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Review

Synthesis of chiral building blocks for organic synthesis via lipase-catalyzed reaction: New method of enhancing enzymatic reaction enantioselectivity

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Abstract

The optically active building blocks for organic synthesis: tertiary carbinols, antitumor lignan, liquid crystals, 1,3-diene and biscyclopropyl compounds were synthesized through lipase-catalyzed reaction. This paper discusses ways in which organic chemists can expand the applicability of lipase-catalyzed reactions for use in designing a synthetic strategy. Several excellent examples are described in which lipase-catalyzed reactions were involved as the key steps. Because lipase-catalyzed reactions often offer insufficient enantioselectivity, a new method to enhance the enantioselectivity of a lipase-catalyzed reaction was demonstrated. Thiacrown ether technology was typically used to synthesize new optically active α,α-difluoro-γ-lactone. © 1997 Elsevier Science B.V.

Keywords: Lipase; Asymmetric synthesis; Chiral alcohols; Fluorine compounds; Stannyl compounds; Crown ether; Regulation of enzymatic reactivity

1. Introduction

Lipases are the most frequently used enzymes in organic synthesis because of their stability, availability and their acceptance of a broad range of substrates [1–6]. The synthetic value of lipases has been well recognized because their reactions proceed efficiently and selectively under mild conditions. We wish to discuss ways in which organic chemists can expand the applicability of lipase-catalyzed reactions in preparation of chiral compounds. This paper consist of two parts: the first offers examples of the use of lipase-catalyzed reactions for organic synthesis, the importance of designing the building blocks is discussed in Section 2. The second part concerns our methodology of regulating the enantioselectivity in lipase-catalyzed reactions. Because a limited number of lipases and substrates are applicable for practical enantiomer resolution, development of conventional methods to improve enzyme efficiency in enantioselectivity is very important. We found that two methodologies were effective in achieving this aim: one of them is modification of the substrate and the other is use of a

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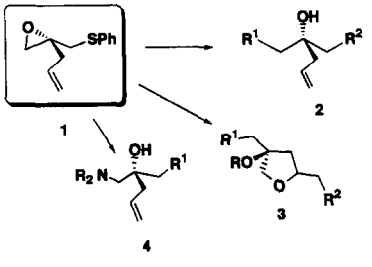
thiacrown ether as an additive that regulates the reactivity of a lipase. Based on the thiacrown ether technology to enhance enantioselectivity of a lipase-catalyzed reaction, we were able to synthesize a new chiral α,α-difluoro-γ-lactone.

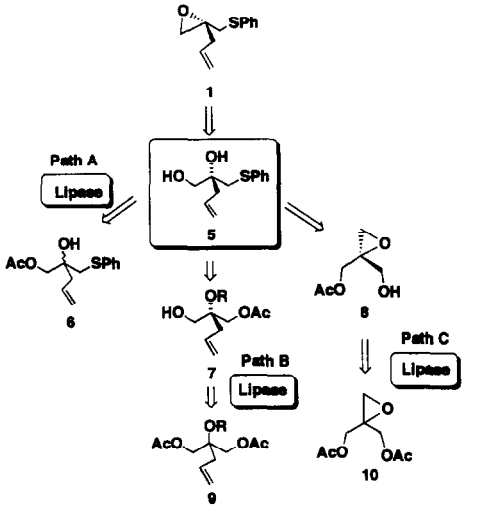
2. Lipase-catalyzed reactions in asymmetric synthesis

2.1. New chiral building block for synthesizing optically active tertiary carbinols

Optically active tertiary alcohol derivatives are widely found in physiologically important compounds [7]. A reasonable way to construct a chiral tertiary hydroxyl moiety is using a synthesis which begins with a chiral building block that possesses a tertiary hydroxyl group with a certain chirality [7–15]. To obtain both enantiomers of tertiary alcohols, we synthesized glycidyl sulfide **1** as a multi-useful chiral building block for tertiary carbinols (Scheme 1) [16]. Subsequent reaction with a variety of nucleophiles gives optically pure β-hydroxyl sulfide, which can again be transformed into a new optically active epoxide while retaining the chiral center [17]. Further, the nucleophilic ring-opening of the newly formed epoxide can lead to a wide variety of optically active tertiary carbinols **2**, **3** and **4**. Therefore, both enantiomeric forms of tertiary alcohols can be obtained by changing the order of the two nucleophilic ring-opening reactions of the optically active glycidyl sulfides **1**.



