**Supplementary data**

**Enzymatic dynamic reductive kinetic resolution towards 12% w/v *(S)*-2-phenylpropanol**

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**1 Reduction of racemic 2-phenylpropanal by isolated D51A *Ct*XR**

*Figure S1*. Time course of the reduction of 0.5mM substrate with 240 U/mL isolated D51A *Ct*XR. Blue dots (*S*)-2-phenylpropanal, orange dots (*R*)-2-phenylpropanol.

**2 Biotransformation of 100mM racemic 2-phenylpropanal**

*Table S1*. Data for Figure 1.

**3 Reversed phase, chiral HPLC**

*Figure S2*. Overlay of HPLC traces for rac-2-phenylpropanal (1), acetophenone (2), 1-phenylethanol (3), (*R,S*)-2-phenylpropanol (4), reaction buffer with NAD+ (5), reaction buffer (6), bioreduction sample 1M 2-phenylpropanal reacted with 40gCDW/L and 6mM NAD+ (7).

*Table S2 (data to Figure S1)*. Retention times for main products and by-products obtained from reversed phase, chiral HPLC.

**4 Chiral GC-FID**

*Figure S3*. Selected GC traces for bioreduction replicates (N=6) of 1M 2-phenylpropanal reacted with 40gCDW/L and 6mM NAD+.

*Table S3 (data corresponding to Figure S2)*. Retention times for main products and by-products obtained from chiral GC.

**5 1H-NMR**

*Figure S4*. 1H-spectra in MeOD. Graph shows a spectrum of isolated product of a reaction with 78 % analytical yield (HPLC). Spectra were recorded immediately after work-up. Ethyl acetate related signals at 1.75, and 3.9ppm; acetophenone related signal at 2.45ppm.

**1 Reduction of racemic 2-phenylpropanal by isolated D51A *Ct*XR Time course**

*Figure S1*. Time course of the reduction of 0.5mM substrate with 240 U/mL isolated D51A *Ct*XR. Blue dots (*S*)-2-phenylpropanal, orange dots (*R*)-2-phenylpropanol.

