CaL-B Immobilized on Single Walled Carbon Nanotubes as Efficient Biocatalyst for the Kinetic Resolution of 1-(Hetero)aryl -Ethanols

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Introduction

Numerous enantiopure heteroaryl alcohols are known for their biological activity, or represent precursors in the synthesis of a large number of pharmaceutical products.[1]

An efficient method for the synthesis of enantiopure secondary alcohols is the lipase-catalyzed kinetic resolution of racemic alcohols.

Immobilization can modify and improve properties of enzymes, may enable their recovery and reuse, can provide a favorable environment for the enzymes, may increase their rigidity and may improve their enantioselecivity.[2]

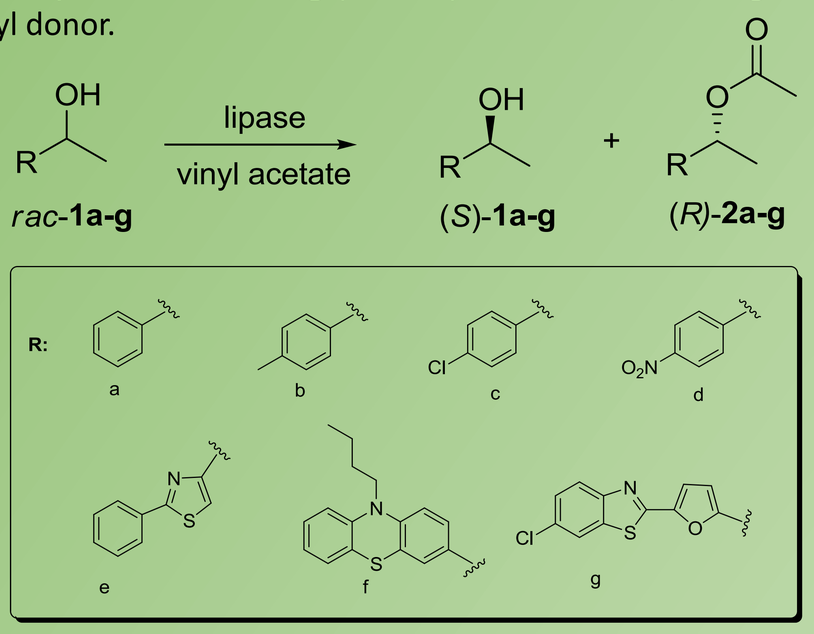
Carbon nanotubes are widely used for the immobilization of biomacromolecules, due to their mechanical, thermal, electrical properties and general biocompatibility since they can offer a support with large surface area, low diffusion limitation and easy recovery.[3]

Experimental

The commercially available $SwCNT_{COOH}$ was activated with carbonyldiimidazol, followed by coupling with 1,3-diaminopropane. The SwCNT carrier was further treated with the glycerol diglycidyl ether crosslinker (Scheme 2). Immobilization of the lipase B from *Candida antarctica* (CaL-B) was carried out in the presence of Tween 80, a non-ionic surfactant. Furthermore, the optimal loading of the enzyme on the support material was also tested. The $SwCNT_{COOH}$CaL-B biocatalyst was used in the kinetic resolution of several secondary alcohols (Scheme 1).

Discussion

The obtained enzyme preparations were characterized by reproducibility and high immobilization yields (>99 % of the enzyme bound to $SwCNT_{COOH}$). The immobilized lipase showed high enantioselectivity and activity in the enzymatic kinetic resolution of the tested racemic secondary alcohols *rac*-**1a-g** (Table 1) in acetonitrile, using vinyl acetate as acyl donor.



Scheme 1. Enzymatic kinetic resolution of racemic alcohols *rac*-**1a-g**. Reactants and solvents: vinyl acetate, $SwCNT_{COOH}$CaL-B , acetonitrile.

Conclusions

Covalent immobilization of CaL-B on carboxylated single-walled carbon nanotubes after amidation and bisepoxide treatment resulted in a highly efficient and stable biocatalyst which proved to be useful in the kinetic resolution of various secondary alcohols.