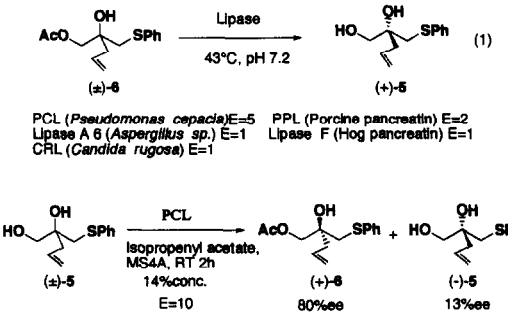
Scheme 1.



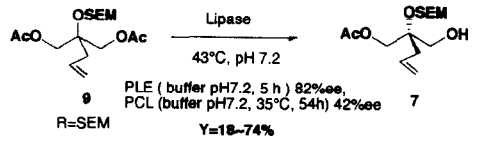
Scheme 2.

Scheme 2 shows a retrosynthetic analysis of glycidyl sulfides **1**. We identified the three pathways, A, B and C, in preparing **1** through lipase-catalyzed reaction. The enantio-discriminating step is the kinetic resolution of racemic substrate **6** in path A, while the concept in path B and path C is an enantioselective reaction of prochiral compounds **9** or **10**, respectively. Path A is the shortest one; therefore, we initially examined this route.

Unfortunately, we found no enzyme capable of hydrolyzing acetate (±)-**6** with a satisfactory enantioselectivity (Eq. 1 in Scheme 3). The highest E value [18,19] recorded was only 5 when (±)-**6** was hydrolyzed by *Pseudomonas cepacia* lipase (PCL) among 40 types of enzymes tested. Acylation of (±)-**5** in organic



Scheme 3.

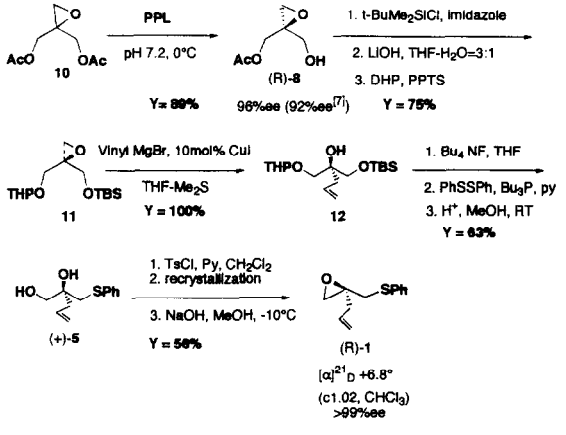


Scheme 4.

media seemed to give a better result, but the reaction had to be stopped at only 7% conversion to obtain the product, (+)-**6**, with high optical purity (Eq. 2 in Scheme 3). Therefore, path A was inadequate from a practical aspect and we thus tried a second pathway.

Recently, Ohta and his coworkers reported highly enantioselective preparation of monoacetate **7** through *Chromobacterium viscosum* lipase-catalyzed hydrolysis [20,21]. The key point of their success in obtaining **7** in high enantioselectivity was the proper choice of the 2-(trimethylsilyl)ethoxymethyl (SEM) unit as a hydroxyl-protecting group (Scheme 4). We found that pig liver esterase (PLE)-catalyzed reaction similarly afforded **7** with high enantioselectivity (Scheme 4), although the expensive SEM group was essential for enantioselective reaction and the chemical yield was insufficient.

The best way to prepare the starting optically active material was path C in which diacetate **10** was hydrolyzed by porcine pancreatic lipase (PPL) to give glycidol diacetate **8** with 96%ee



Scheme 5.

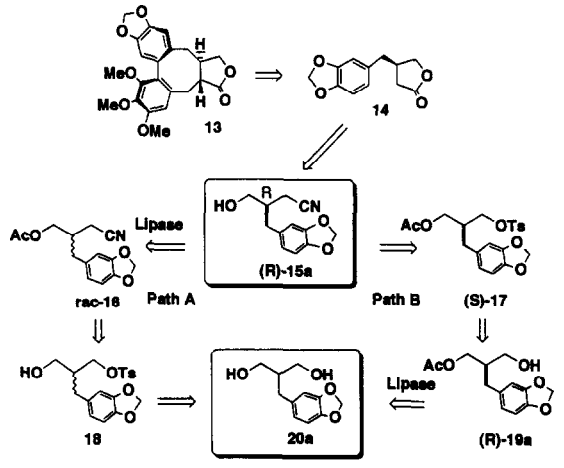
in 89% yield (Scheme 5) [22] [1]. Synthesis of optically active glycidyl sulfide **1** was thus accomplished as shown in Scheme 5. The hydroxyl group of (R)-**8** was first protected as *t*-butyldimethylsilyl (TBDMS) ether. Hydrolysis of the acetyl group and reprotection of the hydroxyl group appeared as tetrahydropyranyl (THP) ether **11**; then reaction with vinyl Grignard reagent gave carbinol **12** in excellent yield. Deprotection of the silyl group and subsequent substitution reaction of the hydroxyl to the phenylthio group [23] and deprotection of THP ether afforded diol (+)-**5**. Tosylation and subsequent alkaline treatment gave epoxide **1** in 56% yield. However, the optical purity of **1** obtained was 86%ee by capillary GPC analysis [2], although the starting material (R)-**8** was 96%ee. We assume that a slight racemization occurred during the step in which **11** was derived from (R)-**8**. Fortunately, recrystallization was successful as the tosylate of (+)-**5** and we obtained epoxide **1** with a satisfied high optical purity (> 99%ee) (Scheme 5).