**2 Biotransformation of 100mM racemic 2-phenylpropanal**

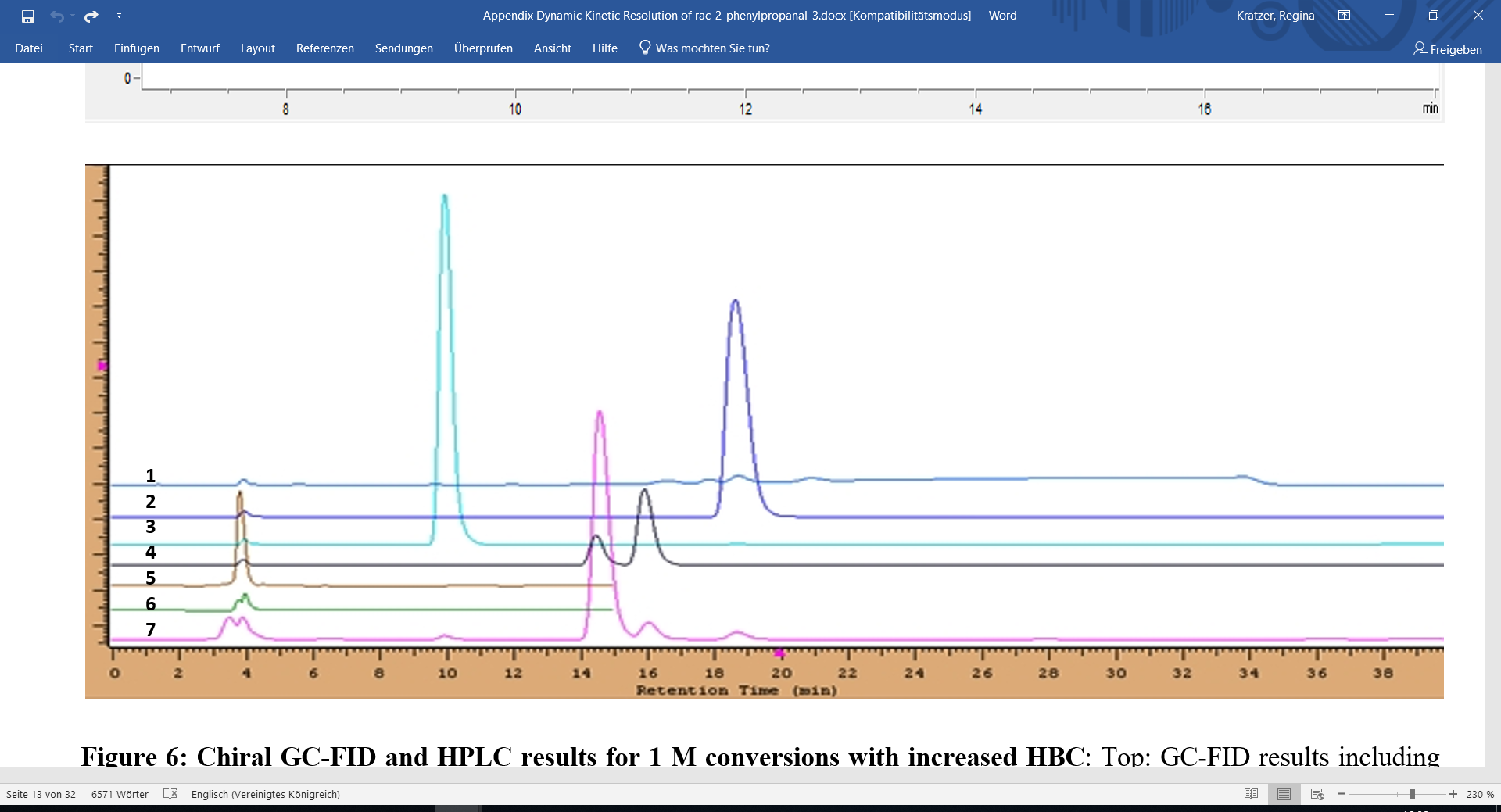
*Table S1 (data for Figure 1)*. Conversions and product e.e.-values of 100mM racemic 2-phenylpropanal reduction by lyophilized whole-cell catalyst and cell-free catalyst. Effects of catalyst form and loadinga.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Whole-cell catalyst  (gCDW/L) | Cell-free catalyst  (gCDW/L) | Conversion  (%) | e.e.  (%) | Ratio  (gsubstrate/gCDW) |
| 4 | 0 | 41.3 | 95.3 | 3.4 |
| 10 | 0 | 66.5 | 61.3 | 1.4 |
| 20 | 0 | 69.4 | 49.0 | 0.7 |
| 40 | 0 | 72.4 | 45.8 | 0.3 |
| 0 | 4 | 29.1 | 95.7 | 3.4 |
| 0 | 10 | 72.7 | 58.1 | 1.4 |
| 0 | 20 | 69.6 | 41.7 | 0.7 |
| 0 | 40 | 76.9 | 27.5 | 0.3 |

aNAD+ concentration 6mM, reaction time 24h. Data are based on HPLC analysis.

**3 Reversed phase, chiral HPLC**

Separation of main products and by-products on HPLC (stationary phase Chiralpak® AD-RH column from Daicel, mobile phase 25 % acetonitrile in ddH2O, 40°C).



*Figure S2*. Overlay of HPLC traces for rac-2-phenylpropanal (1), acetophenone (2), rac. 1-phenylethanol (3), (*R,S*)-2-phenylpropanol (4), reaction buffer with NAD+ (5), reaction buffer (6), bioreduction sample 1M 2-phenylpropanal reacted with 40gCDW/L and 6mM NAD+ (7).

*Table S2 (data corresponding to Figure S1)*. Retention times for main products and by-products on reversed phase, chiral HPLC.

|  |  |
| --- | --- |
| Analyte | Retention time (min) |
| rac-2-Phenylpropanal | broad peak, not applicable |
| (*R*)-2-Phenylpropanol | 15.8 |
| (*S*)-2-Phenylpropanol | 14.5 |
| Acetophenone | 18.6 |
| rac.- 1-Phenylethanol | 9.9 (no separation) |

