Supplementary Material (ESI) for Chemical Communications

Electronic Supplementary Information

Accessing Tetrahydrofuran Based Natural Products by Microbial Baeyer-Villiger Biooxidation

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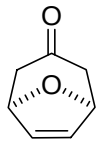
Supporting Information

3	**8-Oxabicyclo-[3.2.1]oct-6-en-3-one**



Cu/Zn couple (20.6g; 0.31mol), furan (100ml, 1.39mol) and catalytic amounts of dibromoethane were suspended in dry MeCN (80mL). The reaction mixture was cooled to 10°C under N_2 atmosphere and was sonificated for 30min under subsequent addition of tetrabromoacetone (38.2g; 1.02mol) dissolved in dry MeCN.

The temperature has to be maintained below 25°C. The conversion of the reaction was monitored by GC-MS. After complete conversion the reaction mixture was filtered through a pad of Celite®. The crude solution of 1,5-dibrom-8-oxabicyclo-[3.2.1]oct-6-en-3-one was used without further work up or purification for the next reaction step.

Cu/Zn couple (47.3g; 0.72mol) and NH_4Cl (26.0g; 0.49mol) were suspended in dry EtOH and cooled to -78°C. Maintaining a reaction temperature below -50°C, 80% of crude 1,5-dibrom-8-oxabicyclo-[3.2.1]oct-6-en-3-on in MeCN was added slowly. After 15min the remaining 20% of the solution were added and the reaction mixture was warmed to r.t. Reaction monitoring was performed by GC-MS. After conversion had reached completion, Cu/Zn couple was removed by filtration and the solid residue was washed with dichloromethane. The combined organic phases were reduced under *vacuo*. The crude product was cooled with an ice bath upon neutralization with saturated bicarbonate. The maintaining suspension was filtered again and washed intensively with dichloromethane. The combined organic layers were separated from the aqueous layer, dried over Na_2SO_4 and concentrated (bath temp. below 30°C!).

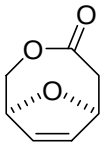
The purity of the crude product was checked by NMR and GC/MS. After evaporating of all volatiles and drying *in vaccuo* 9.11g (0.74mmol, 72%) of 8-oxabicyclo-[3.2.1]oct-6-en-3-one was obtained as beige crystals ; m.p.: 36-38°C (lit.[1] 38°C).

^{1}H NMR (200 MHz, $CDCl_3$): 2.30 (d, J= 16Hz, 2H), 2.80 (dd, J = 16Hz, J = 5Hz, 2H), 5.05 (d, J = 5Hz, 2H), 6.20 (s, 2H)

^{13}C-NMR: (50 MHz, $CDCl_3$): 46.6 (t), 77.1 (d), 133.3 (d), 205.2 (s)

[1] Hoffmann, H.M.R.; Kim, H. *Eur. J. Org. Chem.* **2000**, 2195

Supporting Information

4	**(1S,6S)-3,9-Dioxabicyclo[4.2.1]non-7-en-4-one**	

Standard procedure for Baeyer Villiger oxidation by "non–growing" cells

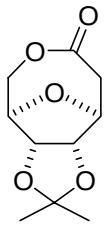
A New Brunswick Bioflow 110 fermenter containing 1L of sterile TB medium supplemented with 200mg/L ampicillin was inoculated with 20mL (2vol%) overnight culture of DH5α/CPMO grown on LB medium (50mg/mL ampicillin). The temperature was maintained at 37°C and the pH was kept constant at 7.00±0.05 by adding 3N NaOH or 3N H_3PO_4 automatically. The 1L culture was grown with an air flow of 5L min^{-1} and stirring rates at 500 rpm. The growth was continued until the culture density reached 3.01-3.44 g/L dcw. Then the temperature was decreased to 25°C and IPTG was added to a final concentration of 0.25mM. After one hour the fermentation culture was supplemented with 4g/L glucose (20% sterile solution). Two hours after induction 20mL of cell culture were taken and activity tests were performed. After passing the activity tests the pre loaded resin and any additives were added. The glucose level was measured periodically as the bioconversion progressed.

