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Lipase-catalyzed resolution of 4-aryl-substituted β-lactams: effect of substitution on the 4-aryl ring

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This paper is dedicated to Professor David Crout to mark the occasion of his retirement

Abstract—*Pseudomonas cepacia* lipase (PS-30) was used in hydrolytic resolution of 3-acetoxy-4-aryl-substituted azetidin-2-ones (>97% ee). Twenty-three β-lactam substrates with varied substituents at the C-4 center of the ring were synthesized and subjected to lipase-PS catalyzed hydrolysis in phosphate buffer (pH 7.2, 0.2 M) at 25°C. The reactions occurred with high enantioselectivity and substrate conversion. The effect of substitution on the C-4 aryl ring on lipase hydrolytic activity was dependent upon the steric and electronic nature of the substituent and its position on the aryl ring. The stereopreference of the lipase PS-30 for the (3*S*,4*R*) enantiomer was rationalized using a known active site model. Absolute stereochemistry of the enantiomers was established using single crystal X-ray crystallographic techniques.

1. Introduction

The use of lipolytic enzymes, such as lipases, provides one of the most useful and versatile biocatalytic methods in asymmetric synthesis and resolution of organic substrates with high efficiency and selectivity.[1–4] Lipases, in this regard, are excellent biocatalysts since they have the remarkable ability of assuming a variety of conformations to accommodate substrates of varying sizes and complexities.[5–9] Above all lipases do not require the use of cofactors, can be recycled, and are effective under mild, environmentally benign conditions. As a result of this broad substrate specificity and their distinctive stereopreferences, lipases have been used in numerous applications ranging from enantioselective syntheses to the resolution of racemic mixtures.[5–9]

Stereoselective synthesis of enantiomerically pure compounds is required not only for clinical compounds but also for fine chemical industries and agrochemicals. It is possible that an enantiomer in a racemic mixture may have detrimental side effects. Syntheses of such homochiral compounds, however, are often too difficult to obtain via conventional chemical synthetic methods. Recently, we have reported the preparation of optically active substituted ε-caprolactones,[10] versatile building blocks in organic synthesis, through lipase-catalyzed kinetic resolution in absence of added solvent. In another report,[11] we have described solventless stereoselective polymerizations of racemic substituted caprolactones catalyzed by *Candida antarctica* lipase.

The β-lactam (azetidin-2-one) is an interesting ring system found in penicillins, one of the most utilized antibacterials in clinical medicine.[12,13] Moreover, β-lactams can be readily synthesized via the Staudinger reaction starting from an amine and an aldehyde (Scheme 1) and serve as chiral synthons in organic synthesis.[13] For example, β-lactam have been used for assembly of the C-13 side chain in the anti-tumor drug paclitaxel,[14] in the asymmetric synthesis of human leukocyte elastase inhibitors,[15] and as key intermediates for penems and carbepenems.[16] Recently, the stereocontrolled synthesis of monocyclic β-lactams has been an active area of chemical research owing to their utility as synthons for a variety of natural products[14] and activity against MRSA bacterial strains.[13] Since these compounds are interesting from chemical and pharmacological aspects, our goal was to study lipase catalyzed resolution of monocyclic β-lactams in order to obtain them in optically pure form. The previous results on enzymatic resolution of racemic β-lactams have largely concentrated on resolution of *N*-hydroxymethylated derivatives[17–22] via lipase catalyzed asymmetric acylation of the primary hydroxyl on the *N*-hydroxymethyl group or by the

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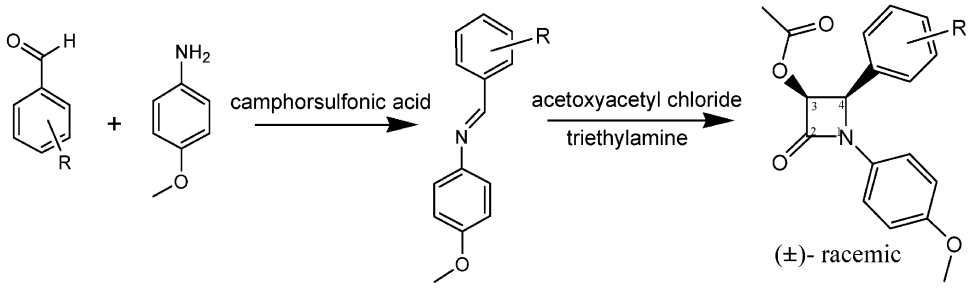


Scheme 1. General synthesis of 4-aryl-substituted β-lactams. **1** R=H; **2** C4=(−CH=CH−Ph); **3** C4=(−C≡C−Ph); **4** C4=(−C_4H_3S); **5** R=4-OCH_3; **6** R=4-C(O)OCH_3; **7** R=4-NO_2; **8** R=3-NO_2; **9** R=4-CN; **10** R=4-$(CH_3)_2N$; **11** R=3-F; **12** R=4-F; **13** R=4-Cl; **14** R=3-Cl; **15** R=2-Cl; **16** R=2,4-di-Cl; **17** R=2-Br; **18** R=3-Br; **19** R=2-I; **20** R=4-Br; **21** R=4-CF_3; **22** R=4-$C(CH_3)_3$; **23** R=4-$CH(CH_3)_2$.

hydrolysis of their corresponding ester derivatives.[17–22] In a different approach, the β-lactam ring has been enantiospecifically hydrolyzed via enzymatic catalysis to the corresponding β-amino acid.[23,24] This approach, however, results in loss of the β-lactam ring in the hydrolyzed enantiomer and would not be synthetically appealing especially if the hydrolyzed enantiomer is needed for subsequent synthetic transformations. In their quest to prepare the C-13 side-chain of paclitaxel, Brieva et al.[14] obtained enantiomerically enriched 3-hydroxy-4-phenyl-β-lactams derivatives via lipase-catalyzed hydrolysis and transesterification of racemic ester and alcohols, respectively. These authors[14] subjected three β-lactams with different substituents (H−, PhCO−, and 4-CH_3OPh−) on the ring nitrogen and observed that lipase-catalyzed resolutions were influenced by the nature of the substituents on the lactam nitrogen.[14] For example, introduction of the *N*-benzoyl substituent, an electron withdrawing group (EWG), caused the cleavage of the β-lactam ring in *tert*-butyl methyl ether in the presence of CH_3OH as a nucleophile.[14]

Our goal in this study was to develop an enzyme system that invokes a resolution of the monocyclic β-lactams upon hydrolysis of its 3-acetoxy derivatives and to probe the effects of the nature of the lactam ring C-4 substituents on enzyme selectivity. In this report, kinetic enzymatic resolution of the 3-acetoxy derivatives was investigated as an attractive alternative to the classical salt resolution or the resolution of the *N*-hydroxymethyl derivatives.[25] The kinetic resolution led to quantitative yields and enantiomerically enriched (>97% ee) β-lactams were thus prepared. Importantly, the nature of the substituent on the C-4 position affected the enzymatic resolution to a great extent. Our efforts to understand the effect of the C-4 substituent on the selectivity of the lipase catalyst are reported herein.

2. Results and discussion

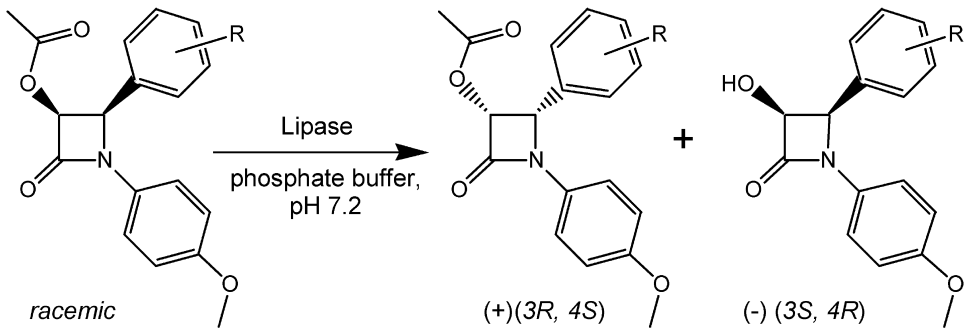
2.1. Synthesis of 4-aryl-substituted beta-lactams (±1–±23)

The racemic 3-acetoxy-1,4-diaryl-azetidin-2-ones (±**1**–±**23**) were synthesized in a three-step reaction scheme as described previously[13] starting with the acid-catalyzed Schiff base formation from the corresponding substituted benzaldehyde and *para*-methoxyaniline. The Schiff base was then reacted with acetoxyacetyl chloride and triethylamine in dichloromethane to yield the β-lactam via a [2+2]-cycloaddition reaction (Scheme 1). In all reactions, only one diastereoisomer possessing the C-3 acetoxy and C-4 aryl groups in the *cis* configuration was produced resulting in formation of (±)-racemates.

2.2. Lipase-catalyzed asymmetric deacetylation of ±1–±23

Our experiences with lipase catalyzed resolution of esters and lactones[6] in anhydrous organic solvents prompted investigation of the deacetylation of the β-lactams in anhydrous solvents. *Pseudomonas fluorescens* (AK), *Porcine pancreatic lipase* (PPL) and *C. antarctica* (Novozym-435) lipases did not catalyze the transesterification reaction between the *n*-butanol and β-lactam acetoxy group in anhydrous tetrahydrofuran (THF), which we have previously found to be a solvent of choice for lipase catalyzed transesterifications.[26] *Pseudomonas cepacia* (PS-30) lipase did catalyze the transesterification in anhydrous THF, however, deacetylation proceeded at an exceedingly slow rate and prolonged reaction times (>6 days) were required for any reasonable substrate conversion. For example, hydrolysis of compound **7** in buffer (pH 7.2, 0.2 M) led to 50% conversion within 48 h, while deacetylation in dry THF/*n*-butanol gave only 35% substrate conversion after 6 days.

The synthesis of the stereochemically pure β-lactam, therefore, was conceived via hydrolysis (Scheme 2) of the racemic acetates (±)**1**–(±)**23**, which were exposed to a series of lipases from different sources (PS-30, Novozym-435, AK, PPL) in aqueous buffer. Generally, microbial lipases exhibit pH optima ranging from 5.6 to 8.5 and at temperature between 30 and 40°C; we found that highest conversions were obtained in pH 7.2 (0.2 M) phosphate buffer at 25°C. Of the enzymes screened, lipases Novozym-435, AK and PPL did catalyze the hydrolysis of the β-lactam acetates in buffer solution, however, long reaction times (>10 days) were required. With *P. cepacia* lipase (PS-30) reactions proceeded with moderate to excellent substrates conversion and hence it was identified as the most suitable biocatalyst for hydrolysis. Brieva[14] et al. also had previously described the lipase PS-30 as a suitable catalyst for enantioselective hydrolysis of the racemic 3-acetoxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one (**1**) in sodium phosphate buffer (pH 7.5, 0.2 M). For optimal production of the alcohols (−)**1a**–(−)**23a** (Scheme 2), water insoluble



Scheme 2. Lipase catalyzed asymmetric deacetylation of 4-aryl-substituted β-lactams. **1** R=H; **2** C4=(–CH=CH–Ph); **3** C4=(–C≡C–Ph); **4** C4=(–C_4H_3S); **5** R=4-OCH_3; **6** R=4-C(O)OCH_3; **7** R=4-NO_2; **8** R=3-NO_2; **9** R=4-CN; **10** R=4-$(CH_3)_2N$; **11** R=3-F; **12** R=4-F; **13** R=4-Cl; **14** R=3-Cl; **15** R=2-Cl; **16** R=2,4-di-Cl; **17** R=2-Br; **18** R=3-Br; **19** R=2-I; **20** R=4-Br; **21** R=4-CF_3; **22** R=4-$C(CH_3)_3$; **23** R=4-$CH(CH_3)_2$.

racemic acetates (±)**1**–(±)**23** were dissolved in a minimal amount of acetone and dispersed in an aqueous phosphate medium (pH 7.2, 0.2 M) at 25°C. The reaction was initiated by addition of predetermined amounts of lipase PS-30. The product alcohol and the unreacted acetate were extracted with ethyl acetate and separated readily by silica-gel column chromatography (Table 1). Using a stepwise elution of ethyl acetate/petroleum ether (15:85) followed by ethyl acetate/petroleum ether (30:70), unreacted acetate and the alcohol were isolated in successive order.

