

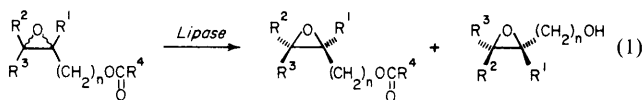
Lipase-Catalyzed Hydrolysis as a Route to Esters of Chiral Epoxy Alcohols¹

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Received May 9, 1984

A current objective of organic chemistry is the development of methods for chiral synthesis.³⁻⁶ This paper describes initial studies of an enantioselective, enzyme-catalyzed hydrolysis of esters of epoxy alcohols which provides an alternative to the Sharpless reaction as a route to these useful chiral synthons (eq 1; this



Compd	R ¹	R ²	R ³
1, 2, 3, 4, 5,			
6, 7, 8, 9, 10		C ₃ H ₇	
11	CH ₃		
12	CH ₃		
13	CH ₃	CH ₃	
14	CH ₃	CH ₃	
15 (n=2)		C ₂ H ₅	
16 (n=2)		C ₂ H ₅	
17 (n=2)		C ₂ H ₅	

equation implies absolute stereochemistry only for 1). The structure of R⁴ is indicated for each compound in Figure 1. The substituents are hydrogens unless indicated otherwise. We are interested in this reaction partly because it offers a practical synthetic route to certain members of a useful class of chiral substances and partly because it provides an opportunity to compare the characteristics of enantioselective biological and abiological synthetic methods.

An initial survey of enzymes^{7,8} that hydrolyze glycidol esters indicated that lipase (E.C. 3.1.1.3, Sigma Type II, from porcine pancreas) has the best combination of activity, selectivity, and cost.⁹ Racemic esters of several epoxy alcohols and related substances were subjected to hydrolysis using this lipase.¹⁰ To facilitate comparison, most reactions were carried to 60% completion (based on the quantity of acid released) under the same conditions (pH 7.8, T = 25 °C). Unhydrolyzed ester was recovered by extraction (recoveries ranged 60–90% of the amount estimated to be present based on acid released). Enantiomeric excesses of recovered ester were measured using Eu(hfc)¹¹ (Figure 1).

These data provide the basis for several observations bearing on the utility of this method. Lipase is not deactivated by reaction

- (1) Supported by the National Institutes of Health, Grant GM 30367.
- (2) NATO postdoctoral research associate 1983–1984.
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- (4) Jones, J. B. In "Asymmetric Synthesis"; Morrison, J. O., Ed.; Academic Press: New York, 1983.
- (5) Wong, C.-H.; Whitesides, G. M. *Aldrichimica Acta* **1983**, *16*, 27–34.
- (6) Sharpless, K. B. *Aldrichimica Acta* **1983**, *16*, 67–79.
- (7) Pig liver esterase, lipase from *Candida cylindracea* and *Rhizopus arrhizus*, and α -chymotrypsin were used in survey experiments.
- (8) Cambou, B.; Klibanov, A. M. *J. Am. Chem. Soc.* **1984**, *106*, 2687–2692.
- (9) The price is <\$25 for 500 g of lipase; it was not immobilized. This enzyme has been used previously for resolutions of alcohols: Laroyre, J.; Verrier, J.; Baratti, J. *Biotech. Bioeng.* **1982**, *24*, 2175–2187. Iriuchijima, S.; Kojima, N. *Agric. Biol. Chem.* **1982**, *46*, 1153–1157. Iriuchijima, S.; Keiyu, A.; Kojima, N. *Agric. Biol. Chem.* **1982**, *46*, 1593–1597. Wang, Y. F.; Chen, C. S.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1984**, *106*, 3695–3696.
- (10) Epoxy alcohols were prepared by use of the procedure of Sharpless (Sharpless, K. B.; Verhoeven, T. R. *Aldrichimica Acta* **1979**, *12*, 63–74) and esters by use of reaction with acid chloride in pyridine/Et₂O (Sonntag, N. O. *V. Chem. Rev.* **1953**, *52*, 237–416).
- (11) The accuracy of the reported values of ee is $\pm 5\%$, except for values >90%; for certain substances in this range, analysis indicated only a value between 90% and 100%.

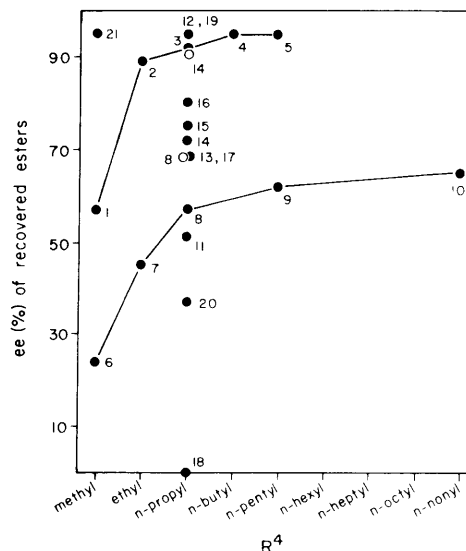


Figure 1. Observed enantiomeric excesses of esters isolated following partial hydrolysis of racemic esters with lipase. Filled points correspond to 60% hydrolysis of the total ester originally present (100% conversion would correspond to complete hydrolysis of both enantiomers of the racemic mixture); open points correspond to 80% hydrolysis.

with the epoxide moiety, and it hydrolyzes a wide variety of structures with useful enantioselectivity.¹² The enzyme is active at water–organic interfaces, and solubility of the organic substrate in water is not necessary. The enantioselectivity depends on the structure of the acid components of the ester (R⁴, eq 1), with better results obtained with longer *n*-alkyl groups (although foaming and emulsification becomes an experimental problem for R⁴ larger than C₅H₉). Since the values of enantiomeric excess increase with extent of conversion in a fashion that is, in principle, well understood,¹³ simply extending reaction times and extent of conversion can be made to yield highly enantiomerically enriched product, albeit at the expense of decreased yield.