**4Chiral GC-FID**

*(S)-*2-phenylpropanol

|  |
| --- |
| *(R)-*2-phenylpropanol  *rac*-2-phenylpropanal  Acetophenone |

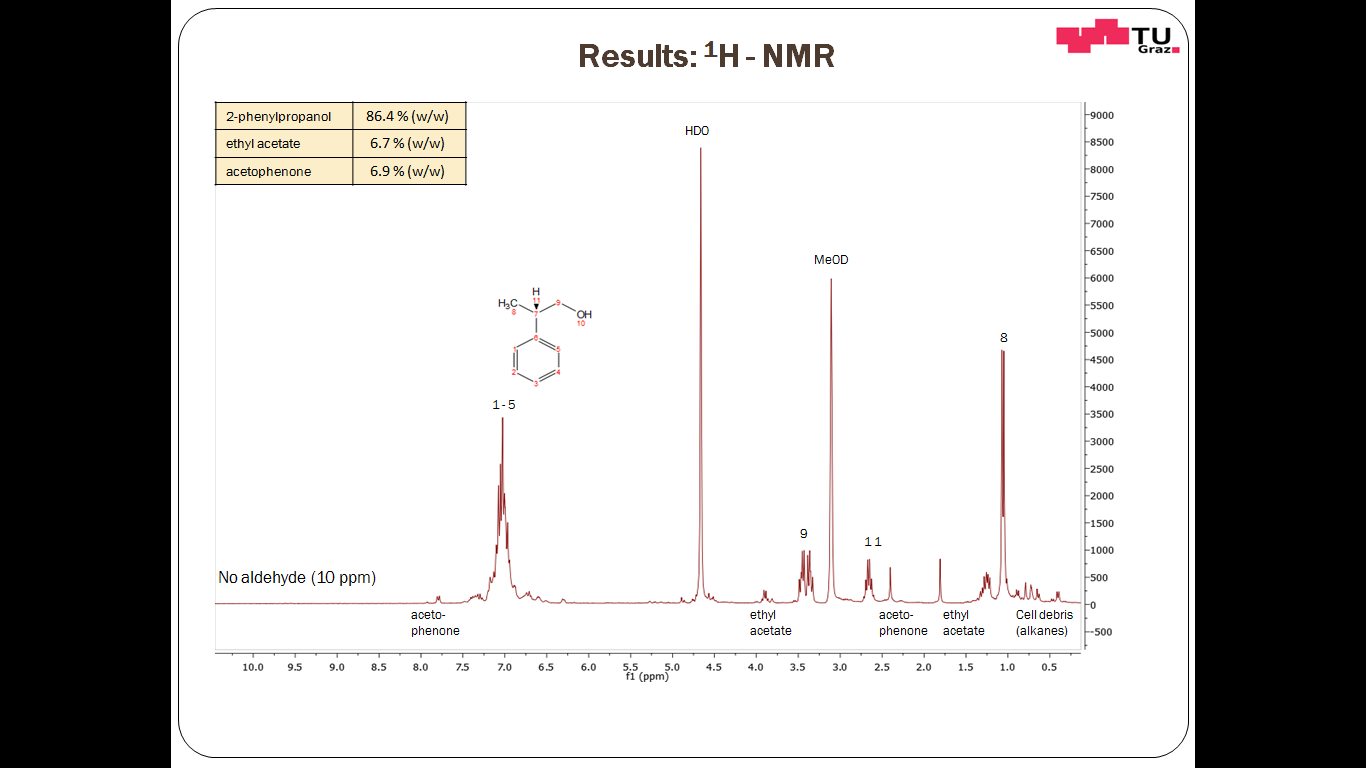
*Figure S3*. GC traces for bioreduction replicates (N=6) of 1M 2-phenylpropanal reacted with 40gCDW/L and 6mM NAD+.

*Table S3 (data corresponding to Figure S2)*. Retention times for main products and by-products on chiral GC.

|  |  |
| --- | --- |
| Analyte | Retention time (min) |
| (*R*)-2-Phenylpropanal | 8.2 |
| (*S*)-2-Phenylpropanal | 8.8 |
| (*R*)-2-Phenylpropanol | 14.8 |
| (*S*)-2-Phenylpropanol | 15.4 |
| Acetophenone | 6.9 |

**5 1H-NMR**

**Bioreduction analysis**

The isolate (extraction with ethylacetate) of a bioreduction mixture from 1M 2-phenylpropanal reacted with 40gCDW/L and 6mM NAD+ was analyzed by 1H-NMR.

*Figure S4*. 1H-spectra in MeOD. Graph shows a spectrum of isolated product of a reaction with 78 % analytical yield (HPLC). Spectra were recorded immediately after work-up. Ethyl acetate related signals at 1.75, and 3.9ppm; acetophenone related signal at 2.45ppm.