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Enzymatic resolution by CALB of organofluorine compounds under conventional condition and microwave irradiation

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ABSTRACT

Enzymatic kinetic resolution of organofluorine *rac*-alcohols by CALB yielded (−)-(*R*)-2,2,2-trifluoro-1-phenylethanol (**2a**), (−)-(*R*)-1-(3-bromophenyl)-2,2,2-trifluoroethanol (**2b**), (−)-(*R*)-1-(4-bromophenyl)-2,2,2-trifluoroethanol (**2c**), (−)-(*S*)-1-(2,4,5-trifluorophenyl)ethanol (**2d**), (+)-(*S*)-2,2,2-trifluoro-1-phenylethyl acetate (**3a**), (+)-(*S*)-1-(3-bromophenyl)-2,2,2-trifluoroethyl acetate (**3b**), (+)-(*S*)-1-(4-bromophenyl)-2,2,2-trifluoroethyl acetate (**3c**) and (+)-(*R*)-1-(2,4,5-trifluorophenyl)ethyl acetate (**3d**) in high enantiomeric excess (up to >99% *ee*). The reactions were conducted under conventional conditions (orbital shaking) and microwave irradiation in toluene and vinyl acetate as acylating agent. The CALB showed excellent selectivities and good yields in the transesterification of fluorinated aromatic compounds.

1. Introduction

Organofluorine compounds possess unique physical properties that are exploited in a wide range of applications such as agrochemicals, pharmaceuticals, antibacterials, and antifungals. In medicinal chemistry, for example, important physiological properties can be conferred on molecules by adding a fluorine atom that enhances binding selectivity, elevates lipophilicity and improves metabolic stability [1,2].

The synthesis and application of chiral organofluorine compounds for therapeutic purposes have attracted significant attention. For example, the syntheses of drugs used in the treatment of depression, such as fluoxetine (Prozac®) and risperidone (Risperdal®), which is the drug most often used to treat schizophrenia, bipolar disorder and psychotic depression [3]. Certainly, following the synthesis, enantiomeric separations and purity determination are essential for the use of such compounds [4].

Lipase-catalyzed resolution of racemic secondary alcohols by esterification and transesterification in non-aqueous media is a more suitable approach than asymmetric bioreduction of prochiral compounds in aqueous media. Above all, the ready availability of lipases from sources such as plants, bacteria and fungi makes them attractive biocatalysts [5].

In recent years, lipases have been extensively used as biocatalysts for organic synthesis, because of their stability in non-aqueous media, their ability to accept a broad range of substances and the use of mild reaction conditions [6–9]. However, few applications of enzymatic resolution of organofluorine compounds are described in the literature. For example, the chemoenzymatic resolution of racemic trifluoro(aryl)ethanols has been investigated with a lipase from *Pseudomonas aeruginosa* [10,11], and enzymatic synthesis of optically active *gem*-difluorinated allylic alcohols and enantiopure 1,2-bis(hydroxymethyl)-3,3-difluorocyclopropane derivatives with several commercial lipases [12,13].

On the other hand, enantiomerically pure fluorinated aryl alcohols and their esters have been obtained by efficient methodology via nonenzymatic kinetic resolution, using (*R*)-benzotetramisole as catalyst [14].

Microwave irradiation has been reported to be a very attractive tool for organic synthesis. Recent investigations by our group showed that lipase-catalyzed kinetic resolution of (±)-mandelonitrile under microwave irradiation by CALB afforded enhancements in both rate and enantioselectivity [15]. The purpose of this study is to show the kinetic resolution of racemic compounds by lipase from *Candida antarctica* (CALB) under conventional conditions (orbital shaking) and microwave irradiation, to obtain enantiopure and/or enriched organofluorine compounds.