2.2. *Efficient synthesis of (+)-isostegan*

Our next synthetic target was (+)-isostegan, **13**, which is known as an anti-cancer drug [24] and Scheme 6 is a retrosynthetic analysis of (+)-isostegan. The starting optically pure compound for our isostegan synthesis was γ-hydroxynitrile, (R)-**15a** and it should be prepared from a common prochiral starting material **20a** through two pathways. Path A involved a lipase-catalyzed kinetic resolution of a racemate and path B was a lipase-catalyzed enantioselec-

[1] The authors reported the preparation of optically active **8** (92%ee) using the same enzyme while we were still engaged in writing the manuscript cited in Ref. [16].

[2] The optical purity of **1** was confirmed by GPC analysis using a chiral column (Chiraldex G-TA, ∅0.25×20 m; He gas, 70 ml/min; split ratio = 100:1; $Rt_{(S)}$ = 78 min; $Rt_{(R)}$ = 82 min). Optical purity of the product is shows > 99%ee when no isomer was detected by GPC analysis, while it is > 98%ee by 200 MHz ^{1}H NMR or 188 MHz ^{19}F NMR analysis.

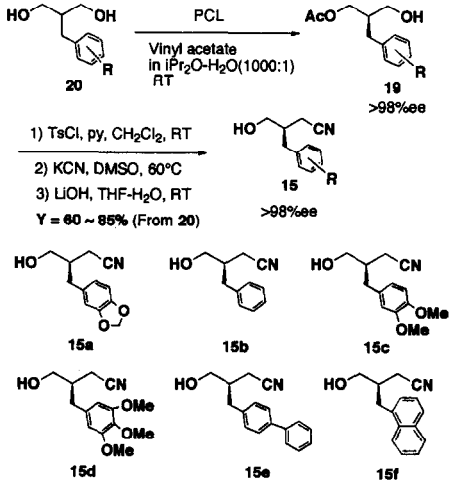


Scheme 6.

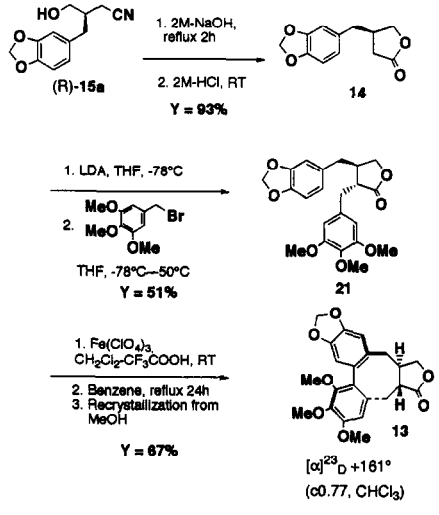
tive conversion of the prochiral diol **20a**. We again learned the importance of strategy for enzymatic reaction in this project. Path A, the kinetic resolution, did not provide the desired compound with sufficient enantioselectivity, although we tested more than 40 types of enzymes.

The asymmetric synthesis approach shown in path B gave excellent results (Scheme 7) [25–28]. Diols **20** were treated with 50% of the weight of PCL in diisopropyl ether as solvent in the presence of vinyl acetate as acyl donor to give the desired monoacetates **19** in excellent yield in optically pure form (> 98%ee). Subsequent tosylation followed by a substitution reaction with potassium cyanide and finally, treatment with lithium hydroxide afforded the γ-hydroxynitriles **15** in 60–85% overall yield [25].