The enantiopurity of the unreacted acetates were calculated from their ^{1}H NMR spectra acquired in presence of (+)-Eu(hfc)$_3$, a chiral shift reagent (Fig. 1). The resonance signal of the acetoxy protons in the racemic mixture, a singlet in absence of the chiral shift reagent, was split into two signals of equal intensity for the two enantiomers in the presence of (+)-Eu(hfc)$_3$. The product alcohol was acetylated and enantiomeric purity was determined using (+)-Eu(hfc)$_3$ in ^{1}H NMR experiments.

The activity of different lipases was greatly dependent upon its source i.e. while lipases CALB (Novozym-435, immobilized form from *C. antarctica*), AK (from *Pseudomonas fluorescens*) and (PPL) showed little activity, the lipase PS-30 (from *P. cepacia*) was highly efficient in buffer solution. The effect of the enzyme concentration on the substrate conversion was studied using a 1:1 (w/w) and 2:1 (w/w) lactam **1** to lipase PS-30 ratio (Fig. 2). The rate of acetate hydrolysis, as expected, was higher using more lipase in the reaction, for example, 50% conversion was achieved after 11 h using 1:1 while more than 18 h were required using 2:1

Table 1. Lipase PS-30 catalyzed hydrolysis of substituted β-lactams (±)**1**–(±)**23**

Substrate[a]	Conv[b] (%)	(3*R*,4*S*)-acetoxy-			*E*	(3*S*,4*R*)-hydroxy-		
		ee_s[c]	% Yield[d]	$[\alpha]_D^{25}$[e]		ee_p[c]	% Yield[d]	$[\alpha]_D^{25}$[e]
–4-Phenyl (**1**)	50	>97	71	+11.6	277	>97	98	−184.6
–4-(2-Phenylethyenyl) (**2**)	48	91	99	+59.6	209	>97	96	−348.0
–4-Phenylethynyl (**3**)	48	>97	99	+149	277	>97	96	−353.0
–4-Thiophen-2-yl (**4**)[f]	49	>97	88	+19.2	277	>97	98	−115.0
–4-(4-Methoxyphenyl) (**5**)	50	>97	87	+16.7	277	>97	95	−216.3
–4-(4-Methoxycarbonylphenyl) (**6**)	50	>97	98	+43.3	277	>97	95	−404.4
–4-(4-Nitrophenyl) (**7**)	50	>97	98	+56	277	–	99	0
–4-(3-Nitrophenyl) (**8**)	48	>97	84	+37.7	277	>97	96	−194.2
–4-(4-Cyanophenyl) (**9**)	29	40	66[g]	+15.1	97	>97	60	−203.6
–4-(4-Dimethylaminophenyl) (**10**)	39	71	80	+29.5	140	>97	69	−175.4
–4-(3-Fluorophenyl) (**11**)	50	>97	97	+13.3	277	>97	98	−328.0[h]
–4-(4-Fluorophenyl) (**12**)	50	>97	93	+2.2	277	>97	99	−203.9
–4-(4-Chlorophenyl) (**13**)	50	>97	88	+21.5	277	>97	66	−208.2
–4-(3-Chlorophenyl) (**14**)	49	>97	92	+12.5	277	>97	96	−338.0
–4-(2-Chlorophenyl) (**15**)	35	56	90	+62.4	115	>97	72	−284.7
–4-(2,4-Dichlorophenyl) (**16**)	28	29	57[g]	+20.1	87	>97	48	−238.4
–4-(2-Bromophenyl) (**17**)	36	64	85	+229.0	127	>97	56	−187.0
–4-(3-Bromophenyl) (**18**)	19	34	68f	+4.7	91	>97	46	−138.6
–4-(2-Iodophenyl) (**19**)	21	15	68[g]	+6.5	76	>97	10	−153.1
–4-(4-Bromophenyl) (**20**)	50	>97	98	+265	277	>97	99	−174.4
–4-(4-Trifluoromethylphenyl) (**21**)	50	>97	99	+196.5	277	>97	98	−150.0[h]
–4-(4-Tertbutylphenyl) (**22**)	17	10	80	+90.5	72	>97	15	−183.5
–4-(4-Isopropylphenyl) (**23**)	27	68	75	+194.5	134	>97	25	−187.2

[a] Represents the aryl substituents on the β-lactam ring.
[b] Determined from ^{1}H NMR of crude product mixture after 48 h.
[c] Determined from ^{1}H NMR spectra in the presence of Eu(hfc)$_3$, by integration of CH_3C=O.
[d] Yield 100% at 50% conversion.
[e] *c*=0.01 (g/mL), $CHCl_3$
[f] Configuration of unreacted substrate (3*R*,4*R*) and that of reacted substrate (3*S*,4*S*).
[g] Yield 100% at 0% conversion.
[h] *c*=0.01 (g/mL), CH_3OH.

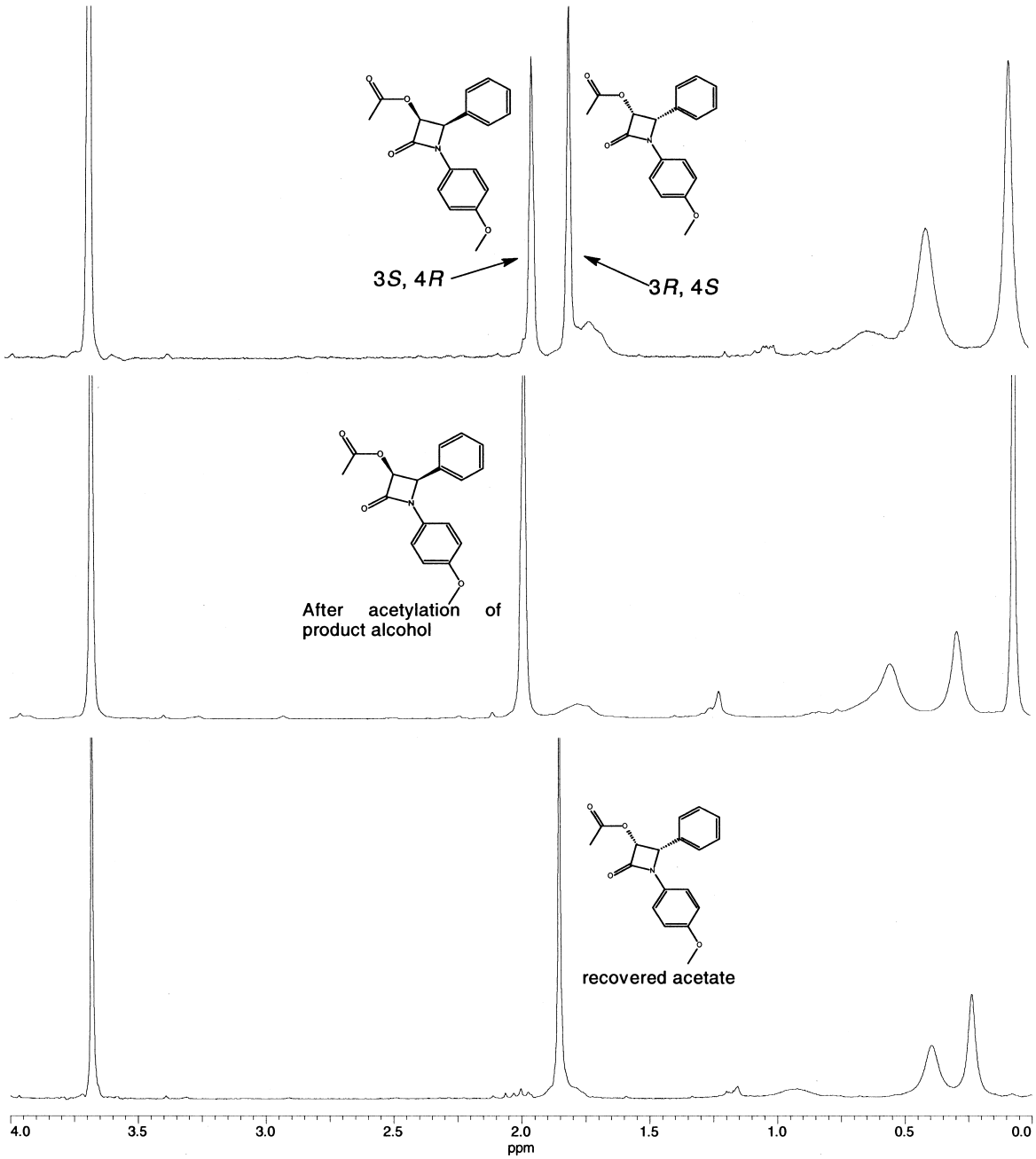


Figure 1. (a) ^{1}H NMR of racemic 3-acetoxy-1-(4-methoxyphenyl)-4-phenyl-β-lactam with Eu(hfc)$_3$. (b) ^{1}H NMR of unreacted (+) (3*R*,4*S*)-3-acetoxy-1-(4-methoxyphenyl)-4-phenyl-β-lactam with Eu(hfc)$_3$. (c) ^{1}H NMR of acetylated (−) (3*R*,4*S*)-3-hydroxy-1-(4-methoxyphenyl)-4-phenyl-β-lactam with Eu(hfc)$_3$.

substrate to lipase ratios. For subsequent studies 1:1 (w/w) substrate to lipase ratio was used (Table 1).

To understand the effect of β-lactam ring substitution on hydrolytic activity of lipase PS-30, 23 β-lactam substrates with different substituents on C-4 were synthesized and subjected to lipase-catalyzed hydrolysis in phosphate buffer (pH 7.2, 0.2 M). The results of the *P. cepacia* lipase-catalyzed hydrolysis after 48 h are shown in Table 1. Even though lipase PS-30 accepted substituted β-lactam rings with phenyl (**1**), thiophen-2-yl (**4**), phenylethenyl (**2**), and phenylethynyl (**3**) at the C-4 position as substrates, the C-4 substituent on the β-lactam ring influenced the hydrolysis to a great extent and substrate conversions were greatly affected (Table 1). For all substrates, the lipase-catalyzed hydrolysis proceeded with high enantioselectivity towards the 3*S* enantiomer. For example, in compound **1**, prolonged reaction time (>72 h, not shown) beyond 50% conversion did not result in hydrolysis of the 3*R* enantiomer (Fig. 2) indicating that the enzymatic reaction proceeded with great efficiency as measured by enantioselectivity (E=277) and substrate conversion (~50%). The lipase enantioselectivity (E) values were determined from the following equation: $E=\ln[(1-c)(1-ee_s)]/\ln[(1-c)(1+ee_s)]$ where the $ee=[(S)-(R)]/[(S)+(R)]\times 100\%$. The enantiomeric excesses were determined from ^{1}H NMR spectra recorded in presence of the (+)-[Eu(hfc)$_3$] (Fig. 1).