2. Results and discussion

Recently this group reported the esterification of *rac*-mandelonitrile with vinyl acetate as acylating agent in toluene catalyzed

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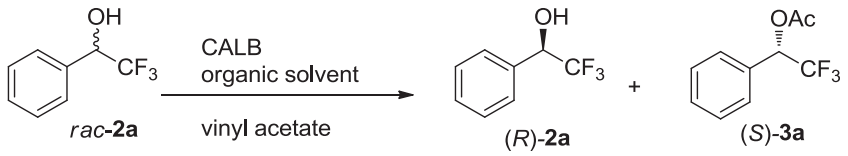
Table 1
Enantioselective acetylation of (±)-2,2,2-trifluoro-1-phenylethanol (**2a**) catalyzed by CALB under various conditions.



Entry	Solvent	Time (h)	*c* (%)[a] **2a**	*ee* (%) **2a** [ac]	*c* (%) **3a**	*ee* (%) **3a** [ac]
Conventional condition (orbital shaker, 32 °C, 130 rpm)						
1	Toluene	48	75	Nd [R]	25	99 [S]
2		96	68	Nd [R]	32	99 [S]
3		120	54	Nd [R]	46	99 [S]
4		168	51(28)[a]	62 [R]	49(18)[a]	99 [S]
5	Dichloromethane	48	96	Nd [R]	4	99 [S]
6	Methanol	48	100	–	Nc	Nc
7	Hexane	24	87	Nd [R]	13	99 [S]
8		48	76	Nd [R]	24	99 [S]
Microwave irradiation (45 °C, 200 W)						
9	Toluene	1	100	–	Nc	Nc
10		2	97	Nd [R]	3	99 [S]
11		4	96	Nd [R]	4	99 [S]
12		5	95	Nd [R]	5	99 [S]
Oil bath (45 °C)						
13	Toluene	2	97	Nd [R]	3	99 [S]
14		4	95	Nd [R]	5	99 [S]
15		24	86	Nd [R]	14	99 [S]
16		48	80	Nd [R]	20	99 [S]

c: concentration of unreacted alcohol determined by GC–FID analysis.
c: conversion to acetate determined by GC–FID analysis.
[a] Isolated yield of (*R*)-**2a**: 28% (62% *ee*) after derivatization with Ac_2O/py and (*S*)-**3a**: 18% (99% *ee*, $E > 200$).
ee: enantiomeric excess.
Nc: reaction did not occur.
Nd: not determined.
ac: absolute configuration.

by CALB under orbital shaker and microwave irradiation. The protocol has proved to be an efficient method for preparing chiral alcohols [15]. In the present study, five aromatic *rac*-alcohols **2a–e** containing fluorine atoms were used.

In order to optimize the conditions for transesterification of (±)-2,2,2-trifluoro-1-phenylethanol (**2a**), various organic solvents were used: toluene, dichloromethane, hexane and methanol (Table 1). Initially, the reactions were carried out in an orbital shaker and catalyzed by CALB.

The best result was obtained in toluene, a hydrophobic solvent, yielding (*S*)-**3a** with excellent optical purity (99% *ee*, $E > 200$) and good conversion (Entry 4, Table 1). Although the reaction showed excellent selectivity and conversion, the reaction time was very long (168 h). Dichloromethane and methanol exhibited a low conversion (0–4%) for (*S*)-**3a** in 48 h (Entries 5 and 6, Table 1). The reaction in hexane showed a similar kinetic resolution (KR) of fluorinated *rac*-**2a** to that with toluene and low conversion at 48 h (Entries 1 and 8, Table 1). Regardless of the time, the reactions gave excellent selectivities for acetate (*S*)-**3a** (99% *ee*, $E > 200$) under conventional conditions (32 °C, 130 rpm).

The transesterification reaction was also carried out in an oil bath with toluene at 45 °C, and yielded (*S*)-**3a** with 20% conversion at 48 h (Entry 16, Table 1). When the reaction was conducted under microwave irradiation in toluene at 45 °C for 5 h, it exhibited a low conversion ($c = 5\%$). This result was somewhat unexpected, since phenylethanol is easily resolved by CALB under MW [16].