(+)-Isostegan was very conveniently synthesized from optically pure **15a** (Scheme 8) [25]. Nitrile (R)-**15a** was converted to lactone **14** in 93% yield. The ester enolate of **14** was reacted with 3,4,5-trimethoxyphenylmethylbromide to give α,β-disubstituted lactone **21**. This step is stereospecific; only the trans isomer of **21** was obtained. Lactone **21** was then oxidized by $Fe(ClO_4)_3$ in TFA-CH_2Cl_2 solution realizing intramolecular oxidative coupling reaction to give biphenyl lactone [29]. Although the coupling reaction proceeded with excellent regioselectivity, the resulting biphenyl lactone was found by ^1H NMR analysis to be a 6:1 mixture of two diastereoisomers of the desired natural (+)-isostegane **13** and undesired (−)-stegane. Fortunately, the undesired (−)-stegane was isomerized completely to the desired **13** by heating under reflux conditions in benzene for 24 h. Thus, optically pure **13** was obtained from **21** in 67% yield after recrystallization from methanol. We succeeded in the efficient total synthesis of an anti-tumor lignan from optically pure γ-hydroxy nitrile **15a** that was obtained via an enzymatic reaction. Our present synthesis is so sim-



Scheme 7.



Scheme 8.

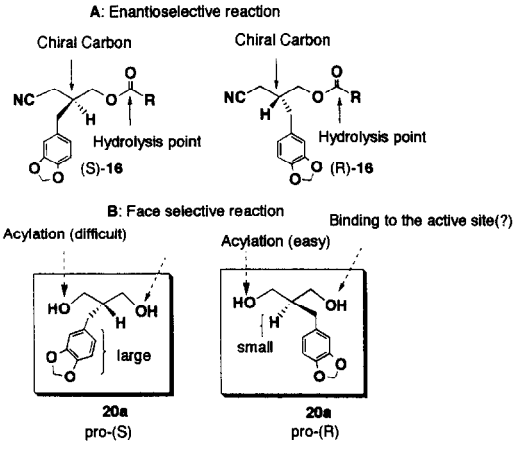


Fig. 1. The enantioselective strategy versus the face selective strategy.

ple that it becomes one of the most promising methods for synthesizing anti-tumor lignans.

The projects described in Sections 2.1 and 2.2 taught us the importance of strategy in preparing optically active compounds even in an enzymatic reaction. For instance, path A in Scheme 6 involves a kinetic resolution, and the hydrolysis point is separate from the chiral carbon in these compounds, so that it might be difficult to distinguish the two isomers, (S)- and (R)-**16**, even for an enzyme (Fig. 1). On the contrary, excellent results were obtained in the reaction of the prochiral substrate **20a** in path B. The observed difference may be due to the fact that **20a** contains an extra OH group to the racemate. This extra hydroxyl group may be able to bind to the active site of the lipase, resulting in a higher degree of enantioselection. In the racemic substrate, **16**, the equivalent position is occupied by a cyano group which presumably will not bind as effectively as OH group. We may be able to explain the difference in enantioselectivity observed between the kinetic resolution of racemate **16** and the asymmetric conversion of the prochiral substrate **20a** from another viewpoint: path B is an easily distinguishable face selective reaction of prochiral substrate and thus offers the desired compound with higher enantioselectivity than path A. The terminal OH group may be effective in that the enzyme distinguishes the preferable face. Therefore, the face selective strategy should be chosen in designing the substrate for enzymatic reactions, if possible.

Incidentally, an empirical rule that predicts which enantiomer of a primary alcohol reacts faster in reactions catalyzed by PCL was proposed by Kazlauskas and Weissflosh [30]. The authors suggest that high enantioselectivity toward primary alcohols requires not only a significant difference in the size of the substituents, but also control of the conformation along the C(1)–C(2) bond; the oxygen at C(2) stabilizes a gaush orientation of the oxygen at the stereocenter and this stabilization may change the favored orientation along the C(1)–C(2) bond thereby changing the enantiopreference [30]. Their empirical rule seems to be useful for designing suitable substrates to the PCL-catalyzed reaction. While our racemic substrate **6** possesses OH group at the stereocenter, PCL-catalyzed reaction was not highly enantioselective (Section 2.1). The attempt to rationalize the enantiopreference of PCL is still in a state of confusion.

2.3. Synthesis of optically active trifluoromethylalkanols

Ferroelectric liquid crystals (FLC) are important high-speed switching devices and their response time strongly depends on the magnitude of their spontaneous polarization (Ps) [31–34]. A recent study revealed that chiral FLC compounds, which involve optically active 1,1,1-trifluoromethylalkanol moieties possess remarkable characteristics: a wide temperature range of the Sc * phase, a large spontaneous polarization

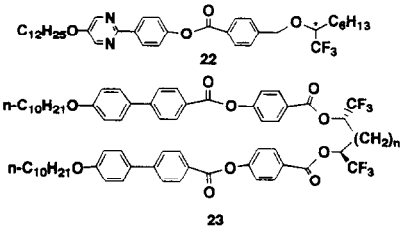


Fig. 2. Chiral ferro- or antiferroelectric liquid crystals.