To understand how the C-4 substituent affected lipase PS-30's ability to hydrolyze the acetoxy group, a number of

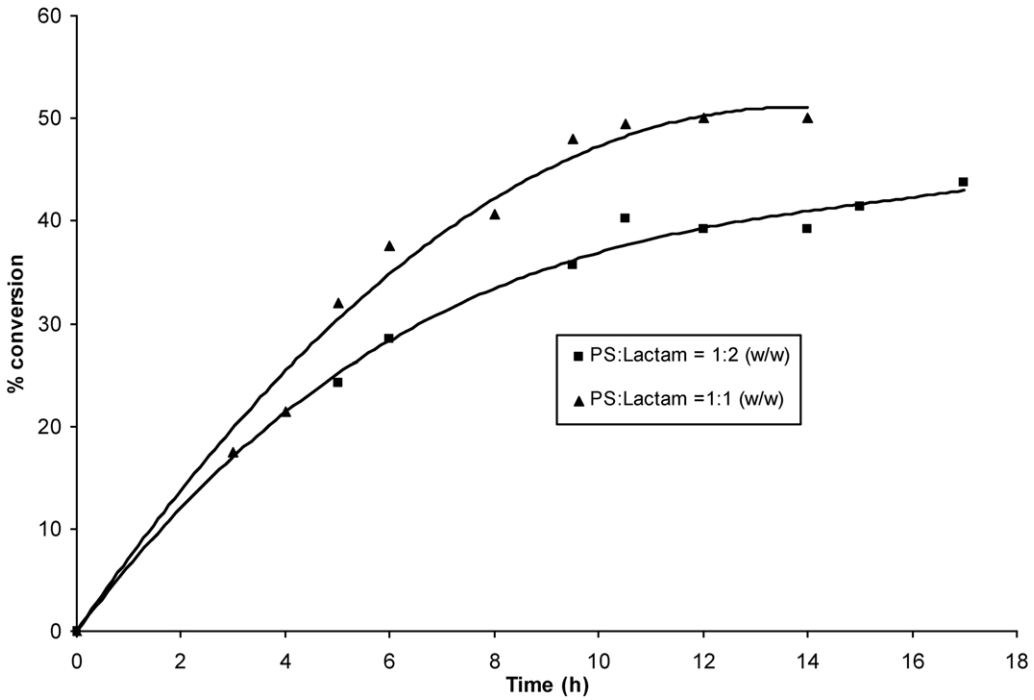


Figure 2. Variation in substrate conversion with time using lactam **1** to PS-30 ratio (w/w) of 1:1 (▲) and 2:1 (■).

substrates that differ in the position, size and electronic nature of substituent(s) on the C-4 phenyl ring were subjected to hydrolysis and substrate conversions were carefully monitored as a function of reaction time by ^{1}H NMR (Fig. 3). From Figure 3, it is important to note that:

- The hydrolytic ability of the lipase PS-30, as illustrated by substrate conversion, was greatly affected by the substituent(s) on the C-4 aryl ring.
- Substrates **6**, **14**, **13**, **20** and **21** with EWG on the aryl ring typically had quantitative conversions (~50%) within 48 h, while compounds **5**, **10**, **22** and **23** with electron donating groups (EDG) had lower substrate conversions and more than 48 h were needed to achieve reasonable conversion (Fig. 3 and Table 1).
- Among the EWGs screened, the position of the substituent(s) on the phenyl ring was of importance in realizing high conversion. In general, a group at the C-4′ position i.e. further away form the lactam ring, on the phenyl moiety gave higher conversion. For example, with C-4′ chloro (in **13**) or bromo (in **20**) substituents quantitative conversions were achieved but the C-2′ chloro (in **15**) and bromo (in **17**) analogs gave only 32 and 27% substrate conversions, respectively, in 13 h (Fig. 3). In the case of compound **16** having two chloro groups at C-4′ and C-2′, the substrate conversion were

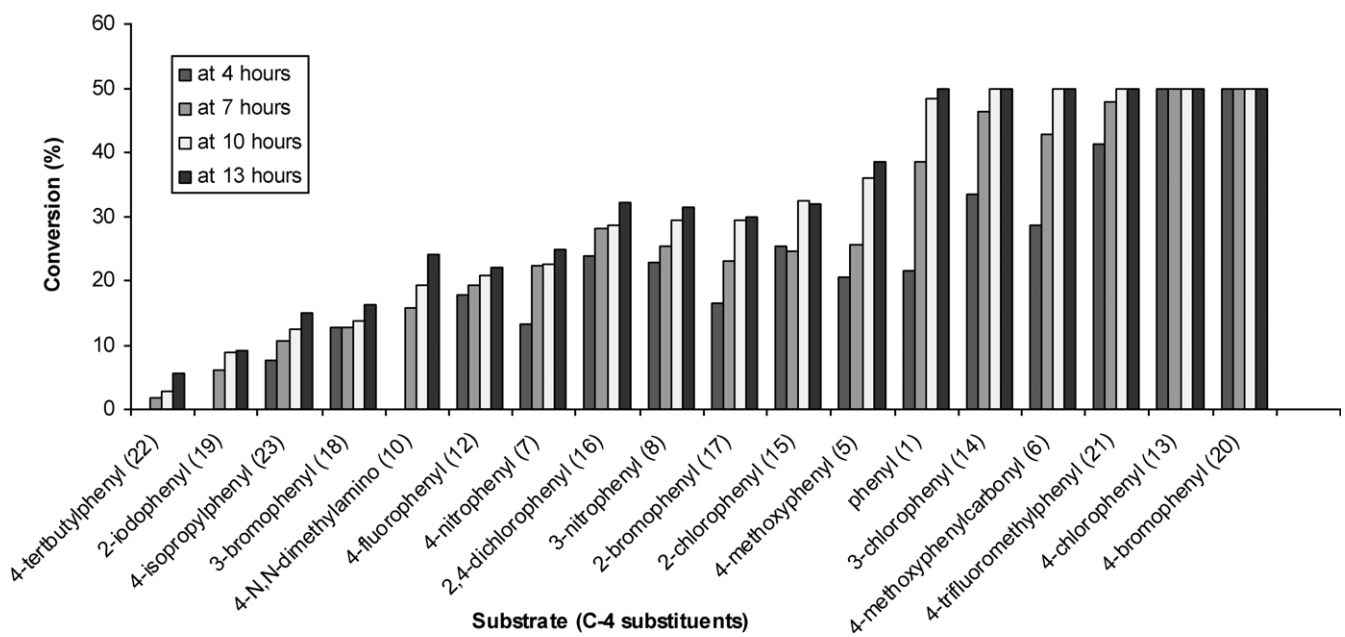
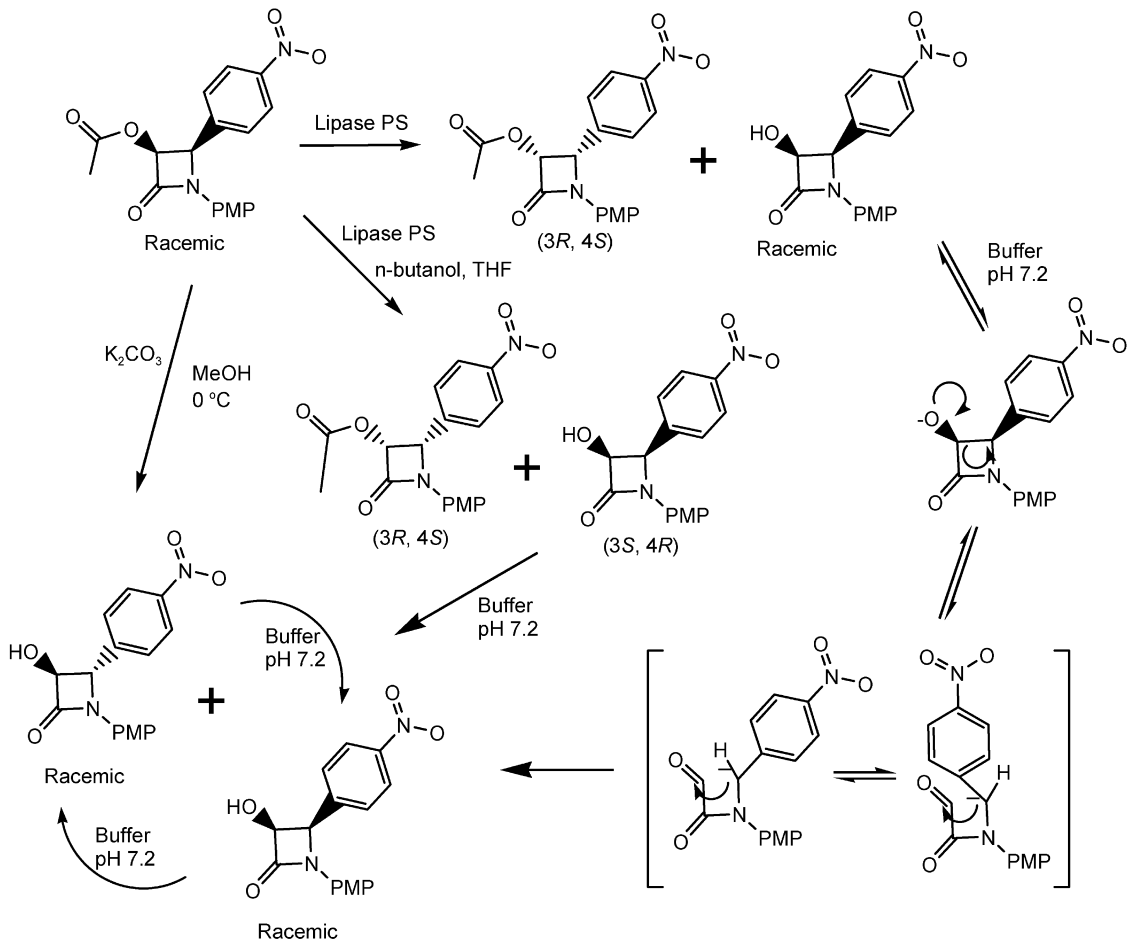


Figure 3. Effect of C-4 substitution on substrate conversion in PS-30 catalyzed hydrolysis.



Scheme 3. Schematic representation of different chemical and enzymatic processes leading to the double epimerization of 3-hydroxy-4-(4-nitrophenyl)azetidin-2-one.

about 32% i.e. much lower than that in compound **13** with a C-4′ chloro substituent.