Bioconversion of ketone (3) to (1S,6S)-3,9-dioxabicyclo[4.2.1]non-7-en-4-one (4)

According to the general procedure for "non" growing cells 8-oxabicyclo[3.2.1]oct-6-en-3-one was biooxidized to the corresponding lactone **4**. Ketone **3** (5g, 40mmol) was dissolved in ethanol (10ml) and was subsequently added to Lewatit VPOC 1163 resin (50g wet resin, load X^{eq} = 0.2) and 100mL of LB_{Amp}. β-Cyclodextrin (10mol%) and the substrate-resin mixture were added to the fermentation broth. After 36 hours of fermentation the resin was recovered by filtration. Cells were separated from the fermentation broth by centrifugation. The resin was submitted to continuous solid phase extraction using dichloromethane overnight. The fermentation broth was also continuously extracted using dichloromethane overnight. The combined organic layers were dried and concentrated. Purification by column chromatography (LP/EtOAc=2/1; 200g SiO_2) gave the desired lactone **4** in 70% yield (3.9g, 95% ee) as colorless crystals; m.p.: 98-100°C; $[\alpha]_D^{20}$: +85.2 (c = 0.2, $CHCl_3$)

^{1}H NMR (200 MHz, $CDCl_3$): 2.90 (dd, J = 5Hz, J = 16Hz, 1H), 3.20 (dd, J = 3Hz, J = 16Hz, 1H), 4.05 (dd, J = 3Hz, J = 12Hz, 1H), 4.40 (d, J = 12Hz, 1H), 4.70 (d, J = 3Hz, 1H), 4.85 (d, J = 3Hz, 1H), 6.10 (d, J = 6Hz, 1H), 6.35 (d, J =6Hz, 1H).

^{13}C-NMR (50 MHz, $CDCl_3$): 46.7 (t), 71.0 (t), 76.3 (d), 81.6 (d), 129.0 (d), 133.4 (d), 172.0 (s).

5	**(1S,2S,6R,7R)-4,4-Dimethyl-3,5,9,12-tetraoxa-tricyclo[5.4.1.0^{2,6}]dodecan-10-one**	

Asymmetric dihydroxylation of 8-oxabicyclo[3.2.1]oct-6-en-3-one was performed by OsO_4 in the presence of N-methylmorpholine oxide. Lactone **4** (50mg, 0.36mmol) and NMO*H_2O (58mg, 0.43mmol, 1.2eq) were dissolved in dry dichloromethane. Subsequently catalytic amounts (5mg) of OsO_4 were added and the reaction was stirred at r.t. until complete conversion was observed (TLC control; LP/EtOAc=1/3). The crude reaction mixture was concentrated *in vacuo* (20°C!) and traces of water were removed by repeated azeotropic distillation using dichloromethane (4times, 10mL dichloromethane). The crude diol was used without further work up or purification in the following protection step. The reaction mixture was dissolved in dry acetone and cooled to 0°C under N_2-atmosphere. Freshly sublimed $AlCl_3$ (100.9mg, 0.76mmol, 2.1eq) was dissolved in dry, cooled diethylether and added slowly to the reaction mixture. After complete addition of $AlCl_3$ the reaction mixture was heated to 40°C until complete conversion of the diol was observed (TLC control, LP/EtOAc=1/1). Hydrolysis was performed with saturated and cooled bicarbonate solution and the aqueous phase was extracted 4 times with EtOAc. The combined organic layers were dried over sodium sulfate and the solvents were evaporated.