Table 1
CAL-catalyzed hydrolysis of **26**

Substrate	*n*	Time (h)	% Yield of **27**	% Yield of **28** (optical purity)	% Yield of **29** (optical purity)
26a	7	140	**49**	**20** (> 99%ee)	**20**(< 99%ee)
26b	9	47	**39**	**25**(> 99%ee)	**24**(> 99%ee)
26c	11	30	**39**	**25**(> 99%ee)	**20**(> 99%ee)

and a short response time [33] (Fig. 2). Trifluoromethylalkanols are important components of the liquid crystal compounds **22** [33] and **23** [35] which display remarkable antiferroelectric liquid crystal characteristics.

Lipases are useful in preparing optically pure 1,1,1-tirfluoro-2-alkanols (Scheme 9) [36,37] [3]. We found that the lipase from *Candida antarctica* (CAL) was the best enzyme among 40 types tested. It should be emphasized that the present lipase resolution is the only means of preparing these compounds in the optically pure state (> 99%ee). In addition, because **24e** has a benzyloxy group at the terminal position, this compound is converted to various types of 1,1,1-trifluoro-2-alkanols. For preparing optically active 1,1,1-trifluoro-2-nonanol, **24**, the trans-esterification method gave a product with better %ee than hydrolysis of the corresponding acetate in 0.1 M phosphate buffer at pH 7.2, though the hydrolysis reaction was faster than trans-esterification [37]. We re-examined CAL-catalyzed hydrolysis of 1,1,1-trifluoro-2-nonyl acetate in the mixed solvent system of buffer and acetone (10:1) and found that the alcohol was formed in an optically pure state with good yield. The mixed solvent system seemed effective to suppress the non-enzymatic hydrolysis reaction of the acetate. Therefore, effective enantiomer resolution of 1,1,1-trifluoro-2-alkanols was achieved by both hydrolysis- and trans-esterification methods using CAL.

Lipase-catalyzed hydrolysis of diacetate (±)-**26** gave excellent results (Table 1). Perfect enantiomer resolution was accomplished very easily and diol **28** and diacetate **26** were obtained in optically pure form (> 99%ee) (Scheme 10) [42] [4]. This enzymatic reaction can be used in a large-scale preparation, and the present method therefore affords a valuable means of preparing optically pure key compounds for making antiferroelectric liquid crystals.

2.4. Synthesis of optically active stannyl compounds

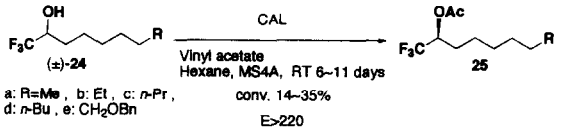
The utilization of organotin compounds in modern synthesis continues to grow at an impressive rate [44,45]. These compounds are fairly stable and can be handled easily; most are stable in air and moisture and are storable without special precautions. Nevertheless, the tin compounds are more reactive than the corresponding silicon compounds and exhibit wide reactivity. Among organotin compounds, hydroxy stannanes are especially valuable for organic synthesis because they contain two functional groups with which it is possible to design a wide variety of synthetic strategies. We previously reported that three types of stannyl compounds, **29** [46] [5], **30** [47] and **31** [48], were obtained with high optical purity through PCL-catalyzed reaction (Scheme 11). We focus here on our recent results to expand the utility of vinylstannane **31** for organic synthesis.

The synthesis of dienes, which possess chiral carbons in their structure, continues to be an area of great interest in organic synthesis [49–58] [59–61]. The copper(II) salt-mediated reaction of alkenylstannanes was reported to provide high stereospecificity for the homocoupling re-

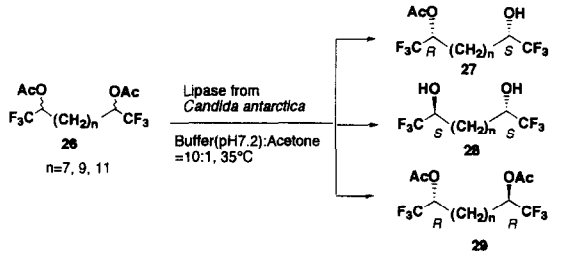
[3] For the synthesis of FLC compounds which involve fluorinated functional groups based on the enzymatic method see Refs. [38–41].

[4] Optical purity of monoacetates, **27**, remains unknown. We tried to determine it by HPLC analysis using a chiral column or ^{19}F NMR analysis of the corresponding (+)-α-methoxy-α-trifluoromethyl-α-phenylacetate (MTPA) [43] of **27**, but none of our efforts has yet been successful.