- The size of a substituent on the C-4 phenyl ring was another important factor that affected substrate conversion. For an EWG at C-4′ the conversion increased with increasing size (**12**<**7**<**21**<**13**<**20**), while for group at C2′ or C-3′ the conversion decreased with increasing size of the substituent (**15**<**17**<**19**). For example, only 21% substrate conversion in 48 h was achieved for compound **19**, with a large iodo group at C-2′, while 35% substrate conversion was observed for compound **15** with chloro, a relatively smaller group, at C-2′. Compounds **15**, **17**, and **19** with chloro, bromo and iodo groups have decreased substrate conversion, which follow the order of their size

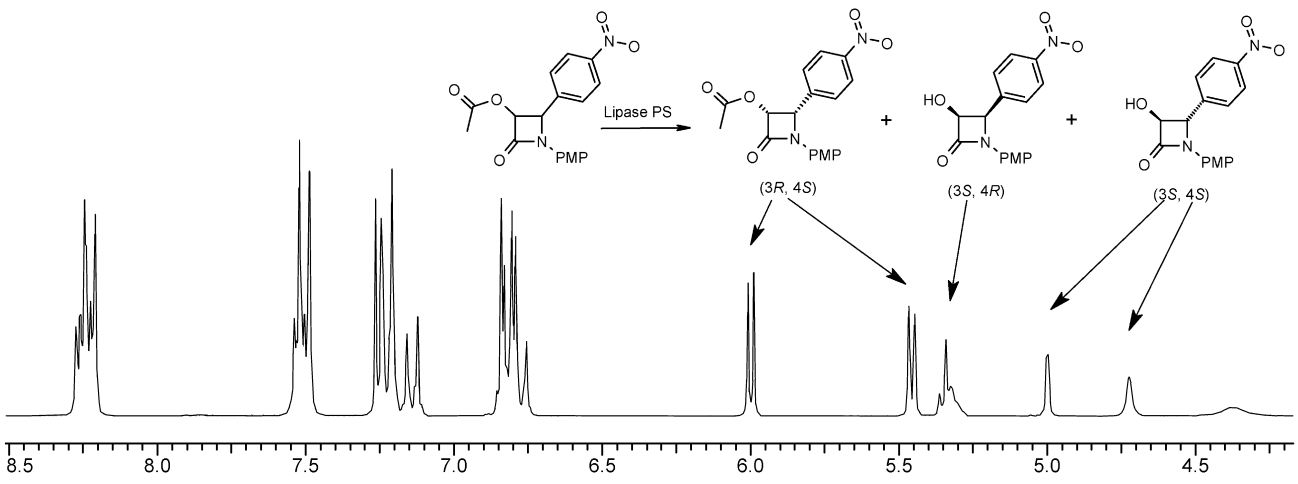


Figure 4. ^{1}H NMR spectrum of the crude product mixture form lipase-catalyzed hydrolysis of 3-acetoxy-1-(4-methoxyphenyl)-4-(4-nitrophenyl)azetidin-2-one (±)-**7**.

(Cl<Br<I, Fig. 3). For compounds with EDGs on the C-4 phenyl ring the substrate conversion decreased as the size of the substituent increased i.e. 4-*tert*-butyl<4-isopropyl<4-*N*,*N*-dimethylamino<4-methoxy<phenyl. The lower substrate conversion in these substrates may be attributed to their steric bulkiness, which could result in van der Waals repulsive interactions in the enzyme active site.

- The dependence of the substrate conversion on the size and electronic nature of the C-4 substituent suggested that steric as well as electronic characteristic of the substrates drive the lipase PS-30 catalyzed hydrolysis.

In compound **7**, analysis of the ^{1}H NMR spectra (Fig. 4) of the reaction mixture at 50% conversion revealed that the product alcohol was a 1:1 racemic mixture of the *cis* and *trans* alcohols. The reaction mixture was subjected to flash column chromatography to obtain the recovered (3*R*,4*S*)-acetate (50%, $[\alpha]_D^{25}=+56°$, %ee=>97), racemic-*cis*-alcohol (25%, $[\alpha]_D^{25}=0°$, %ee=0), and racemic-*trans*-alcohol (25%, $[\alpha]_D^{25}=0°$, %ee=0) (Fig. 2). The formation of the *cis*- and *trans*-alcohols from a *cis*-acetate can only be explained via an unusual ring opening across the lactam C3–C4 bond to a benzylic carbanion (stabilized by the 4′-nitro group) that regenerates the lactam ring by attack on the planar carbonyl group resulting in formation of the racemic *cis*- and *trans*-3-hydroxy-β-lactam derivatives upon double epimerization (Scheme 3).[27] The observation that the recovered β-lactam derivative i.e. (3*R*)-acetoxy-(4*S*)-(4-nitrophenyl) was optically active ($[\alpha]_D^{25}=+112°$) but the hydroxy compounds were not only racemic but a mixture of *cis*- and *trans*-stereoisomers suggested that the ring opening across the C3–C4 bond in the hydroxyl compound might involve a non-enzymatic process. However, it was not obvious if the double epimerization involved an enzymatic process or was just a reaction artifact. To uncover the role of enzyme in the epimerization process, 3-hydroxy-4-(4-nitrophenyl)-1-(4-methoxyphenyl)azetidin-2-one was sought. The compound **7**, however, was stable to hydrolysis in phosphate buffer (pH 7.2, 0.2 M) in the absence of the lipase, indicating that the hydrolysis observed in the presence of lipase indeed was an enzymatic process. Several attempts to obtain the *cis*-hydroxy compound upon chemical hydrolysis of racemic *cis*-**7** (Scheme 3) using K_2CO_3/acetone and K_2CO_3/CH_3OH at room temperature or 0°C led to the formation of the diastereomeric racemic *cis*- and *trans*-alcohols, suggesting that the epimerization of the hydroxy lactam (under basic conditions) was a chemical process. Further, when the *cis*-3-hydroxy-4-(4-nitrophenyl)-1-(4-methoxyphenyl)azetidin-2-one, isolated upon column chromatography from the *cis*-/*trans*-mixture, was stirred in phosphate buffer (pH 7.2, 0.2 M without lipase) it rapidly led to an equilibrium mixture of the *cis*-/*trans*-hydroxy products. To elucidate the role of buffer in this racemization reaction, we removed the buffer from the enzymatic reaction and instead the lipase catalyzed deacetylation of **7** was carried out in dry THF/*n*-butanol. Though deacetylation in THF/*n*-butanol was slow, optically pure *cis*-hydroxy compound ($[\alpha]_D^{25}=-71.2°$) and unreacted optically enriched (+)-*cis*-acetate were isolated after 6 days. The positive and negative sign of the optical rotation for the recovered acetate and the hydroxy product, respectively, established that the enantiopreference of the lipase PS remained unchanged between the buffer and the THF reaction media. The optically pure (−)-*cis*-hydroxy compound was subsequently stirred in phosphate buffer (pH 7.2, 0.2 M) in absence of lipase. The isolated compound was found to be racemic mixture of the *cis*- and *trans*-hydroxyl compounds. The formation of the *trans*- isomer from the *cis*-isomer in absence of the lipase confirmed that the double epimerization was a result of a non-enzymatic reaction and that a free hydroxyl group was required for the double epimerization. In light of these observations, it was concluded that (1) racemization of the product alcohol was an artifact and not a result of the enzymatic catalysis and (2) a non-enzymatic cleavage of the C3–C4 bond followed by ring closure involving double epimerization, it is believed, resulted in formation of a racemic, diastereomeric mixture of the hydroxyl products (Scheme 3).

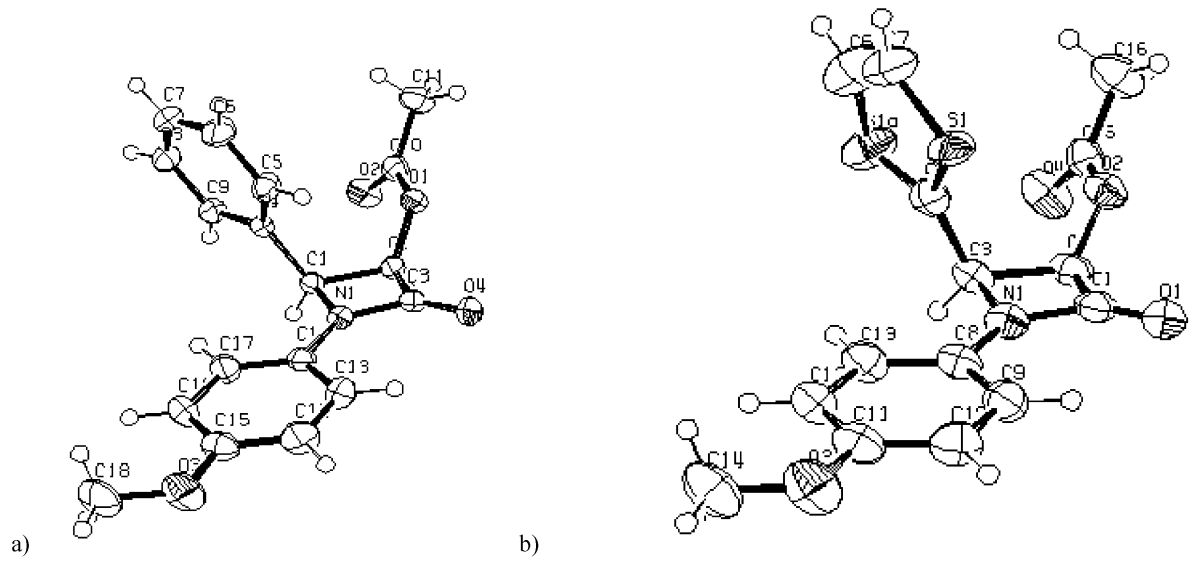


Figure 5. Ortep plot for the X-ray structures (a) of (+)-(3*R*,4*S*)-3-acetoxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one (**1**) and (b) (+)-(3*R*,4*R*)-3-acetoxy-1-(4-methoxyphenyl)-4-thiophen-2-ylazetidin-2-one (**4**).

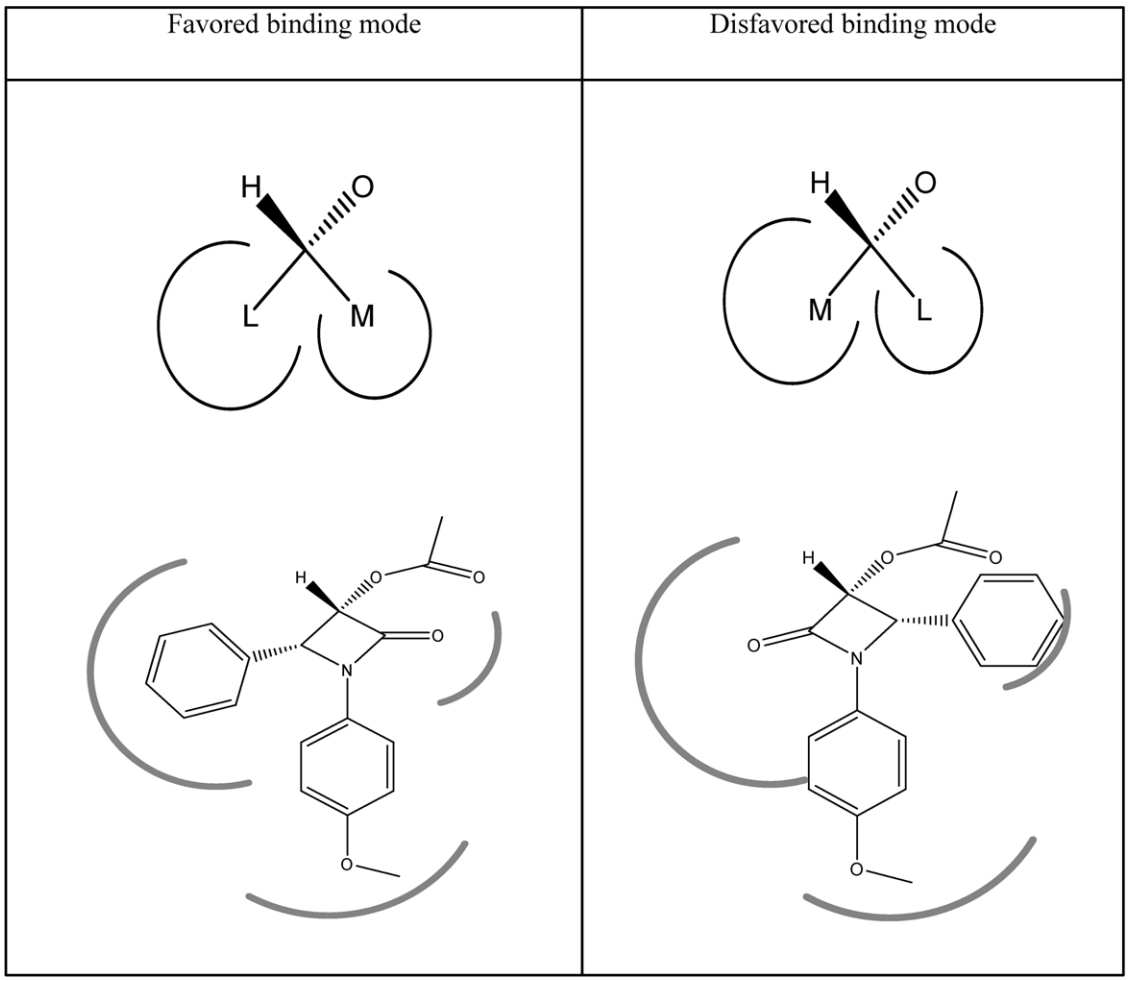


Figure 6. Schematic representations of the orientation of favored (3*S*,4*R*) and disfavored (3*R*,4*S*) enantiomer in active site of the lipase PS-30. M, medium, L, large.