Selected references

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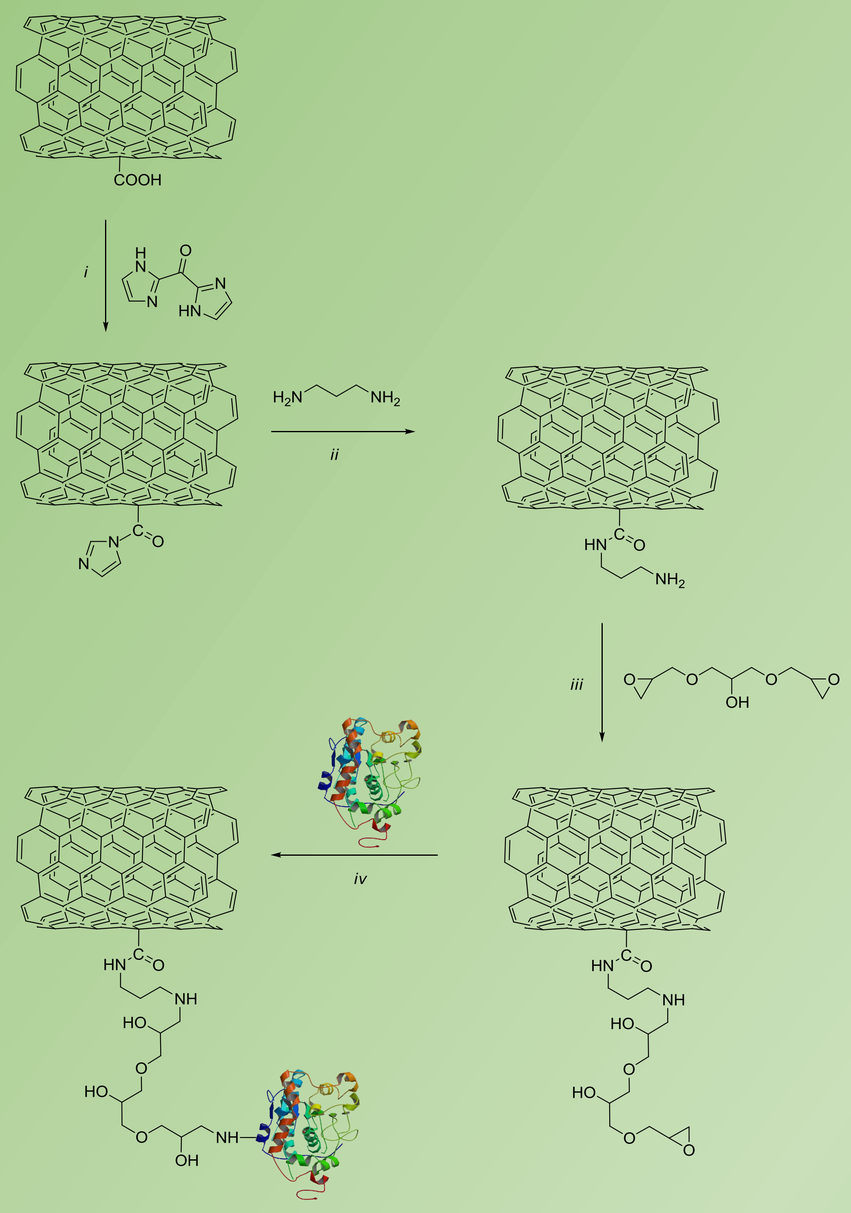

Scheme 2. Immobilization of CaL-B on $SwCNT_{COOH}$ i) CDI in CH_2Cl_2; ii) $H_2N(CH_2)_3NH_2$ in water; iii) glycerol diglycidyl ether in CH_2Cl_2; iv) CaL-B in PBS buffer (20 mM Na_2HPO_4, 150 mM NaCl, pH 7)

Table 1. Enantioselective acylation of *rac*-**1a-g** mediated by $SwCNT_{COOH}$CaL-B after 36 h (10 mg substrate, 1 mg neat enzyme, 5 µL vinyl acetate, 1 mL acetonitrile).