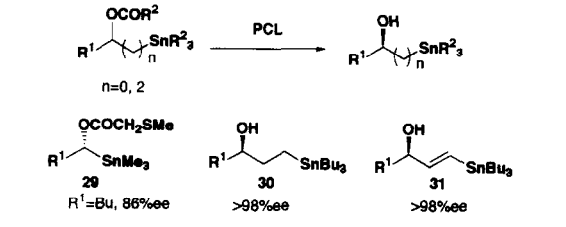
[5] Chong and Mar reported that trans-esterification of α-hydroxystannanes by PPL provided the corresponding acetate with much higher enantiomeric excess [103].



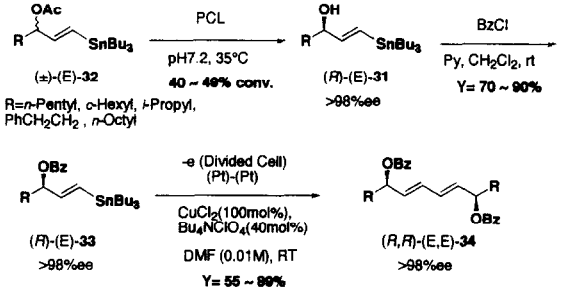
Scheme 9.



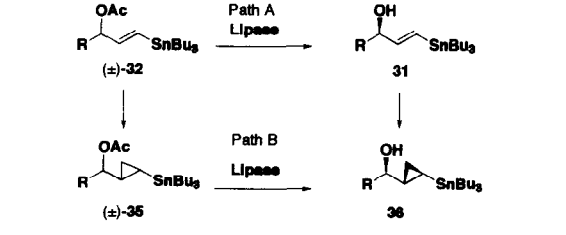
Scheme 10.



Scheme 11.



Scheme 12.

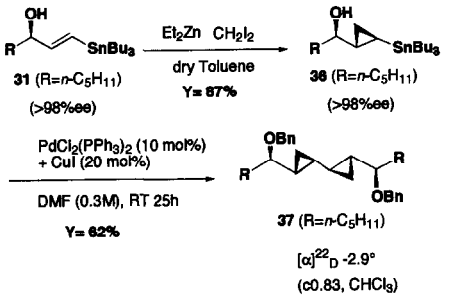


Scheme 13.

action to symmetrical dienes, though it required an excessive amount of copper salt [61]. Because of its highly stereoselective nature, we refined the copper-mediated coupling reaction of alkenylstannanes **31** using electro-oxidation. A combination of electro-oxidation and copper(II) chloride-mediated homocoupling of γ-acyloxyvinylstannanes **33** afforded 1,3-dienes **34** [62]. The stereoselectivity strongly depended on the solvent system and no isomerization occurred when the reactions were carried out in DMF (0.01 M). The synthesis of the four optically active types of dienes, **34**, was thus accomplished by the combined method of electro-oxidation and copper(II)-mediated coupling. This methodology was beneficial in the preparation of various types of optically pure 2,4-diene-1,6-diol derivatives (Scheme 12).

Chiral cyclopropylstannanes are viewed as useful building blocks for synthesizing chiral multi-cyclopropyl compounds of which the moieties are found in some biologically active compounds such as anti-fungal or anti-cancer drugs [63], [64–67] [6]. Synthesis of cyclopropylstannanes [68] were thus demonstrated through two synthetic pathways using a lipase-catalyzed reaction (path A and path B in Scheme 13). Path A might be the preferred route to obtain the target molecule because we have already established the procedure for preparation of these optically pure compounds [48]. Path B is interesting from a biological aspect because cyclopropyl compounds sometimes act as serious inhibitors of an enzyme in microbes, so that this route was initially examined. PCL-catalyzed reaction of (±)-**35** provided the corresponding alcohol **36** with perfect enantioselectivity (> 98%ee), though the reaction stopped at low conversion. We assume that the enantioselectivity of the enzymatic reaction is excellent; however, the reaction appears to be inhibited by the small amount of by-product formed, because

[6] Leading references on the synthesis of multi-cyclopropane compounds.



Scheme 14.

neither the addition of the product nor the substrate inhibited the hydrolysis reaction of PCL. The reaction in path A was useful in preparing chiral cyclopropane derivatives. Optically active tributylstannylcyclopropane derivatives, **36**, were prepared from the corresponding vinylstannane **32** in 87% yield (Scheme 14). Biscyclopropane **37** was thus synthesized via homocoupling reaction using a palladium catalyst [69] [7].

3. Regulation of lipase-catalyzed reactions in asymmetric synthesis

3.1. Enhanced enantioselectivity of lipase-catalyzed reaction

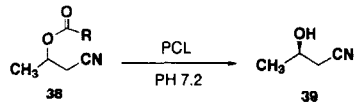
Lipases can hydrolyze a wide variety of compounds, although sometimes low enantioselectivities are observed as mentioned in Section 1. For instance, the best enantiomeric excess of **39** produced was ca. 60%ee in the hydrolysis of **38a** (R = Me) at 45% conversion using PCL after testing 24 types of commercial enzymes [70]. This result is not acceptable from a practical viewpoint. Two methods were demonstrated to improve lipase-catalyzed reaction performance: modification of the substrate and use of an additive that regulates the reactivity.