After purification *via* column chromatography (LP/EtOAc=3/1, 5g basic SiO_2) 36mg (47%) of compound **5** were obtained as colorless crystals; m.p.: 163-167°C (lit.[2] 167-168.5°C); $[\alpha]_D^{20}$: +73.0 (c = 0.66, $CHCl_3$) (lit.[2] +82.6, c = 0.6, $CHCl_3$)

^{1}H NMR (200 MHz, $CDCl_3$): 1.32 (s, 3H), 1.49 (s, 3H), 2.96 (d, 2H, 4.0 Hz), 4.23-4.30 (m, 3H), 4.31-4.39 (m, 1H), 4.65 (d, 2H, 5.5Hz), 4.95 (d, 2H, 5.5Hz);

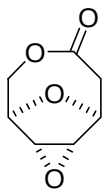
^{1}H-NMR data are according to the literature.[3]

^{13}C-NMR (50 MHz, $CDCl_3$): 24.3 (q), 25.9 (q), 42.5 (t), 71.5 (t), 78.3 (d), 81.5 (d), 82.4 (d), 83.5 (d), 112.4 (s), 172.2 (s).

[2] Cruickshank, K.A.; Reese, C.B. *Synthesis* **1983**, 3, 199.
[3] Gensler, W.J.; Chan, S.; Ball, D.B. *J. Org. Chem.* **1981**, *46*, 3407.

Supporting Information

6	**(1R,6S,7S,9R)-3,8,10-Trioxa-tricyclo[4.3.1.0^{7,9}]decan-4-one**



m-CPBA (3891 mg, 22.7 mmol) was added to a solution of lactone **4** (390 mg, 2.27 mmol) in dry CH_2Cl_2 (100 mL) at room temperature. The reaction mixture was refluxed for 2 days until no starting material could be detected by TLC.

Subsequently, the mixture was concentrated and the residue was purified by column chromatography on silica gel (hexane/EtOAc) to give epoxide **6** (438 mg, 98%) as a colorless crystals; mp: 78-80°C.

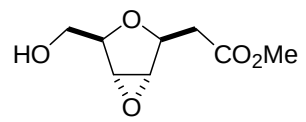
MW: 156.14 $C_7H_8O_4$ $[\alpha]_D^{22}$ = +94.9 (*c*= 0.79, $CHCl_3$)

^{1}H NMR (200 MHz, $CDCl_3$): 2.92 (dd, 2H, J=3.8 and 16.5), 3.05 (dd, 1H, J= 2.5 and 16.5), 3.76 (s, 2H), 4.19 (dd, 1H, J= 2.7 and 3.6), 4.20-4.50 (m, 2H).

^{13}C-NMR (50 MHz, $CDCl_3$): 41.9 (t), 52.6 (d), 54.2 (d), 69.3 (d), 71.3 (t), 74.2 (d), 171.5 (s).

Supporting Information

7	**(1S,2S,4R,5R)-(4-Hydroxymethyl-3,6-dioxa-bicyclo[3.1.0]hex-2-yl)-acetic acid methyl ester**



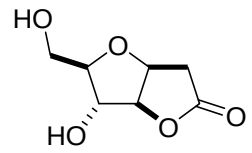
K_2CO_3 (20.0 mg, 0.143 mmol) was added to the stirred solution of compound **6** (0.047g, 0.249 mmol) in MeOH/H_2O (1 mL, 8:2) at room temperature.

The reaction mixture was stirred until the lactone had reacted completely (TLC, 10 seconds). The mixture was quenched with saturated aqueous solution of ammonium chloride and extracted with EtOAc (5 x 15mL). The organic layer was dried (Na_2SO_4) and evaporated. Purification by flash chromatography on silica gel eluting with EtOAc /hexane, gave the methylester **7** (0.056 g, 98%) as a colorless oil.

MW: 188.18 $C_8H_{12}O_5$ $[\alpha]_D^{22}$ = -32.09 (*c*= 6.19, $CHCl_3$)

^{1}H-NMR (200 MHz, $CDCl_3$): 2.43 (br s, 1H), 2.57 (d, 2H, J= 7.5), 3.67 (s, 3H), 3.50-3.75 (m, 4H), 4.11 (t, 1H, J= 4.3), 4.43 (t, 1H, J= 7.5).

^{13}C-NMR (50 MHz, $CDCl_3$): 38.2 (t), 52.1 (d), 58.6 (d), 59.9 (d), 62.7 (t), 74.8 (q), 79.7 (d), 171.2 (s).