2.3. Determination of absolute configuration

The absolute configuration of the two enantiomers was determined by X-ray crystallography. Single crystals suitable for X-ray crystallographic analysis were selected following examination under a microscope. Single-crystal X-ray diffraction data for the compound were collected on a Bruker-AXS SMART APEX/CCD diffractometer. Diffracted data were corrected for absorption using the SADABS program. Direct methods and the structure solution solved the structures and refinement was based on *F2*. All non-hydrogen atoms were refined with anisotropic displacement parameters whereas hydrogen atoms were placed in calculated positions and given isotropic *U* values based on the atom to which they are bonded. All crystallographic calculations were conducted with the SHELXTL 6.1 program package.

For the crystal of (+)-(3*R*,4*S*)-3-acetoxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one, two molecules were found in each unit cell. The compound crystallizes in a monoclinic space group *P2* (1), with cell dimensions a=5.7617 (8) Å, b=8.1159 (11) Å, c=16.889 (2) Å. A total of 4910 unique reflection data were obtained to give a final *R* index [$I>2\sigma(I)$] of 0.0466 and a flack parameter of 0.0 (9). The results of the structure determination study confirmed the *cis*-(3*R*,4*S*) configuration of the unreacted substrate, (+)-(3*R*,4*S*)-3-acetoxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one (Fig. 5). In the crystal of (+)-(3*R*,4*R*)-3-acetoxy-1-(4-methoxyphenyl)-4-thiophen-2-ylazetidin-2-one, two molecules were found in each unit cell. The compound crystallized in a monoclinic space group *P2* (1), with cell dimensions a=5.3082 (10) Å, b=8.4869 (16) Å, c=17.005 (3) Å. A total of 4638 unique reflection data were obtained to give a final *R* index [$I>2\sigma(I)$] of 0.0618 and flack parameter of 0.19 (16). The results of the structure determination study confirmed the *cis*-(3*R*,4*R*) configuration (Fig. 5).

2.4. Stereopreference of the lipase PS-30

X-Ray crystallographic data (Fig. 5) collected using a single crystal of the unreacted enantiomer of the 4-phenyl (**1**) and 4-thiophen-2-yl (**4**) β-lactam supported the (*S*)-enantiopreference of the *P. cepacia* lipase. The data confirmed (3*R*,4*S*) and (3*R*,4*R*) as the absolute configurations for the unreacted enantiomer of **1** and **4**, respectively. Hence the reacted enantiomers must possess the (3*S*) absolute configurations.

Modeling experiments[28–30] have indicated that *P. cepacia* has a stereochemical preference for the (*R*)-substrate over the (*S*)-substrate but its stereoselectivity seems to be dependent on the chemical nature and/or physical state of

the substrate, as has been observed for other lipases.[31] The stereopreference of the lipase PS-30 for the (3*S*,4*R*) enantiomer was rationalized using a known active site model (Fig. 6) for the lipase as described by Zuegg et al.[28] While the hydrogen at the asymmetric center of both enantiomers is directed towards the same site (referred to as H-alignment), the medium sized group (the carbonyl function) and the large group (the phenyl attached to the C-4 stereogenic center) trade places. This allows the medium-sized group to fit into the smaller hydrophobic pocket in the favored enantiomer whereas the larger-sized group is made to accommodate the smaller hydrophobic pocket in the disfavored enantiomer.

3. Experimental

3.1. Materials and methods

Lipases were obtained from Amano Enzymes (Elgin, IL, USA) and Novozymes (Franklinton, NC, USA). ^{1}H NMR spectra were recorded on a Bruker 250 MHz spectrometer and a Bruker 360 MHz spectrometer in $CDCl_3$ with TMS or CD_3OD as the internal standard. Optical rotations were measured with an AutoPol-IV (Rudolph research analytical) automatic polarimeter. Elemental analysis was performed using a NC-2100 Soil Analyzer, CE Elantech, Inc. Thin-layer chromatography (TLC) was performed on glass plates coated with 0.25 mm thickness of silica-gel. All solvents were distilled prior to use and organic solvent extracts dried over Na_2SO_4.

3.2. X-Ray crystallography

Single-crystal X-ray diffraction data for the compounds **1** and **4** were collected on a Bruker-AXS SMART APEX/CCD diffractometer using a Mo radiation source. Diffracted data were corrected for absorption using the SADABS program. Crystallographic data (excluding structure factors) for the structures in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication numbers CCDC 191837 and 191838. Copies of the data can be obtained, free of charge, on application to CCDC, 12 Union road, Cambridge CB2 1Ez, UK (Fax: +44-1223-336033 or e-mail: deposit@ccdc.cam.ac.uk).

3.3. General procedure for synthesis of racemic 3-acetoxy-4-arylazetidin-2-ones 1–23[32]

A solution of acetoxyacetyl chloride (7.30 mmol) in dry dichloromethane (12 mL) was added drop wise over 1 h at room temperature to a mixture of the appropriately substituted Schiff base (8.76 mmol) and triethylamine (17.52 mmol) in dry dichloromethane (18 mL). The reaction mixture was stirred for an additional hour at room temperature and was then washed with water (2×50 mL). The organic layer was separated and dried over sodium sulfate. Evaporation of the solvent followed by purification by column chromatography (dichloromethane/ether, 9:1) afforded the desired product.

3.3.1. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one (±)-1.[14,32] With the procedure described above **1** was prepared as a white solid, mp 163–165°C (lit.[32] 158–160°C). ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.67 (3H, s, $CH_3C{=}O$), 3.75 (3H, s, CH_3O), 5.34 (1H, d, *J*=4.9 Hz, 4-*H*), 5.94 (1H, d, *J*=4.9 Hz, 3-*H*), 6.78–7.34 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.5, 61.4, 77.3, 114.1, 114.4, 118.8, 127.9, 128.5, 128.8, 130.3, 132.2, 156.6, 161.3, 169.3.

3.3.2. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(2-phenylethyenyl)azetidin-2-one (±)-2.[33] With the procedure described above **2** was prepared as a yellow solid, mp 138–140°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 2.07 (3H, s, $CH_3C{=}O$), 3.77 (3H, s, CH_3O), 4.94 (1H, dd, *J*=4.9, 8.0 Hz, 4-*H*), 5.90 (1H, d, *J*=4.9 Hz, 3-*H*), 6.16 (1H, dd, *J*=8.0, 16.1 Hz, 5-*H*), 6.80 (1H, d, *J*=16.1 Hz, 6-*H*), 6.83–7.43 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 20.3, 55.4, 60.1, 76.3, 114.1, 114.4, 118.7, 121.4, 126.7, 128.7, 130.6, 135.5, 137.1, 156.6, 161.1, 169.5.

3.3.3. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(phenylethynyl)azetidin-2-one (±)-3. With the procedure described above **3** was prepared as a white solid, mp 94–96°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 2.21 (3H, s, $CH_3C{=}O$), 3.80 (3H, s, CH_3O), 5.12 (1H, d, *J*=4.7 Hz, 4-*H*), 5.88 (1H, d, *J*=4.7 Hz, 3-*H*), 6.90–7.54 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 20.3, 51.7, 55.3, 76.7, 81.0, 114.2, 118.7, 121.5, 128.3, 129.1, 130.0, 132.0, 156.6, 161.1, 169.5. Anal. calcd for $C_{20}H_{17}NO_4{\cdot}0.22H_20$: C, 70.78; H, 5.05; N, 4.13. Found: C, 70.92; H, 5.06; N, 3.95.

3.3.4. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-thiophen-2-ylazetidin-2-one (±)-4.[34] With the procedure described above **4** was prepared as a white solid, mp 148–149°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.87 (3H, s, $CH_3C{=}O$), 3.76 (3H, s, CH_3O), 5.62 (1H, d, *J*=4.7 Hz, 4-*H*), 5.96 (1H, d, *J*=4.7 Hz, 3-*H*), 6.80–7.38 (7H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 20.0, 55.4, 57.6, 114.4, 118.8, 126.8, 127.4, 128.4, 130.0, 135.6, 156.7, 161.0, 169.3.

3.3.5. Racemic 3-acetoxy-1, 4-bis-(4-methoxyphenyl)azetidin-2-one (±)-5.[33] With the procedure described above **5** was prepared as a white solid, mp 144–145°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.66 (3H, s, $CH_3C{=}O$), 3.68 (3H, s, CH_3O), 3.73 (3H, s, CH_3O), 5.22 (1H, d, *J*=4.8 Hz, 4-*H*), 5.82 (1H, d, *J*=4.8 Hz, 3-*H*), 6.71–7.22 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.9, 55.4, 61.1, 113.6, 113.9, 114.3, 118.8, 124.0, 129.2, 130.3, 156.5, 159.9, 161.4, 169.3.

3.3.6. Racemic 3-acetoxy-4-(4-methoxycarbonylphenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-6. With the procedure described above **6** was prepared as a white solid, mp 151–152°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.62 (3H, s, $CH_3C{=}O$), 3.68 (3H, s, CH_3O), 3.85 (3H, s, CH_3O), 5.32 (1H, d, *J*=4.9 Hz, 4-*H*), 5.89 (1H, d, *J*=4.9 Hz, 3-*H*), 6.72–7.97 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 52.3, 55.5, 61.1, 76.3, 114.5, 118.7, 127.9, 129.7, 130.0, 130.6, 137.6, 156.7, 160.9, 166.5, 169.2. Anal. calcd for $C_{20}H_{19}NO_6{\cdot}0.82H_2O$: C, 62.52; H, 4.98; N, 3.64. Found: C, 62.64; H, 4.99; N, 3.48.

3.3.7. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(4-nitrophenyl)azetidin-2-one (±)-7.[27] With the procedure

described above **7** was prepared as a yellow solid, mp 162–164°C (lit.[27] 165°C). ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.74 (3H, s, C*H*$_3$C═O), 3.76 (3H, s, C*H*$_3$O), 5.45 (1H, d, *J*=4.9 Hz, 4-*H*), 5.99 (1H, d, *J*=4.9 Hz, 3-*H*), 6.80–8.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.9, 55.5, 60.6, 114.6, 118.6, 123.7, 128.9, 129.6, 140.0, 148.2, 156.9, 160.6, 169.1.