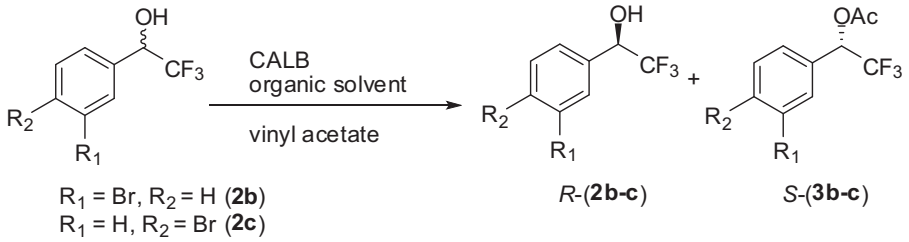
Aiming to extend the enzymatic methodology for KR under orbital shaker and microwave irradiation to organofluoro compounds, the fluoroalcohols **2b–e** were used as substrates. The choice of solvent is important for use in microwave irradiation, as it can influence the enantioselectivity of enzymatic reaction. Toluene has a higher boiling point than hexane, and appears to be an excellent solvent for the investigation of microwave-assisted kinetic resolution [17]. Thus, reactions with organofluoro compounds **2b–d** were carried out in toluene.

As shown by the data in Table 2 for *rac*-**2b**, the time required for kinetic resolution of this compound under microwave irradiation was almost 10 times shorter than the time required under conventional conditions. When the KR was performed on *rac*-**2b** under MW irradiation, (*S*)-1-(3-bromophenyl)-2,2,2-trifluoroethyl acetate (**3b**) was produced with lower optical purity (78% *ee*, $E = 12$), but in a shorter time (14 h) (Entry 14, Table 2). The kinetic resolution of *rac*-**2b** yielded *S*-**3b** with good conversion and selectivity (c 51%, 82% *ee*, $E = 19$) on the orbital shaker. However, the KR took longer (144 h) (Entry 4, Table 2). In this reaction, the unreacted alcohol *R*-**2b** was obtained with good isolated yield (36%) and selectivity (61% *ee*).

A similarly *rac*-**2c** yielded the alcohol *R*-**2c** (c 51%, 70% *ee*) and acetate *S*-**3c** (c 49%, 98% *ee*, $E > 200$) with good conversion and selectivity under MW (Entry 20, Table 2). The effect of raised temperature on the activity and enantioselectivity of CALB under microwave irradiation can be seen, since the reaction was carried out in toluene at 80 °C for *rac*-**2b–c**. The collision rate between the enzyme and substrate molecules increased possibly favoring the formation of enzyme–substrate complex and thus enhancing the reaction rate [18].

The acylation reaction and the enantioselectivity depend on how strong an interaction there is between the aromatic ring of the substrate and the active site of lipase. This interaction is a sum of electronic and steric effects [16]. The KR showed that the activity and selectivity dependent on the position and electronegativity of the substituent group attached to the aromatic ring. Comparing the

Table 2
Enantioselective acetylation of organofluoro alcohols (**2b–c**) by lipase CALB under various conditions.



R_1 = Br, R_2 = H (**2b**)
R_1 = H, R_2 = Br (**2c**)

R-(**2b-c**) *S*-(**3b-c**)

Entry	Time (h)	*c* (%)	*ee* (%) [ac]	*c* (%)	*ee* (%) [ac]
Conventional condition (orbital shaker, 32 °C, 130 rpm)					
#	#	**2b**	**2b**	**3b**	**3b**
1	48	66	31 [*R*]	34	83 [*S*]
2	72	60	40 [*R*]	40	83 [*S*]
3	120	52	55 [*R*]	48	83 [*S*]
4	144	49(36)[a]	61 [*R*]	51(34)[a]	82 [*S*]
#	#	**2c**	**2c**	**3c**	**3c**
5	2	95	4 [*R*]	5	99 [*S*]
6	4	94	6 [*R*]	6	99 [*S*]
7	24	65	42 [*R*]	35	99 [*S*]
8	48	51(20)[a]	72 [*R*]	49(21)[a]	99 [*S*]
Microwave irradiation (80 °C, 200 W)					
#	#	**2b**	**2b**	**3b**	**3b**
9	1	94	5 [*R*]	6	83 [*S*]
10	3	84	22 [*R*]	16	82 [*S*]
11	6	74	22 [*R*]	26	81 [*S*]
12	9	65	32 [*R*]	35	80 [*S*]
13	12	62	38 [*R*]	38	81 [*S*]
14	14	58	43 [*R*]	42	78 [*S*]
#	#	**2c**	**2c**	**3c**	**3c**
15	1	88	12 [*R*]	12	99 [*S*]
16	3	75	27 [*R*]	25	99 [*S*]
17	6	65	43 [*R*]	35	99 [*S*]
18	9	57	57 [*R*]	43	99 [*S*]
19	12	54	66 [*R*]	46	98 [*S*]
20	14	51	70 [*R*]	49	98 [*S*]