8	**(3aS,5R,6R,6aR)-6-Hydroxy-5-hydroxymethyl-tetrahydro-furo[3,2-b]furan-2-one**	

A solution of compound **7** (0.081 g, 0.430 mmol) in dry dichloromethane (3.0 mL) was treated with tin tetrachloride in dichloromethane (1 mL, 100µL/mL) at -78°C.

The reaction mixture was stirred for 1 h until the starting material had reacted completely (TLC). Silica gel was added, the mixture was concentrated and directly submitted to purification by chromatography (hexane/EtOAc) to yielded **8** (0.073 g, 98%) as a gum.

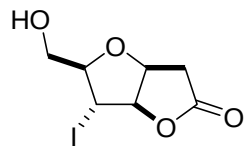
MW: 174.15 $C_7H_{10}O_5$ $[\alpha]_D^{25}$ = - 30.1 (*c*= 0.86, CH_3OH)

^{1}H NMR (200 MHz, $CDCl_3$, CD_3OD): 2.50 (d, 1H, J = 18.4Hz), 2.72 (dd, 1H, J = 4.9, 18.8Hz), 3.21 (br s, 2H), 3.48 (dd, 1H, J= 5.9, 11.9Hz), 3.68 (dd, 1H, J= 3.7, 11.9 Hz), 3.74 (app.dt, 1H, J = 3.7, 5.9), 4.06 (d, 1H, J = 5.5 Hz), 4.76-4.80 (m, 2H).

^{13}C NMR (50 MHz, CD_3OD): 35.1 (t), 61.1 (t), 75.5 (d), 77.2 (d), 86.4 (d), 90.4 (d), 176.3 (s).

^{1}H-NMR and ^{13}C NMR data are according to the literature.[4]

[4] Cagnolini, C.; Ferri, M.; Jones, P.R.; Murphy, P.J. *Tetrahedron*, **1997**, 53, 4815

9	**(3aS,5R,6R,6aR)-5-Hydroxymethyl-6-iodo-tetrahydro-furo[3,2-b]furan-2-one**	

KOH (230 mg, 4.11 mmol) was added to a stirred solution of lactone **4** (0.550 g, 3.918 mmol) in MeCN/H_2O (2.3:1; 8.6 mL).

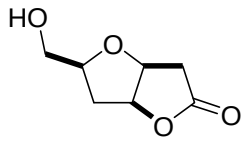
The reaction mixture was stirred at room temperature for 4 h until TLC showed disappearance of the starting material. Afterwards, a mixture of I_2 (1.092 g, 4.301 mmol) and KI (2.145 g, 12.93 mmol) was added. The resulting mixture was stirred in the dark for 5 days at 40°C. The reaction was quenched with a 10% of $Na_2S_2O_3$ solution until the mixture became colorless and then extracted with EtOAc (5 x 15mL). The combined organic layers were washed with brine, dried (Na_2SO_4) and evaporated. Purification by flash chromatography on silica gel eluting with EtOAc/hexane gave 0.835 g (75%) of **9** as a beige oil which slowly solidified upon standing in the refrigerator (m.p.: 84-86°C).

MW: 284.05 $C_7H_9IO_4$ $[\alpha]_D^{22}$ = -70.2 (*c*= 1.28, $CHCl_3$)

^{1}H-NMR (200 MHz, $CDCl_3$): 1.79 (br s, 1H), 2.72 (d, 2H, J= 3.3), 3.62 (dd, 1H, J= 4.6 and 12.4), 3.80 (dd, 1H, J= 3.0 and 12.4), 4.15 (dd, 1H, J= 2.1 and 7.5), 4.27 (ddd, 1H, J= 3.0, 4.6 and 7.5), 4.75-4.82 (m, 1H), 5.15 (dd, 1H, J= 2.1 and 4.5).

^{13}C-NMR (50 MHz, $CDCl_3$): 20.0 (d), 36.2 (t), 61.2 (t), 78.2 (d), 90.7 (d), 92.8 (d), 173.8 (s).