Design of the acyl part of the substrate can enhance enantioselectivity of the product, because enantioselectivity is dependent on the size and nature of the acyl part. A detailed study of the enantio-preference of PCL was carried out using various esters **38** (Scheme 15) [71]. Table 2 indicates that introduction of a hetero-atom into the acyl group clearly enhanced the *E* value. Among the acyl groups examined, methylthioacetate was confirmed as the one affording the highest enantioselection.

The additive method is more advantageous. It is simple to use, but only several compounds have been reported to enhance enantioselectivity of the lipase reaction [72] [8], [73] [9], [74] [10], [75] [11], [76] [12], [77] [13], [78] [14], [79] [15]. Guo and Sih first reported that dextromethorphan and levomethorphan functioned as enantioselective inhibitors in *Candida cylindracea* lipase-catalyzed hydrolysis of (±)-aryloxy- or arylpropionic esters [72]. We found that *l*-methioninol similarly enhanced enantioselectivity in the PCL-catalyzed hydrolysis of **38a** [73]. Each of these remarkably improved the enantioselectivity but rarely increased the reaction rate. Crown ethers are known as complexing agents for several proteins [80] and Reinhoudt et al. reported that serine proteases were activated by crown ethers [81–86] [16]. We recently found that some crown ethers had potential to enhance both the enantioselectivity and reaction rate in the lipase-catalyzed hydrolysis of acetate **38a** (Scheme 16) [87–89]. We chose acetate **38a** and PCL as the substrate and enzyme in the study,

[7] Optical purity of monoacetates, **37**, remains undecided. Recently an excellent dimerization protocol for the cyclopropylstannane has been reported in Refs. [66,67] and the references cited therein.

[8] Dextromethorphan.
[9] l-Methioninol.
[10] Sodium chloride.
[11] Sodium chloride.
[12] Calcium chloride.
[13] Potassium chloride.
[14] Triton X-100.
[15] D-Sorbitol.
[16] 18-Crown-6 enhanced enantioselectivity of α-chymotrypsin-catalyzed trans-esterification of N-acetyl-D,L-phenylalanine esters in organic solvent.



Scheme 15.

because the nitrile **39** produced is an employable chiral building block and PCL is applicable to various substrates [30]. To avoid complexation between the crown ether and the metal cation, hydrolysis was generally performed in non-buffered aqueous solution. The enantioselectivity depended strongly on the nature of the additive. We examined ten crown ethers, one cryptand, eight hetero-macrocycles, eight armed macrocycles and five acyclic analogs as additives. They vary widely in structure and guest-binding property. Scheme 16 summarizes typical examples of crown ether additives which significantly enhanced both enantioselectivity and reaction efficiency in the hydrolysis. In particular, 1,4,8,11-tetrathiacyclotetradecane (**43**) was confirmed as the best additive. The highest E value recorded was 37 when the hydrolysis was carried out in the presence of additive **43** (Scheme 16) [87].

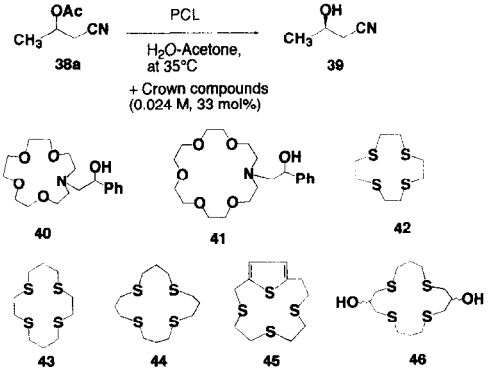
Our employed crown ether additive cannot change the original stereochemistry of the product but does enhance its potential ability to a level at which the reaction can be used practically. We recently found that addition of thiacrown ether **43** greatly enhanced the reactivity of the lipase toward several allyl alcohols [90] [17]. There were clearly differences in the additive effect in the regioselectivity depending on the enzyme's origin. The action of PCL and *Candida rugosa* lipase (CRL) was particularly strongly modified by the thiacrown ether additive [18]. We currently assume that two factors are