c: concentration of unreacted alcohol determined by GC–FID analysis.
c: conversion of acetate determined by GC–FID analysis.
[a] Isolated yield of (*S*)-**3b**: 34% (82% *ee*) and (*S*)-**3c**: 21% (99% *ee*, $E > 200$).
ee: enantiomeric excess.
ac: absolute configuration.

data shown for *rac*-**2b** and *rac*-**2c** (Entries 14 and 20, Table 2), it was observed that the bromo atom in the *para* position relative to the alcohol group in **2c** enabled a better conversion and selectivity in the transesterification of that than the same atom in the *meta* position in **2b**, for the reaction under MW (14 h). In addition, it is important to observe that the *rac*-**2b** showed excellent conversion and enantiomeric excess, however, a longer reaction time was needed under conventional conditions (144 h) (Entry 4, Table 2) than for *rac*-**2c** (48 h) (Entry 8, Table 2).

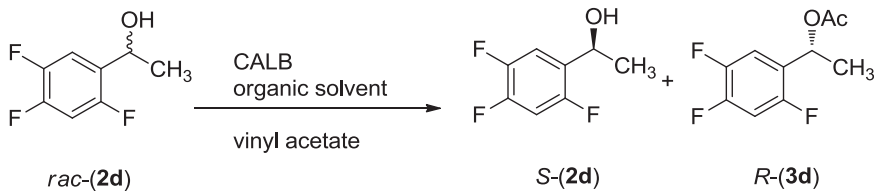
In the case of *rac*-**2d** the halogen substituent has increased electronegativity and there are a large number of fluorine atoms on the aromatic ring. Interestingly, the best result of KR by conventional condition and MW was achieved with *rac*-**2d**. Kinetic resolution of *rac*-**2d** occurred after 30 min under microwave irradiation and the conversion and *ee* remained stable in 8 h of reaction (Entries 7–10, Table 3). Under conventional conditions the reaction was also very fast (2 h) (Entry 1, Table 3).

The presence of three fluorine atoms on the aromatic ring certainly favored a perfect fit of the substrate on the active site of the lipase. Both electronic and steric effects must have promoted the excellent performance of the reaction, providing enantiomerically pure fluorinated products, *S*-**2d** (c = 50%, 99% *ee*) and *R*-**3d** (c = 50%, 99% *ee*, $E > 200$) (Table 5).

The technique of microwave irradiation is very attractive for applications in organic synthesis. Previous studies of lipase-catalyzed kinetic resolution of secondary alcohols under microwave irradiation have in several cases shown enhancements in both rate and/or enantioselectivity [15]. In fact, the temperature has a significant influence on enzyme activity and enantioselectivity. An increase in temperature can reduce mixture viscosity and enhance solubility and diffusion of substrates, thus reducing resistance to mass transfer and favoring interactions between enzyme and substrates that lead to enhancement of enzyme activity and rate of reaction [5]. However, it is also important to observe that the steric and electronic features of the substrates also influence the performance of reactions, as was evident in the results reported here, with under the MW and conventional conditions.