Supporting Information

10	**(3aS,5S,6aS)-5-Hydroxymethyl-tetrahydro-furo[3,2-b]furan-2-one**	

Tributyltin hydride (0.500 mL, 1.852 mmol) was added dropwise to a solution of iodolactone **9** (0.190 g, 0.712 mmol) in dry toluene (15 mL) at room temperature.

The mixture was stirred for 48 h at 60 °C until the iodolactone had reacted completely (TLC). The solvent was removed by evaporation and the residue was purified by column chromatography (silica gel, EtOAc/Hexane) to yielded lactone **10** (0.105 g, 99%) as a colorless oil.

MW: 158.15 $C_7H_{10}O_4$ $[\alpha]_D^{25}$ = -53,3 (*c*= 0.42, CH_3OH)

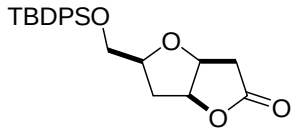
^{1}H NMR (200 MHz, $CDCl_3$): 1.89 (br s, 1H), 2.12 (ddd, 1H, J= 3.2, 7.1, 14.6 Hz), 2.39 (ddd, 1H, J= 6.9, 7.8, 14.6 Hz), 3.60 (dd, 1H, J= 6.3, 11.8 Hz), 2.75 (d, 2H, J= 3.2 Hz), 3.74 (dd, 1H, J= 3.2, 11.9 Hz), 4.18 (ddd, 1H, J= 3.2, 7.2, 10.4 Hz), 4.63 (dd, 1H, J= 3.2, 7.3 Hz), 5.05 (ddd, 1H, J= 1.7, 4.2, 6.3 Hz).

^{13}C NMR (50 MHz, $CDCl_3$): 34.0 (t), 36.4 (t), 64.6 (t), 79.0 (d), 80.7 (d), 84.4 (d), 175.2 (s).

^{1}H-NMR and ^{13}C NMR data are according to the literature.[5]

[5] a) H.B. Mereyala and R.R. Gadikota, *Tetrahedron Asymm.* 2000, **11**, 743. b) R.R. Gadikota, C.S. Callam and T.L. Lowary, *J. Org. Chem.*, 2001, **66**, 9046.

Supporting Information

11	**(3aS,5S,6aS)-5-(*tert*-Butyl-diphenyl-silanyloxymethyl)-tetrahydro-furo[3,2-b]furan-2-one**	

Imidazole was added (20.0 mg, 0.290 mmol) to a solution of alcohol **10** (8.5 mg, 0.053 mmol) in dry DMF (0.5 mL) followed by *tert*-butylchlorodiphenylsilane (30 mg, 0.108 mmol). The mixture was stirred for 24 h at room temperature until the alcohol **10** had reacted completely (TLC). Then, the reaction salutation was directly purified by chromatography (hexane/EtOAc) to yielded **11** (20.0 mg, 92%) as a colorless oil.

MW: 396.55 $C_{23}H_{28}O_4Si$ $[\alpha]_D^{26}$ = -23.8 (*c*= 0.75, $CHCl_3$)

^{1}H NMR (200 MHz, $CDCl_3$, δ): 1.05 (s, 9H), 2.10-2.26 (m, 1H), 2.26-2.45 (m, 1H), 2.68-2.72 (m, 2H), 3.63-3.79 (m, 2H), 4.05-4.20 (m, 1H), 4.54-4.61 (m, 1H), 4.97-5.05 (m, 1H), 7.30-7.45 (m, 6H), 7.61-7.72 (m, 4H).

^{13}C NMR (50 MHz, $CDCl_3$, δ): 19.2 (s), 26.8 (q), 34.6 (t), 36.4 (t), 65.6 (t), 78.9 (d), 80.7 (d), 84.3 (d), 127.7 (d), 129.7 (d), 133.4 (s), 133.4 (s), 135.5 (d), 135.6 (d), 175.2 (s).

^{1}H-NMR and ^{13}C NMR data are according to the literature.[5]