Electronic Supplementary Material (ESI) for RSC Advances

Tuning lipase B from *Candida antarctica* C-C Bond Promiscuous Activity by immobilization on Poly-Styrene–Divinylbenzene beads

Diana F. Izquierdo, Oveimar Barbosa, M. Isabel Burguete, Pedro Lozano, S.V. Luis,

Roberto Fernadez-Lafuente and Eduardo García-Verdugo

General Procedure for the Preparation of Nitroalcohols. A typical experiment procedure is as follows: Over a suspension of the corresponding aldehyde (0.57 mmol) in water (2.80 mL) were added nitromethane (5.7 mmol) and supported enzyme (200 mg). The mixture was shaken at 40 °C and 250 rpm for the corresponding time. After that time, the reaction was quenched by adding H_2O (5 mL) and the aqueous phase extracted with CH_2Cl_2 (3 × 5 mL). The organic phases were combined, dried over Na_2SO_4 and filtered, and the solvent was removed by distillation under reduced pressure. The reaction crude was analyzed by NMR to determine the conversion.

The products where isolated and purified by column flash chromatography using hexane/AcOEt as already reported by us and others[1],[2]

2-Nitro-1-(4-nitrophenyl)ethanol: *Rf* (20% EtOAc/hexane) 0.25; 1H NMR ($CDCl_3$, 300.13 MHz): δ 2.25 (brs, 1H), 4.58-4.61 (m, 2H), 5.63-5.68 (m, 1H), 7.64 (d, 3*J*HH) 6.5 Hz, 2H), 8.26 (d, 3*J*HH) 6.5 Hz, 2H);

13C NMR ($CDCl_3$, 75.5 MHz): δ 70.4 (CH2), 81.0 (CH), 125.5 (2CH), 127.4 (2CH), 145.5 (C), 148.5 (C).

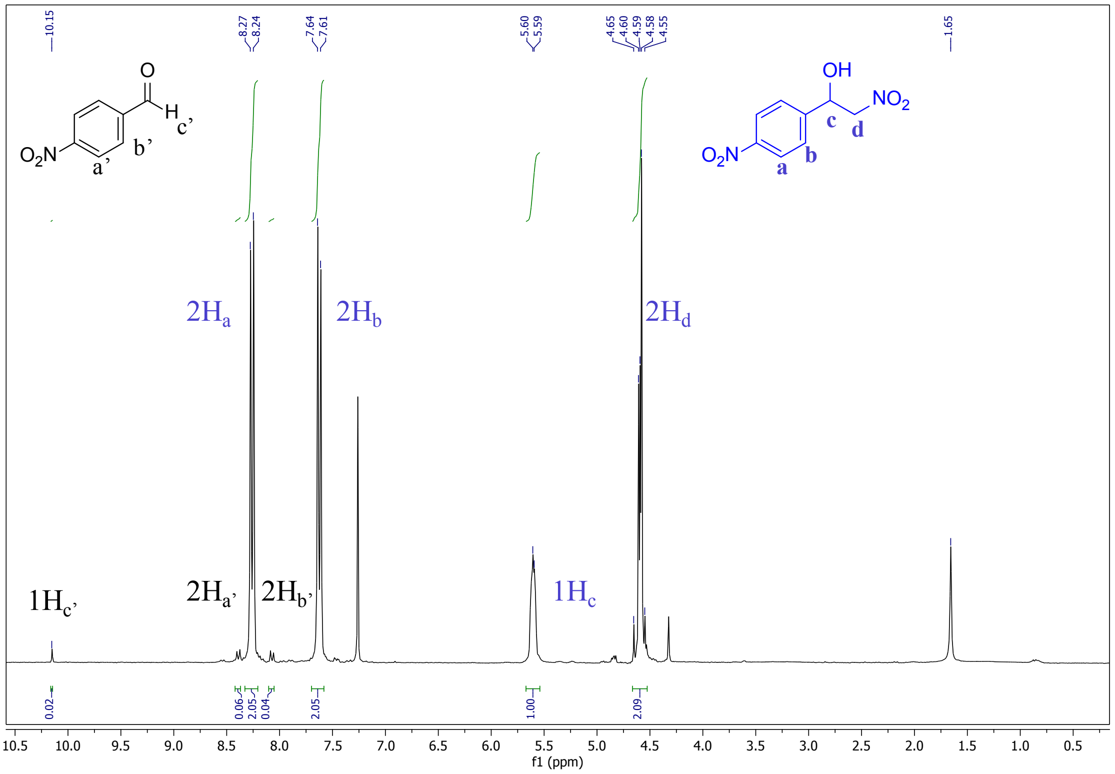

Figure 1. ^{1}H NMR of 2-nitro-1-(4-nitrophenyl)ethanol. . The conversion had been calculated using the following equation:

$$C = \left(A_c/nH_c\right) x100 \Bigg/ \left(\left(A'_c/nH'_c\right) + \left(A_c/nH_c\right)\right)$$