3.3.8. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(3-nitrophenyl)azetidin-2-one (±)-8. With the procedure described above **8** was prepared as a brown solid, mp 89–91°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.73 (3H, s, C*H*$_3$C═O), 3.76 (3H, s, C*H*$_3$O), 5.45 (1H, d, *J*=4.9 Hz, 4-*H*), 5.99 (1H, d, *J*=4.9 Hz, 3-*H*), 6.80–8.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.3, 60.2, 76.3, 113.2, 114.3, 118.6, 122.9, 123.5, 129.6, 133.8, 135.00, 148.3, 156.8, 160.6, 169.0. Anal. calcd for $C_{18}H_{16}N_2O_6{\cdot}0.75H_2O$: C, 58.46; H, 4.36; N, 7.57. Found: C, 58.79; H, 4.39; N, 7.00.

3.3.9. Racemic 3-acetoxy-4-(4-cyanophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-9. With the procedure described above **9** was prepared as a yellow solid, mp 178–180°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.66 (3H, s, C*H*$_3$C═O), 3.69 (3H, s, C*H*$_3$O), 5.32 (1H, d, *J*=4.9 Hz, 4-*H*), 5.90 (1H, d, *J*=4.9 Hz, 3-*H*), 6.73–7.61 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.5, 60.8, 76.4, 112.8, 114.6, 118.2, 118.6, 128.6, 129.7, 132.3, 138.0, 156.9, 160.6, 169.1. Anal. calcd for $C_{19}H_{16}N_2O_4{\cdot}1.6H_2O$: C, 62.26; H, 4.39; N, 7.64. Found: C, 62.83; H, 4.44; N, 6.87.

3.3.10. Racemic 3-acetoxy-4-(4-dimethylaminophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-10. With the procedure described above **10** was prepared as a white solid, mp 198–199°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.67 (3H, s, C*H*$_3$C═O), 2.88 (6H, s, N(C*H*$_3$)$_2$), 3.67 (3H, s, C*H*$_3$O), 5.18 (1H, d, *J*=5.1 Hz, 4-*H*), 5.81 (1H, d, *J*=5.1 Hz, 3-*H*), 6.61–7.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 20.0, 40.6, 55.4, 61.4, 112.9, 114.3, 118.9, 129.0, 130.5, 156.4, 161.6, 169.4. Anal. calcd for $C_{20}H_{22}N_2O_4{\cdot}2.75H_2O$: C, 59.45; H, 5.49; N, 6.93. Found: C, 60.35; H, 5.57; N, 5.71.

3.3.11. Racemic 3-acetoxy-4-(3-fluorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-11. With the procedure described above **11** was prepared as a white solid, mp 166–167°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.67 (3H, s, C*H*$_3$C═O), 3.69 (3H, s, C*H*$_3$O), 5.26 (1H, d, *J*=4.9 Hz, 4-*H*), 5.88 (1H, d, *J*=4.9 Hz, 3-*H*), 6.73–7.22 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.4, 60.8, 76.2, 114.5, 114.7, 115.6, 116.0, 118.7, 123.6, 130.0, 135.0, 135.2, 156.7, 160.7, 161.0, 164.7, 169.2. Anal. calcd for $C_{18}H_{16}FNO_4{\cdot}0.7H_2O$: C, 63.12; H, 4.71; N, 4.09. Found: C, 63.29; H, 4.39; N, 3.98.

3.3.12. Racemic 3-acetoxy-4-(4-fluorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-12.[32] With the procedure described above **12** was prepared as a yellow solid, mp 136°C (lit.[32] 136–137°C). ^{1}H NMR (250 MHz, CD_3OD) δ (ppm): 1.60 (3H, s, C*H*$_3$C═O), 3.62 (3H, s, C*H*$_3$O), 5.39 (1H, d, *J*=4.8 Hz, 4-*H*), 5.84 (1H, d, *J*=4.8 Hz, 3-*H*), 6.73–7.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, CD_3OD): 19.7, 55.9, 62.0, 77.7, 115.2, 115.5, 116.2, 116.5, 120.1, 130.2, 131.4, 158.3, 163.3, 170.5.

3.3.13. Racemic 3-acetoxy-4-(4-chlorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-13. With the procedure described above **13** was prepared as a white solid, mp 172–173°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.67 (3H, s, C*H*$_3$C═O), 3.68 (3H, s, C*H*$_3$O), 5.25 (1H, d, *J*=4.8 Hz, 4-*H*), 5.85 (1H, d, *J*=4.8 Hz, 3-*H*), 6.72–7.28 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 55.4, 58.8, 84.9, 114.4, 118.6, 127.0, 128.9, 129.5, 130.3, 131.1, 133.2, 156.4, 163.6. Anal. calcd for $C_{18}H_{16}ClNO_4{\cdot}1.25H_20$: C, 58.71; H, 4.38; N, 3.80. Found: C, 57.50; H, 4.29; N, 3.35.

3.3.14. Racemic 3-acetoxy-4-(3-chlorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-14. With the procedure described above **14** was prepared as a yellow solid, mp 132–134°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.68 (3H, s, C*H*$_3$C═O), 3.69 (3H, s, C*H*$_3$O), 5.23 (1H, d, *J*=4.9 Hz, 4-*H*), 5.87 (1H, d, *J*=4.9 Hz, 3-*H*), 6.73–7.25 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.4, 60.8, 76.2, 114.2, 114.5, 118.7, 126.1, 127.9, 129.1, 130.0, 134.6, 156.7, 161.0, 169.2. Anal. calcd for $C_{18}H_{16}ClNO_4$: C, 62.52; H, 4.66; N, 4.05. Found: C, 61.23; H, 4.56; N, 3.91.

3.3.15. Racemic 3-acetoxy-4-(2-chlorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-15. With the procedure described above **15** was prepared as a yellow solid, mp 95–97°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.69 (3H, s, C*H*$_3$C═O), 3.69 (3H, s, C*H*$_3$O), 5.71 (1H, d, *J*=5.0 Hz, 4-*H*), 6.09 (1H, d, *J*=5.0 Hz, 3-*H*), 6.74–7.38 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 19.9, 55.4, 58.2, 75.4, 114.5, 118.6, 126.8, 128.7, 129.8, 130.0, 130.2, 133.8, 156.7, 161.4, 168.7. Anal. calcd for $C_{18}H_{16}ClNO_4{\cdot}2.45H_2O$: C, 55.45; H, 4.13; N, 3.59. Found: C, 51.82; H, 4.56; N, 3.56.

3.3.16. Racemic 3-acetoxy-4-(2,4-dichlorophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-16. With the procedure described above **16** was prepared as a white solid, mp 116–118°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.86 (3H, s, C*H*$_3$C═O), 3.75 (3H, s, C*H*$_3$O), 6.07 (1H, d, *J*=5.0 Hz, 4-*H*), 6.21 (1H, d, *J*=5.0 Hz, 3-*H*), 6.80–7.43 (7H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 20.0, 55.4, 57.8, 75.4, 114.5, 118.6, 127.2, 128.9, 129.7, 134.6, 135.2, 156.8, 161.2, 168.7. Anal. calcd for $C_{18}H_{15}Cl_2NO_4$: C, 56.86; H, 3.98; N, 3.68. Found: C, 56.51; H, 3.96; N, 3.57.

3.3.17. Racemic 3-acetoxy-4-(2-bromophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-17.[35] With the procedure described above **17** was prepared as a white solid, mp 79–80°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.77 (3H, s, C*H*$_3$C═O), 3.77 (3H, s, C*H*$_3$O), 5.76 (1H, d, *J*=5.0 Hz, 4-*H*), 6.19 (1H, d, *J*=5.0 Hz, 3-*H*), 6.81–7.64 (8H, m, *Ph*). ^{13}C NMR: 114.2, 114.8, 118.7, 123.7, 127.3, 128.9, 130.0, 131.7, 133.1, 156.7, 161.4, 168.7.

3.3.18. Racemic 3-acetoxy-4-(3-bromophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-18.[36] With the procedure described above **18** was prepared as a yellow solid, mp 150–152°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.75 (3H, s, C*H*$_3$C═O), 3.76 (3H, s, C*H*$_3$O), 5.29 (1H, d, *J*=4.9 Hz, 4-*H*), 5.94 (1H, d, *J*=4.9 Hz, 3-*H*), 6.80–7.46 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.8, 55.4, 60.8, 76.2, 114.5, 118.7, 122.6, 126.6, 130.0, 130.8, 132.1, 134.8, 156.7, 161.0, 169.1.

3.3.19. Racemic 3-acetoxy-4-(2-iodophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-19. With the procedure described above **19** was prepared as a white solid, mp 139–142°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.76 (3H, s, C*H*$_3$C═O), 3.77 (3H, s, C*H*$_3$O), 5.63 (1H, d, *J*=5.0 Hz, 4-*H*), 6.18 (1H, d, *J*=5.0 Hz 3-*H*), 6.82–7.91 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 20.0, 55.5, 65.4, 75.4, 98.8, 114.5, 118.7, 128.2, 128.6, 130.0, 130.3, 134.5, 139.8, 156.7, 161.3, 168.7. Anal. calcd for $C_{18}H_{16}INO_4$: C, 49.45; H, 3.69; N, 3.20. Found: C, 49.37; H, 3.68; N, 3.04.

3.3.20. Racemic 3-acetoxy-4-(4-bromophenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-20. With the procedure described above **20** was prepared as a white solid, mp 178–180°C. ^{1}H NMR (360 MHz, $CDCl_3$) δ (ppm): 1.76 (3H, s, C*H*$_3$C═O), 3.78 (3H, s, C*H*$_3$O), 5.30 (1H, d, *J*=5.0 Hz, 4-*H*), 5.92 (1H, d, *J*=5.0 Hz, 3-*H*), 6.80–7.51 (8H, m, *Ph*). ^{13}C NMR (125 MHz, $CDCl_3$): 19.9, 55.4, 60.9, 76.2, 114.5, 118.7, 122.9, 129.6, 130.0, 131.5, 131.7, 156.7, 161.0, 169.2. Anal. calcd for $C_{18}H_{16}BrNO_4$: C, 55.40; H, 4.13; N, 3.59. Found: C, 55.35; H, 4.07; N, 3.55.

3.3.21. Racemic 3-acetoxy-4-(4-trifluoromethylphenyl)-1-(4-methoxyphenyl)azetidin-2-one (±)-21. With the procedure described above **21** was prepared as a white solid, mp 154–156°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.70 (3H, s, C*H*$_3$C═O), 3.76 (3H, s, C*H*$_3$O), 5.40 (1H, d, *J*=5.0 Hz, 4-*H*), 5.97 (1H, d, *J*=5.0 Hz, 3-*H*), 6.80–7.66 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.7, 55.4, 60.8, 76.3, 114.5, 118.6, 125.4, 128.3, 129.8, 130.7, 136.6, 156.7, 160.8, 169.1. Anal. calcd for $C_{19}H_{16}F_3NO_4$: C, 60.16; H, 4.25; N, 3.69. Found: C, 60.14; H, 4.25; N, 3.74.

3.3.22. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(4-*tert*-butyl phenyl)azetidin-2-one (±)-22. With the procedure described above **22** was prepared as a white solid, mp 175–177°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.22 (9H, s, C(C*H*$_3$)$_3$), 1.56 (3H, s, C*H*$_3$C═O), 3.68 (3H, s, C*H*$_3$O), 5.24 (1H, d, *J*=4.8 Hz, 4-*H*), 5.84 (1H, d, *J*=4.8 Hz, 3-*H*), 6.72–7.29 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.7, 31.2, 34.6, 55.4, 61.2, 114.3, 118.8, 125.3, 127.6, 129.1, 130.3, 151.9, 156.5, 161.3, 169.3. Anal. calcd for $C_{22}H_{25}NO_4$: C, 71.91; H, 6.86; N, 3.81. Found: C, 71.85; H, 6.72; N, 3.74.