The KR of *rac*-**2e** was also investigated under conventional conditions and MW, but an efficient conversion was not observed. Possibly, the CF_3 group in the *ortho* position relative to the alcohol group may have caused a steric hindrance the reaction occurring

Table 3
Enantioselective acetylation of organofluoro alcohol (**2d**) by lipase CALB under various conditions.



rac-(**2d**) *S*-(**2d**) *R*-(**3d**)

Entry	Time (h)	*c* (%)	*ee* (%) [ac]	c (%)	*ee* (%)[ac]
Conventional condition (orbital shaker, 32 °C, 130 rpm)					
#	#	**2d**	**2d**	**3d**	**3d**
1	2	50	99 [*S*]	50	99 [*R*]
2	4	50	99 [*S*]	50	99 [*R*]
3	24	50	99 [*S*]	50	99 [*R*]
4	48	50(33)[a]	99 [*S*]	50(19)[a]	99 [*R*]
Microwave irradiation (65 °C, 200 W)					
#	#	**2d**	**2d**	**3d**	**3d**
5	0.08	71	37 [*S*]	29	99 [*R*]
6	0.16	58	60 [*S*]	42	99 [*R*]
7	0.33	51	80 [*S*]	49	99 [*R*]
8	0.5	48	90 [*S*]	52	99 [*R*]
9	1	50	99 [*S*]	50	99 [*R*]
10	8	50	99 [*S*]	50	99 [*R*]

c: concentration of unreacted alcohol determined by GC–FID analysis.
c: conversion of acetate determined by GC–FID analysis.
[a] Isolated yield of (*R*)-**3d**: 19% (99% *ee*, $E > 200$).
ee: enantiomeric excess.
ac: absolute configuration.

efficiently. In the literature it is reported that bully substituents in the *ortho* position in the aromatic ring to the reacting group are not resolved by CALB [19].

The absolute configurations of fluoroalcohols (*R*-**2a–c** and *S*-**2d**) were assigned on the basis of their specific rotations which were compared with literature values (Table 5). In addition, these data were confirmed with the empirical Kazlauskas rule. This rule shows the enantiopreference of the esterification of secondary alcohols by lipase and provides suggestions for the absolute configuration of products [20].

The absolute configurations of *S*-**3a–c** and *R*-**3d** acetates were, also suggested by the empirical Kazlauskas rule. According to that rule, lipase stereoselectivity is mainly decided by steric interactions between enzyme and substrate. The small differences in the steric bulk and positions of groups on substrates led to the occurrence or absence of reactions catalyzed by lipase CALB [20].

Kato et al. investigated the effect of the trifluoro-methyl group in lipase-catalyzed reactions of trifluoro(aryl)ethanol derivatives in comparison with the non-fluorinated analogs. Interestingly, the lipases resolved with high enantioselectivity and the rate of acetylation increased remarkably compared to the fluorinated compounds [10]. Similar tendency also was observed in hydrolyses of ethyl 4,4,4-tri-fluoro-3-indole-3-butyrate when was subjected to lipase-catalyzed enantioselective hydrolysis [26]. In this case as well, the bulky and low-affinity trifluoro-methyl group reduced not only the reactivity but also the enantioselectivity of lipases. In studies carried out by us, the *rac*-trifluoro-alcohols containing the trifluoromethyl group were obtained with high selectivity by lipase CALB on MW irradiation.

Bogár and Backvall investigated the metalloenzymatic dynamic kinetic resolution of fluorinated aryl alcohols catalyzed by CALB. In this study, two groups surround the alcohol moiety: one is the

Table 4
Temperature programs used for enantioseparation of the alcohols **2a–e** and acetates **3a–e** on chiral columns by GC–FID analysis.