, where A is proton integration area and nH is number of protons of the corresponding signal.

2-Nitro-1-(4-nitrophenyl)propan-1-ol: *Rf* (20% EtOAc/hexane) 0.29; 1H NMR (CDCl3, 300.13 MHz): *δ* 1.35 (d, 3*J*HH) 7.1 Hz, 3H*syn*), 1.52 (d, 3*J*HH) 7.1 Hz, 3H*anti*), 3.20 (brs,1H*syn*+1H*anti*), 4.59-4.72 (m, 1H*syn*+1H*anti*), 5.20 (d, 3*J*HH), 9.3 Hz, 1H*syn*), 5.55 (d, 3*J*HH) 3.5 Hz, 1H*anti*), 7.58-7.63 (m,1H*syn*+1H*anti*), 8.21-8.35 (m, 1H*syn*+1H*anti*); 13C NMR (CDCl3, 75.5 MHz): *δ anti* 11.7 (CH3), 72.3 (CH), 86.6 (CH), 123.8 (2CH), 126.9 (2CH), 145.2 (C), 148.1 (C). *δ syn* 16.1 (CH3), 74.9 (CH), 87.7 (CH), 124.0 (2CH), 127.8 (2CH), 145.5 (C), 148.1 (C).

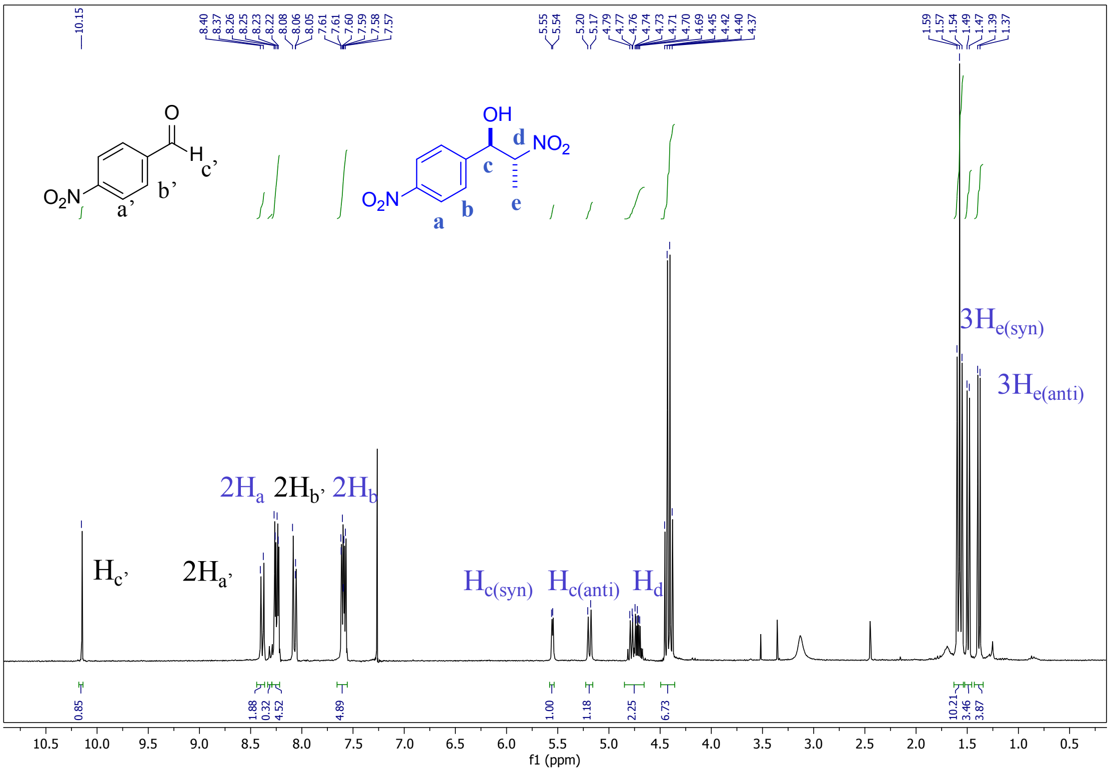

Figure 2. ^{1}H NMR of 2-nitro-1-(4-nitrophenyl)propan-1-ol. The conversion had been calculated using the following equation:

$$C = \frac{\left[\left(A_{c(syn)}/H_{c(syn)}\right) + \left(A_{c(anti)}/H_{c(anti)}\right)\right]}{\left[\left(A'_{c}/H'_{c}\right) + \left(A_{c(syn)}/H_{c(syn)}\right) + \left(A_{c(anti)}/H_{c(anti)}\right)\right]} x100$$

, where A is proton integration area and nH is number of protons of the corresponding signal..

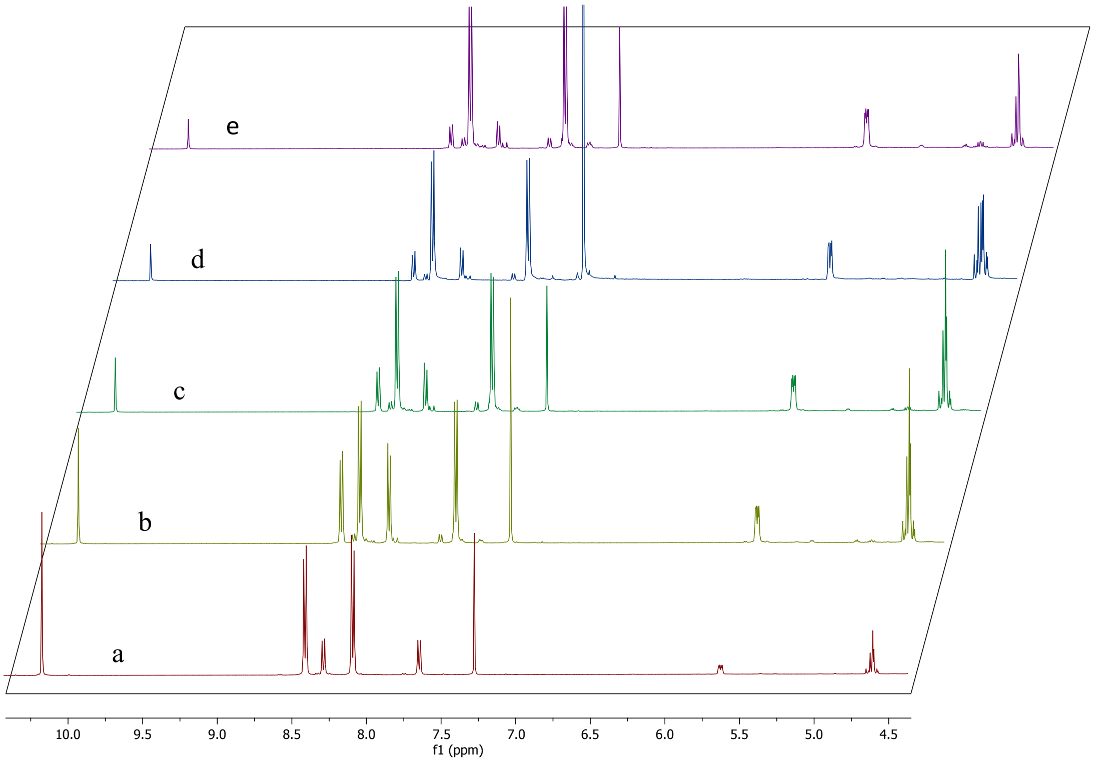

Figure 3. ^{1}H NMR spectrums of kinetic reaction carried out by PS-DVB-CALB at different times; a) 4h, b) 8h, c) 12h, d) 16h y e) 24h.

---

1 M. I. Burguete, H. Erythropel, E. Garcia-Verdugo, S. V. Luis, V. Sans, *Green Chem.* **2008**, *10*, 401.

2 E. Busto, V. Gotor-Fernández, V. Gotor, *Org. Process Res. Dev.* **2010**, *15*, 236.