Substrate	ee_s (%)	ee_p (%)	*c* (%)	E
rac-**1a**	37	>99	27.2	>200
rac-**1b**	68	>99	40.6	>200
rac-**1c**	49	>99	33.1	>200
rac-**1d**	92	>99	48.0	»200
rac-**1e**	>99	>99	50	»200
rac-**1f**	>99	>99	50	»200
rac-**1g**	76	>99	43.1	»200

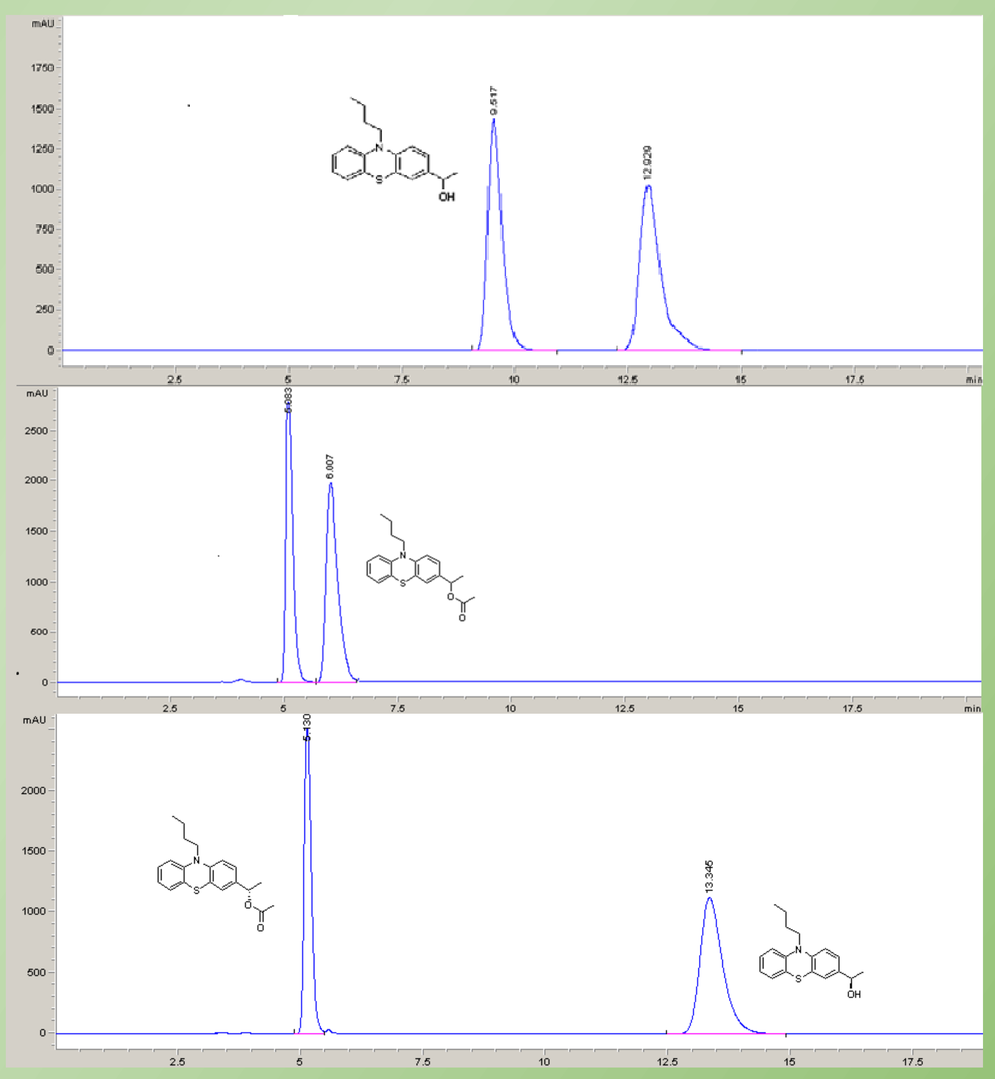

Figure 1. Chromatographic separation of the enantiomers of *rac*-**1f** and *rac*-**2f**; and enantiomeric composition of the products at 50% conversion in $SwCNT_{COOH}$CaL-B mediated enantiomer selective acylation of *rac*-**1f** with vinyl acetate in acetonitrile.