Table 2
Effect of modification of the acyl group on enantioselectivity

R	E	R	E
CH_3	7	CH_2SBu	13
n-C_3H_7	2	CH_2SPh	13
n-C_4H_9	3	CH_2OPh	10
CH_2SMe	29	CH_2SO_2Me	1
CH_2OMe	14	CH_2Cl	6

involved in the reaction. One is the interaction between the thiacrown ether and the enzyme. The employed crown ether may interact with certain sites of the lipase, thereby modifying its local conformation, activating it, and causing a change in the stereoselectivity of the enzymatic reaction as proposed in the CRL-catalyzed reaction after treatment with 2-propanol [91] or sodium deoxycholate [92]. The second factor is the complexation of the crown ether with the product. We observed that thiacrown ether additives induced significant changes in ^{13}C NMR spectra of the products or substrates. Therefore, we assumed that the thiacrown ether may bind such neutral molecules in the course of the reaction, so that chemical equilibrium of the reaction occurring near the active site is modified.

This finding represents not only a significant advance in improvement of lipase-catalyzed organic synthesis but also provides an interesting combined use of crown ethers with enzymes. The approach is therefore recommended as a



Scheme 16.

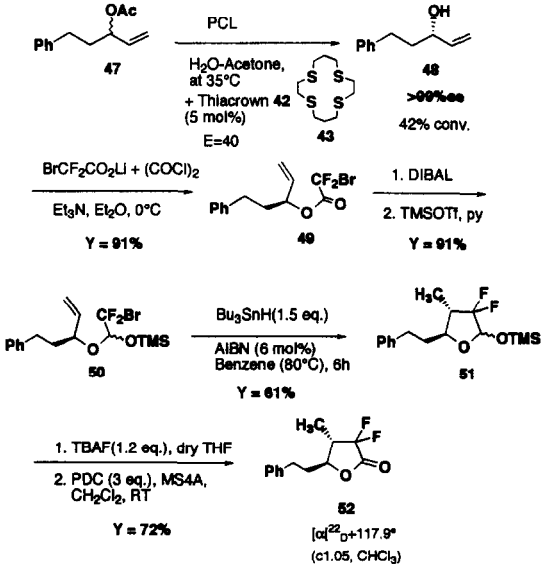
[17] Enantioselectivity in both the hydrolysis of **47** and the acylation of **48** by PCL was enhanced by addition of 5 mol% of thiacrown **43**.

[18] Chemical yield and regioselectivity of the PCL or CRL-catalyzed partial hydrolysis of 4-acetoxy-2-methyl-2-butenyl acetate were also greatly enhanced by addition of 5 mol% of thiacrown ether 43 to the substrate (unpublished results).

new technique by which to regulate enzymatic reactions by chemical reagents.

3.2. Synthesis of new α,α-difluoro-γ-lactones through lipase-catalyzed reaction and intramolecular radical cyclization

Thiacrown ether additive **43** remarkably enhanced the enantioselectivity in the lipase-catalyzed hydrolysis of 5-phenyl-1-penten-2-yl acetate (**47**) [90]. Enantioselective hydrolysis of **47** was carried out in the presence of 5 mol% of thiacrown ether **43** providing the corresponding alcohol **48** with excellent enantioselectivity, an *E* value of more than 40. The reaction provided an *E* value of 15–17 in the absence of the thiacrown. This allyl alcohol **48** was converted to the corresponding α-bromo-α,α-difluoroacetate, **49**, then changed to TMS acetal **50**. The acetal was converted to the corresponding lactol, **51**, through intramolecular radical cyclization [93]. Deprotection and PDC oxidation afforded γ-lactone **52** in 72% yield (Scheme 17). Because intramolecular radical cyclization proceeded highly stereoselectively, *trans*-isomers of the γ-lactone **52** were obtained as a sole product. We thus succeeded in synthesizing optically active α,α-difluoro-γ-lactones using a thiacrown ether-modified lipase-catalyzed reaction as a key reaction.



Scheme 17.

4. Closing remarks

Lipase-catalyzed reactions are particularly useful in preparative organic synthesis. They offer high efficiency and selectivity especially in asymmetric synthesis under mild conditions. The preparation of optically active compounds in lipase-catalyzed reactions has sometimes been criticized as being just the kinetic resolution of racemic substrates, so that the maximum yield is basically 50%, except for the reaction of prochiral substrate [94–97]. Fortunately, recent examples clearly surpassed this limitation by the concept of 'dynamic resolution protocol' in the lipase-catalyzed reaction [98–102].

In this review, we have described our original results as examples of organic synthesis-oriented enzymatic reactions. Specific experimental problems have been overcome by establishing two new methods for enhancing the activity of lipase-catalyzed reactions and a variety of useful chiral molecules have been prepared as mentioned in Chapter 2. Although the number of successful applications to date is limited, a lipase-catalyzed reaction using an artificial substrate will undoubtedly allow us to develop a smarter and more rapid organic synthesis.

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