3.3.23. Racemic 3-acetoxy-1-(4-methoxyphenyl)-4-(4-isopropylphenyl)azetidin-2-one (±)-23. With the procedure described above **23** was prepared as a white solid, mp 131–133°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.15 (6H, d, *J*=7.0 Hz, CH(C*H*$_3$)$_2$), 1.58 (3H, s, C*H*$_3$C═O), 2.82 (1H, sep., *J*=7.0 Hz, C*H*(CH$_3$)$_3$), 3.68 (3H, s, C*H*$_3$O), 5.24 (1H, d, *J*=4.8 Hz, 4-*H*), 5.84 (1H, d, *J*=4.8 Hz, 3-*H*), 6.72–7.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 19.7, 23.9, 33.8, 55.4, 61.3, 114.3, 118.8, 126.5, 127.9, 129.4, 130.3, 149.6, 156.5, 161.4, 169.2. Anal. calcd for $C_{21}H_{23}NO_4$: C, 71.37; H, 6.56; N, 3.96. Found: C, 71.30; H, 6.44; N, 3.85.

3.4. General procedure for lipase catalyzed hydrolysis of the acetoxy β-lactam

The racemic 3-acetoxy β-lactam (**1**–**23**, 200 mg) was dissolved in acetone (5 mL) and then added to phosphate buffer (25 mL, pH 7.2, 0.2 M) in a 50 mL round bottom flask. To this reaction mixture was added the enzyme catalyst (lipase PS-30, 100 mg) and reaction flask was capped to prevent loss of co-solvent due to evaporation. The reaction mixture was stirred at 25°C and progress of the reaction was monitored via TLC until no further change was detected. The products were isolated by extraction of the reaction mixture with ethyl acetate. The crude product mixture was analyzed by ^{1}H NMR to determine the percent conversion. The hydrolyzed product (the secondary alcohol) and the unreacted *O*-acetate were separated by column chromatography using a stepwise elution of ethyl acetate/petroleum ether (15:85) followed by ethyl acetate/petroleum ether (30:70).

For monitoring substrate conversion with time, reactions were done in 20 mL scintillation vials using 1:1 ratio (w/w) of substrate to the lipase PS-30. After predetermined times, aliquot were withdrawn from the reaction, extracted with ethyl acetate, and the crude reaction product after evaporation of the solvent was analyzed by ^{1}H NMR spectroscopy to determine the substrate conversion.

3.5. Non-enzymatic hydrolysis of racemic *cis*-7

In a 50 mL round bottom flask, 200 mg of the racemic *cis*-**7** was added to a suspension of potassium carbonate (100 mg) in 25 mL of methanol. The reaction mixture was stirred for 15 min at 0°C. The potassium carbonate was filtered off and the crude reaction mixture was isolated upon removal of methanol under reduced pressure. The crude mixture was analyzed from its ^{1}H NMR spectrum, which confirmed the formation of the diastereomeric racemic *cis*- and *trans*-alcohols **7a** and **7b**.

3.6. Enzymatic transesterification of racemic *cis*-7

In a 50 mL round bottom flask, 100 mg of the racemic *cis*-**7** was dissolved in dry THF (10 mL) to which 40 μL of *n*-butanol was added. To this reaction mixture, 100 mg PS-30 was added and the reaction was stirred at 40°C for 6 days. The reaction mixture was filtered through a bed of celite and the crude reaction mixture was analyzed by ^{1}H NMR to determine its percent conversion. The hydrolyzed product (the secondary alcohol, ee>97%, $[\alpha]_D^{25}=-71.2°$) and the enantioenriched *O*-acetate were separated by column chromatography using a stepwise elution of ethyl acetate/petroleum ether (20:80) followed by ethyl acetate/petroleum ether (50:50).

3.7. Determination of the enantiomeric excess

The enantiopurity (the ee values) was determined from the ^{1}H NMR spectrum acquired on a Bruker 250-MHz NMR spectrometer in presence of (+)-[Eu(hfc)$_3$, a chiral shift reagent. The resonance signal of the acetoxy protons, a singlet in absence of the chiral shift reagent, was split into two signals of equal intensity for the two enantiomers in the racemic mixture. The ratio of the intensity of the C*H*$_3$C═O signal in the ^{1}H NMR spectra for the respective enantiomer was used to calculate the % enantiomeric excess. The %ee values for the hydrolyzed alcohols were also obtained using (+)-[Eu(hfc)$_3$ as a chiral shift reagent after their acetylation with Ac_2O/DMAP in dry THF.

3.7.1. 3-Hydroxy-1-(4-methoxyphenyl)-4-phenylazetidin-2-one (−)-1a.[14] With the enzymatic hydrolysis procedure described above, the reaction of racemic **1** afforded **1a** as a white solid, mp 190–192°C (lit.[32] 210–212°C). ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.76 (3H, s, C*H*$_3$O), 5.16 (1H, d, *J*=5.0 Hz, 4-*H*), 5.27 (1H, d, *J*=5.0 Hz, 3-*H*), 6.79–7.45 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 62.2, 77.2, 114.1, 114.4, 118.8, 127.4, 129.0, 129.2, 130.4, 133.1, 156.4, 165.3.

3.7.2. 3-Hydroxy-1-(4-methoxyphenyl)-4-(2-phenylethyenyl)azetidin-2-one (−)-2a.[37] With the enzymatic hydrolysis procedure described above, the reaction of racemic **2** afforded **2a** as a white solid, mp 166–167°C (lit.[37] 157–160°C). ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.74 (3H, s, C*H*$_3$O), 4.38 (1H, bs, O*H*), 4.80 (1H, dd, *J*=5.0, 8.0 Hz, 4-*H*), 5.13 (1H, d, *J*=5.0 Hz, 3-*H*), 6.41 (1H, dd, *J*=8.0, 16.0 Hz, 5-*H*), 6.81–6.84 (1H, d, *J*=16.0 Hz, 6-*H*), 6.76–7.44 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 61.6, 77.2, 113.6, 114.0, 114.3, 114.6, 118.8, 123.0, 126.8, 128.6, 130.8, 135.8, 136.5, 156.4, 166.3.

3.7.3. 3-Hydroxy-1-(4-methoxyphenyl)-4-(phenylethynyl)azetidin-2-one (−)-3a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **3** afforded **3a** as a yellow solid, mp 170–171°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.66 (3H, s, C*H*$_3$O), 4.20 (1H, d, *J*=5.0 Hz, 4-*H*), 4.93 (1H, d, *J*=5.0 Hz, 3-*H*), 5.04 (1H, bs, O*H*), 6.66–7.42 (9H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 51.7, 55.3, 76.7, 81.0, 114.2, 118.7, 121.5, 128.3, 129.1, 130.0, 132.0, 156.6, 165.1. Anal. calcd for $C_{18}H_{15}NO_3 \cdot 0.47H_2O$: C, 71.64; H, 5.01; N, 4.65. Found: C, 71.39; H, 4.99; N, 4.46.

3.7.4. 3-Hydroxy-1-(4-methoxyphenyl)-4-thiophen-2-ylazetidin-2-one (−)-4a.[34] With the enzymatic hydrolysis procedure described above, the reaction of racemic **4** afforded **4a** as a white solid, mp 189–191°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.76 (3H, s, C*H*$_3$O), 5.18 (1H, dd, *J*=5.1 Hz, 4-*H*), 5.51 (1H, d, *J*=5.1 Hz, 3-*H*), 6.80–7.38 (7H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 58.7, 114.4, 118.9, 126.6, 127.3, 127.6, 130.2, 136.5, 156.5, 165.5.

3.7.5. 3-Hydroxy-1,4-bis-(4-methoxyphenyl)-azetidin-2-one (−)5a.[38] With the enzymatic hydrolysis procedure described above, the reaction of racemic **5** afforded **5a** as a white solid, mp 158–159°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.67 (3H, s, C*H*$_3$O), 3.72 (3H, s, C*H*$_3$*O*), 5.07 (1H, d, *J*=5.2 Hz, 4-*H*), 5.13 (1H, d, *J*=5.2 Hz, 3-*H*), 6.69–7.22 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.3, 61.9, 114.3, 114.4, 114.6, 118.9, 124.8, 128.8, 130.5, 156.3, 159.9, 166.0.

3.7.6. 3-Hydroxy-4-(4-methoxycarbonylphenyl)-1-(4-methoxyphenyl)azetidin-2-one (−)-6a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **6** afforded **6a** as a white solid, mp 180–182°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.67 (3H, s, C*H*$_3$O), 3.82 (3H, s, C*H*$_3$O), 5.12 (1H, d, *J*=4.9 Hz, 4-*H*), 5.18 (1H, d, *J*=4.9 Hz, 3-*H*), 6.71–7.97 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 52.1, 55.3, 62.3, 114.3, 118.7, 127.6, 129.7, 129.9, 130.2, 139.1, 156.4, 166.3, 166.9. Anal. calcd for $C_{18}H_{17}NO_5 \cdot 0.96H_2O$: C, 62.75; H, 4.97; N, 4.07. Found: C, 62.03; H, 4.91; N, 3.62.

3.7.7. *cis*-3-Hydroxy-1-(4-methoxyphenyl)-4-(4-nitrophenyl)azetidin-2-one (±)-7a.[27] With the enzymatic hydrolysis procedure described above, the reaction of racemic **7** afforded **7a** as a yellow solid, mp 182–183°C, ($[\alpha]_D^{25}$=0°). ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.65 (3H, s, C*H*$_3$O), 5.22 (1H, d, *J*=5.2 Hz, 4-*H*), 5.26 (1H, d, *J*=5.2 Hz, 3-*H*), 6.64–8.16 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.9, 63.4, 78.6, 115.2, 115.8, 119.9, 124.5, 130.0, 131.7, 143.7, 149.2, 158.1, 167.7.

3.7.8. *trans*-3-Hydroxy-1-(4-methoxyphenyl)-4-(4-nitrophenyl)azetidin-2-one (±)-7b.[27] With the enzymatic hydrolysis procedure described above, the reaction of racemic **7** afforded **7b** as a yellow solid, mp 98–99°C, ($[\alpha]_D^{25}$=0°). ^{1}H NMR (250 MHz, CD_3OD) δ (ppm): 3.60 (3H, s, C*H*$_3$O), 4.52 (1H, d, *J*=1.4 Hz, 4-*H*), 4.91 (1H, d, *J*=1.4 Hz, 3-*H*), 6.69–8.13 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.9, 66.4, 85.2, 115.2, 115.5, 120.3, 125.2, 128.4, 131.3, 145.6, 149.3, 158.2, 167.5.

3.7.9. 3-Hydroxy-1-(4-methoxyphenyl)-4-(3-nitrophenyl) azetidin-2-one (−)-8a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **8** afforded **8a** as a brown solid, mp 151–152°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.65 (3H, s, C*H*$_3$O), 4.70 (1H, bs, O*H*), 5.22 (2H, d, *J*=*5.1* Hz, 3-*H*, 4-*H*), 6.69–8.15 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 61.9, 144.5, 118.8, 122.9, 123.5, 129.7, 133.9, 136.0, 148.3, 156.7, 166.2. Anal. calcd for $C_{16}H_{14}N_2O_5 \cdot 0.37H_2O$: C, 59.88; H, 4.40; N, 8.73. Found: C, 59.99; H, 4.41; N, 8.09.

