MHz; CDCl$_3$) $\delta$ 55.9 (OCH$_3$), 66.5 (CH$_2$), 77.1 (CHOH), 127.3, 127.5, 128.6–129.0 (signals superimposed(, 130.0, 130.9, 138.5, 156.4, 166.6 (C=O esters), 201.8 (C=O, ketone); $^{19}$F NMR (282 MHz; CDCl$_3$) $\delta$ –72.2; m/z (ES$^+$) found MH$^+$ 615.09. C$_{29}$H$_{25}$F$_6$O$_8$ requires 615.13.

References