Compounds*	T_i (°C)	T_f (°C)	t_i (min)	t_f (min)	*r* (°C/min)	T_{MW} (°C)
2a[a]	70	150	3.0	26.67	3.0	45
2b[b]	100	180	1.0	40.00	2.0	80
2c[b]	100	170	1.0	38.33	3.0	80
2d[a]	40	150	1.0	40.67	3.0	65
2e[b]	60	140	3.0	28.67	3.0	80
3a[a]	70	150	3.0	26.67	3.0	45
3b[b]	100	180	1.0	40.00	2.0	80
3c[b]	100	170	1.0	38.33	3.0	80
3d[a]	40	150	1.0	40.67	3.0	65
3e[b]	60	140	3.0	28.67	3.0	80

T_i: initial temperature; T_f: final temperature; t_i: initial time; t_f: final time; *r*: rate; T_{MW}: temperature in microwave irradiation.
[a] Chiral column: CP-CP-7502 CP-Chirasil-Dex CB (25 m × 0.25 mm × 0.39 μm).
[b] Chiral column: CP-7500 CP-Cyclodextrin-β-2,3,6-M-19 (25 m × 0.25 mm × 0.25 μm).
* Enantioseparation: (*R*-**2a**, 22.54 min; *S*-**2a**, 22.54 min), (*R*-**2**, 34.66 min; *S*-**2b**, 34.29 min), (*R*-**2c**, 26.30 min; *S*-**2c**, 26.04 min), (*R*-**2d**, 30.15 min; *S*-**2d**, 32.83 min), (*R*-**2e**, 24.26 min; *S*-**2e**, 24.94 min), (*R*-**3a**, 10.37 min; *S*-**3a**, 10.59 min), (*R*-**3b**, 18.16 min; *S*-**3b**, 18.39 min), (*R*-**3c**, 16.11 min; *S*-**3c**, 16.29 min), (*R*-**3d**, 26.14 min; *S*-**3d**, 25.77 min), and (*R*-**3e**, 22.82 min; *S*-**3e**, 22.82 min).

Table 5
Optical rotations of the organofluorine alcohols **2a–e** and acetates **3a–e** obtained by kinetic resolution with lipase B from *Candida antarctica*.

Compounds	$[\alpha]_D^T$ experimental	$[\alpha]_D^T$ literature
(*R*)-2,2,2-trifluoro-1-phenylethanol (**2a**)	$[\alpha]_D^{26}$ −1.77 (*c* 0.12, $CHCl_3$)	$[\alpha]_D^{25}$ −20.41 (*c* 0.48, $CHCl_3$) [21]
(*R*)-1-(3-bromophenyl)-2,2,2-trifluoroethanol (**2b**)	$[\alpha]_D^{25}$ −13.26 (*c* 0.021, $CHCl_3$)	$[\alpha]_D^{25}$ −25.1 (*c* 2.49, EtOH) [22]
(*R*)-1-(4-bromophenyl)-2,2,2-trifluoroethanol (**2c**)	$[\alpha]_D^{32}$ −16.89 (*c* 0.012, EtOH)	$[\alpha]_D^{20}$ −27.5 (*c* 1.06, EtOH) [23]
(*S*)-1-(2,4,5-trifluorophenyl)ethanol (**2d**)	$[\alpha]_D^{26}$ −1.82 (*c* 1.654, $CHCl_3$)	$[\alpha]_D^{20}$ −29.9 (*c* 0.005, $CHCl_3$) [24]
(*S*)-1-(2-trifluoromethyl)phenyl)ethanol (**2e**)	Nd	$[\alpha]_D^{25}$ −23.0 (MeOH) [25]
(*S*)-2,2,2-trifluoro-1phenylethyl acetate (**3a**)	$[\alpha]_D^{26}$ +3.9 (*c* 0.023, $CHCl_3$)	–
(*S*)-1-(3-bromophenyl)-2,2,2-trifluoroethyl acetate (**3b**)	$[\alpha]_D^{25}$ +178.1 (*c* 0.015, $CHCl_3$)	–
(*S*)-1-(4-bromophenyl)-2,2,2-trifluoroethyl acetate (**3c**)	$[\alpha]_D^{32}$ +78.41 (*c* 0.011, $CHCl_3$)	–
(*R*)-1-(2,4,5-trifluorophenyl)ethyl acetate (**3d**)	$[\alpha]_D^{26}$ +1202 (*c* 0.94, $CHCl_3$)	–
(*R*)-1-(2-(trifluoromethyl)phenyl)ethyl acetate (**3e**)	Nd	$[\alpha]_D^{25}$ +48.5 ($CHCl_3$) [25]

Nd: not determined.

larger fluorinated aryl group and the other is the smaller alkyl group. This composition determined the stereochemical outcome of the product in the lipase-catalyzed transesterification reactions, and the enantioselectivity follows Kazlauskas' rule [27].