3.7.10. 4-(4-Cyanophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-9a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **9** afforded **9a** as a white solid, mp 85–88 °C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.69 (3H, s, C*H*$_3$O), 4.17 (1H, bs, O*H*), 5.20 (2H, s, 3-*H*, 4-*H*), 6.72–7.61 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.5, 60.8, 77.2, 112.4, 114.5, 118.4, 118.7, 128.4, 129.8, 132.5, 139.2, 156.8, 165.8. Anal. calcd for $C_{17}H_{14}N_2O_3$: C, 69.38; H, 4.79; N, 9.52. Found: C, 69.25; H, 4.55; N, 9.32.

3.7.11. 3-Hydroxy-4-(4-dimethylaminophenyl)-1-(4-methoxyphenyl)azetidin-2-one (−)-10a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **10** afforded **10a** as a brown solid, mp 187–190°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 2.88 (6H, s, N(C*H*$_3$)$_2$), 3.67 (3H, s, C*H*$_3$O), 5.02 (1H, d, *J*=5.0 Hz, 4-*H*), 5.11 (1H, d, *J*=5.0 Hz, 3-*H*), 6.67–7.25 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 40.5, 55.4, 62.0, 112.9, 114.3, 118.9, 128.4, 130.7, 150.4, 156.2, 165.9. Anal. calcd for $C_{18}H_{20}N_2O_3$: C, 69.21; H, 6.45; N, 8.97. Found: C, 68.98; H, 6.43; N, 8.80.

3.7.12. 4-(3-Fluorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-11a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **11** afforded **11a** as a white solid, mp 217–218°C. ^{1}H NMR (250 MHz, $CDCl_3$/CD_3OD) δ (ppm): 3.67 (3H, s, C*H*$_3$O),

5.08 (1H, d, *J*=5.1 Hz, 4-*H*), 5.12 (1H, d, *J*=5.1 Hz, 3-*H*), 6.71–7.21 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$/CD_3OD): 55.3, 61.1, 76.8, 114.3, 114.7, 115.1, 115.4, 118.7, 123.2, 130.2, 136.3, 136.4, 156.3, 166.4. Anal. calcd for $C_{19}H_{16}FNO_3$: C, 66.89; H, 4.91; N, 4.88. Found: C, 66.43; H, 4.88; N, 4.58.

3.7.13. 4-(4-Fluorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-12a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **12** afforded **12a** as a yellow solid, mp 163–166°C. ^{1}H NMR (250 MHz, $CDCl_3$/CD_3OD) δ (ppm): 3.69 (3H, s, CH_3O), 6.16 (1H, d, *J*=5.1 Hz, 4-*H*), 6.23 (1H, d, *J*=5.1 Hz, 3-*H*), 7.82–8.36 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$/CD_3OD): 55.3, 62.0, 76.6, 114.6, 115.3, 118.7, 129.3, 130.2, 156.3, 160.7, 164.6, 166.5. Anal. calcd for $C_{19}H_{16}FNO_3 \cdot 5.84H_2O$: C, 48.96; H, 3.59; N, 3.57. Found: C, 43.65; H, 3.20; N, 3.02.

3.7.14. 4-(4-Chlorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-13a.[39] With the enzymatic hydrolysis procedure described above, the reaction of racemic **13** afforded **13a** as a white solid, mp 180–181°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.68 (3H, s, CH_3O), 5.13 (1H, d, *J*=5.1 Hz, 4-*H*), 5.16 (1H, d, *J*=5.1 Hz, 3-*H*), 6.71–7.31 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$) δ (ppm): 55.4, 61.9, 114.4, 118.8, 129.0, 129.1, 130.1, 131.8, 134.7, 156.6.

3.7.15. 4-(3-Chlorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-14a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **14** afforded **14a** as a white solid, mp 197–198°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.68 (3H, s, CH_3O), 5.12 (2H, s, 3-*H*, 4-*H*), 6.71–7.28 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 62.0, 77.2, 114.2, 114.4, 118.8, 125.8, 127.7, 129.0, 130.1, 134.9, 135.6, 156.6, 165.8. Anal. calcd for $C_{16}H_{14}ClNO_3 \cdot 0.75H_2O$: C, 60.56; H, 4.45; N, 4.41. Found: C, 60.96; H, 4.48; N, 4.09.

3.7.16. 4-(2-Chlorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-15a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **15** afforded **15a** as a brown solid, mp 214–217°C. ^{1}H NMR (250 MHz, $CDCl_3$/CD_3OD) δ (ppm): 3.69 (3H, s, CH_3O), 5.19 (1H, d, *J*=5.0 Hz, 4-*H*), 5.53 (1H, d, *J*=5.0 Hz, 3-*H*), 6.75–7.38 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$/CD_3OD) δ (ppm): 55.1, 60.2, 114.2, 118.5, 126.6, 128.2, 129.0, 129.3, 130.2, 131.3, 133.1, 156.2, 166.3. Anal. calcd for $C_{16}H_{14}ClNO_3 \cdot 1.13H_2O$: C, 59.30; H, 4.36; N, 3.35. Found: C, 58.30; H, 4.28; N, 3.69.

3.7.17. 4-(2,4-Dichlorophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-16a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **16** afforded **16a** as a brown solid, mp 181–183°C. ^{1}H NMR (250 MHz, $CDCl_3$/CD_3OD) δ (ppm): 3.70 (3H, s, CH_3O), 5.19 (1H, d, *J*=4.6 Hz, 4-*H*), 5.49 (1H, d, *J*=4.6 Hz, 3-*H*), 6.75–7.41 (7H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$/CD_3OD) δ (ppm): 55.3, 59.8, 114.4, 118.5, 127.1, 129.4, 130.1, 130.2, 134.0, 156.5. Anal. calcd for $C_{16}H_{13}Cl_2NO_3$: C, 56.82; H, 3.87; N, 4.14. Found: C, 56.75; H, 3.90; N, 4.05.

3.7.18. 4-(2-Bromophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-17a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **17** afforded **17a** as a white solid, mp 205–206°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.69 (3H, s, CH_3O), 5.32 (1H, d, *J*=5.1 Hz, 4-*H*), 5.56 (1H, d, *J*=5.1 Hz, 3-*H*), 6.74–7.53 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 29.3, 55.1, 62.4, 114.1, 118.5, 127.1, 128.4, 129.3, 130.2, 132.5, 132.8, 156.2, 166.3. Anal. calcd for $C_{16}H_{14}BrNO_3 \cdot 0.23H_2O$: C, 54.54; H, 4.00; N, 3.97. Found: C, 54.71; H, 4.01; N, 3.37.

3.7.19. 4-(3-Bromophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-18a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **18** afforded **18a** as a yellow solid, mp 182–183°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.68 (3H, s, CH_3O), 5.11 (2H, s, 3-*H*, 4-*H*), 6.71–7.44 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 62.0, 77.2, 114.2, 114.5, 118.7, 118.9, 123.0, 126.2, 127.7, 130.1, 130.6, 130.7, 131.9, 135.9, 156.6, 165.9. Anal. calcd for $C_{16}H_{14}BrNO_3$: C, 55.15; H, 4.05; N, 4.02. Found: C, 55.10; H, 4.00; N, 4.05.

3.7.20. 3-Hydroxy-4-(2-iodophenyl)-1-(4-methoxyphenyl)azetidin-2-one (−)-19a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **19** afforded **19a** as a yellow solid, mp 78–80°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 3.69 (3H, s, CH_3O), 5.27 (1H, d, *J*=5.0 Hz, 4*H*), 5.36 (1H, d, *J*=5.0 Hz, 3*H*), 6.73–7.86 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 66.5, 76.9, 98.2, 114.2, 114.5, 118.8, 128.4, 130.1, 130.3, 135.5, 139.7, 156.5, 165.4. Anal. calcd for $C_{16}H_{14}INO_3$: C, 48.63; H, 3.57; N, 3.54. Found: C, 43.65; H, 3.45; N, 3.50.

3.7.21. 4-(4-Bromophenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-20a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **20** afforded **20a** as a pale yellow solid, mp 193–194°C. ^{1}H NMR (360 MHz, $CDCl_3$) δ (ppm): 3.68 (3H, s, CH_3O), 5.05 (2H, s, 3-*H*,4-*H*), 6.62–7.46 (8H, dd, *J*=5.9, 5.5 Hz, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.4, 61.9, 114.5, 118.8, 122.9, 129.3, 130.2, 132.1, 132.4, 156.6, 165.6. Anal. calcd for $C_{16}H_{14}BrNO_3$: C, 55.19; H, 4.05; N, 4.02. Found: C, 55.02; H, 3.97; N, 3.99.

3.7.22. 4-(4-Trifluoromethylphenyl)-3-hydroxy-1-(4-methoxyphenyl)azetidin-2-one (−)-21a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **21** afforded **21a** as a white solid, mp 220–223°C. ^{1}H NMR (250 MHz, $CDCl_3$/CH_3OH) δ (ppm): 3.66 (3H, s, CH_3O), 5.10 (1H, d, *J*=5.0 Hz, 4-*H*), 5.17 (1H, d, *J*=5.0 Hz, 3-*H*), 6.70–7.55 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3$): 55.2, 62.1, 76.8, 114.3, 118.6, 125.3, 128.0, 130.1, 138.0, 156.4, 166.3. Anal. calcd for $C_{17}H_{14}F_3NO_3$: C, 60.54; H, 4.18; N, 4.15. Found: C, 60.32; H, 4.25; N, 4.10.

3.7.23. 3-Hydroxy-4-(4-*tert*-butylphenyl)-1-(4-methoxyphenyl)azetidin-2-one (−)-22a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **22** afforded **22a** as a white solid, mp 180–182°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.30 (9H, s, C(CH_3)$_3$), 3.76 (3H, s, CH_3O), 5.14 (1H, d, *J*=5.0 Hz, 4-*H*), 5.21 (1H, d, *J*=5.0 Hz, 3-*H*), 6.79–7.41 (8H, m, *Ph*).

^{13}C NMR (62.5 MHz, $CDCl_3$): 30.8, 34.2, 55.0, 62.4, 114.0, 118.6, 125.2, 127.1, 130.1, 130.3, 151.0, 156.1. Anal. calcd for $C_{20}H_{23}NO_3$: C, 73.82; H, 7.12; N, 4.30. Found: C, 73.74; H, 7.05; N, 4.19.

3.7.24. 3-Hydroxy-4-(4-isopropylphenyl)-1-(4-methoxyphenyl)azetidin-2-one (−)-23a. With the enzymatic hydrolysis procedure described above, the reaction of racemic **23** afforded **23a** as a white solid, mp 238–240°C. ^{1}H NMR (250 MHz, $CDCl_3$) δ (ppm): 1.17 (6H, d, J=6.9 Hz, $CH(CH_3)_2$), 2.50 (1H, bs, O*H*), 2.83 (1H, sep., J=6.9 Hz, $CH(CH_3)_3$), 3.68 (3H, s, CH_3O), 5.07 (1H, bs, 4-*H*), 5.17 (1H, d, J=5.0 Hz, 3-*H*), 6.71–7.24 (8H, m, *Ph*). ^{13}C NMR (62.5 MHz, $CDCl_3/CH_3OH$): 23.3, 33.5, 54.9, 62.5, 114.0, 118.6, 126.3, 127.3, 130.3, 130.5, 148.7, 156.0, 166.5. Anal. calcd for $C_{19}H_{21}NO_3$: C, 73.29; H, 6.80; N, 4.50. Found: C, 73.15; H, 6.55; N, 4.40.

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