A representative experimental procedure for an ester that hydrolyzes with good enantioselectivity is that for glycidyl butyrate (3). A mixture of 300 g of 3 (2.08 mol) and 300 mL of water was placed in a 1-L three-necked flask equipped with a pH electrode, and the two-phase mixture was stirred vigorously with a magnetic stirring bar. Addition of 7.5 g of crude lipase initiated the hydrolysis. The pH was kept at 7.8 by addition of 7 M NaOH using a pH controller. When 60% of the theoretical amount of base required for complete hydrolysis of 3 had been added (178 mL, 1.25 mol, 6-h reaction time) the reaction was poured into 1 L of dichloromethane. The phases were separated and the aqueous phase reextracted with two 200-mL portions of methylene chloride. The combined organic extracts were washed once with 300 mL of 10% NaHCO₃ and twice with 200-mL portions of water, dried (MgSO₄ containing a small amount of Na₂CO₃), and concentrated on a rotary evaporator. Distillation yielded 107 g of (*R*)-3 (0.74 mol, 89% based on the theoretical yield of one enantiomer), bp 81–82 °C (12 torr), with ee = $\geq 92\%$.^{11,14}

A similar procedure for 16 started with 32.5 g (0.174 mol) of 16 and carried the hydrolysis to 75% conversion. We obtained 7.1 g (38 mmol, 88% based on the theoretical yield of one enantiomer) of material with ee = $\geq 95\%$.¹¹

Alcohols were recovered from the reactions summarized in Figure 1 in 30–80% yield by extraction; values of ee were low (30–65%) as expected for resolutions designed to give high ee for

(12) Lipases accept a wide range of substrates. Desnuelle, P. In "The Enzymes", 3rd ed.; Boyer, P. O., Ed.; Academic Press: New York, 1972; Vol VII, Chapter 19. Brockerhoff, H. "Lipolytic Enzymes"; Academic Press: New York, 1974.

(13) Chen, C.-S.; Fujimoto, Y.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1982**, *104*, 7294–7299.

(14) The observed optical rotation for resolved 3 was $[\alpha]_D -26.96^\circ$ (c 20.465, CHCl₃); lit. $[\alpha]_D -28.4^\circ$ for (*R*)-3: Lok, C. M.; Ward, J. P.; van Dorp, D. A. *Chem. Phys. Lipids* **1976**, *16*, 115–122.

the ester products. Alcohols can be prepared with high values of ee by carrying hydrolyses to low conversions.

This enzyme-catalyzed reaction for preparation of chiral epoxy alcohols has advantages and disadvantages relative to transition-metal-catalyzed asymmetric epoxidation. Its major advantage is that it is experimentally the simpler procedure when applicable. A disadvantage of kinetic resolutions is that the theoretical *maximum* yield of chiral product is usually 50% based on racemic starting material (prochiral compounds such as **21** have a theoretical maximum yield of 100%).

Registry No. **1**, 60456-24-8; (\pm)-**1**, 92418-55-8; **2**, 60456-25-9; (\pm)-**2**, 92418-56-9; **3**, 60456-26-0; (\pm)-**3**, 92418-57-0; **4**, 92418-73-0; (\pm)-**4**, 92315-14-5; **5**, 60456-27-1; (\pm)-**5**, 92418-58-1; **6**, 92418-60-5; (\pm)-**6**, 92315-15-6; **7**, 92315-26-9; (\pm)-**7**, 92419-24-4; **8**, 92418-61-6; (\pm)-**8**, 92315-16-7; **9**, 92418-62-7; (\pm)-**9**, 92315-17-8; **10**, 92418-63-8; (\pm)-**10**, 92315-18-9; **11**, 92418-64-9; (\pm)-**11**, 92315-19-0; **12**, 92418-65-0; (\pm)-**12**, 92345-46-5; **13**, 92418-66-1; (\pm)-**13**, 92315-20-3; **14**, 92418-67-2; (\pm)-**14**, 92315-21-4; **15**, 92418-68-3; (\pm)-**15**, 92315-22-5; **16**, 92418-69-4; (\pm)-**16**, 92345-47-6; **17**, 92418-70-7; (\pm)-**17**, 92315-23-6; (\pm)-**18**, 92345-48-7; (\pm)-**19**, 92315-24-7; (\pm)-**20**, 92418-59-2; (\pm)-**21**, 92315-25-8; E.C. 3.1.1.3, 9001-62-1; glycidol, 57044-25-4; 3-propylglycidol, 92418-71-8; 2-methylglycidol, 86884-89-1; (*R*)-3-methylglycidol, 58845-50-4; (*S*)-3-methylglycidol, 92418-72-9; 2,3-dimethylglycidol, 92315-27-0; oxirane-ethanol, 76282-48-9; (*R*)-3-ethyloxiraneethanol, 91603-22-4; (*S*)-3-ethyloxiraneethanol, 91603-21-3.