In summary, a detailed studies conducted by Soloshonok et al. has shown that the stereochemical outcome of reactions using various fluorinated compounds is strongly influenced by fluoro group. The stereochemical course of the reactions may not only be rationalized by means of steric factors, but also involves the electronic influences fluorine atom, and the nature of the catalyst [28–34].

3. Experimental

3.1. General methods

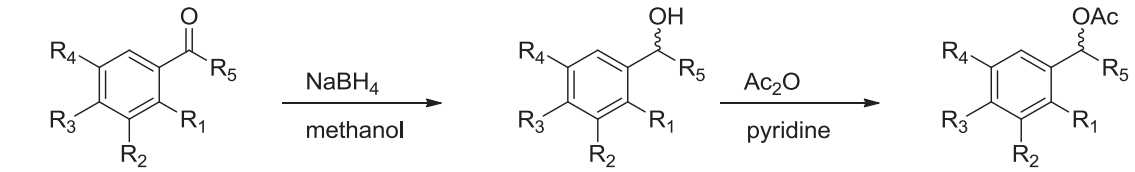
The 2,2,2-trifluoroacetophenone **1a**, 3-bromo-2,2,2-trifluoroacetophenone **1b**, 4-bromo-2,2,2-trifluoacetophenone **1c**, 2,4,5-trifluoroacetophenone **1d** and 2-(trifluoromethyl)acetophenone **1e** were purchased from Sigma–Aldrich. Sodium borohydride and methanol were purchased from Synth and Tedia (Brazil), respectively. Enzymatic kinetic resolution (KR) under conventional conditions (orbital shaking) was carried out in a Tecnal TE-421 orbital shaker. Enzyme reaction products were analyzed with a Shimadzu GC 2010 gas chromatograph equipped with an AOC 20i auto injector and a flame ionization detector (FID). The temperature programs used for the GC–FID analyses of *rac*-alcohols **2a–e** and *rac*-acetates **3a–e** are detailed in Table 4. The injector and detector were maintained at 200 °C, the split ratio of the injector was 1:20 and the carrier gas was N_2 at 60 kPa. The enantiomeric excess (*ee*) of alcohols **2a–e** and acetates **3a–e** were determined by GC–FID analysis (Tables 1–3). The *ee* of unreacted alcohol **2a** was determined after its derivatization with Ac_2O/py by chiral column. All the compounds were separated out by column chromatography (CC) over silica gel (230–400 mesh). The column was eluted with mixtures of hexane/ethyl acetate (9:1 and 8:2) and monitored by TLC, using Sorbent aluminum-backed pre-coated silica gel 60 F254 thin layers.

3.2. Synthesis of (±)-organofluoro alcohols **2a–e**

Racemic secondary alcohols **2a–e** were prepared by reducing ketones **1a–e** with sodium borohydride in methanol. The ketones (**1a**, 7.1 mmol; **1b**, 3.2 mmol; **1c**, 3.2 mmol; **1d**, 3.8 mmol; **1e**, 6.7 mmol), $NaBH_4$ (1.1 equivalents of the quantity of ketone) and methanol (10 mL) were mixed in a 25 mL flask equipped with a magnetic stirrer. The mixtures were stirred for 45 min in on ice bath (Scheme 1). The reactions were then quenched by adding water (1 mL), the methanol was removed by evaporation under vacuum and the residue extracted with ethyl acetate (3× 20 mL). The combined organic phases were dried over anhydrous sodium sulfate (Na_2SO_4) and then filtered. The organic solvent was evaporated under reduced pressure and the residue was purified by silica gel column chromatography, using hexane and ethyl acetate as eluents to produce racemic alcohols **2a–e** in excellent yields (**2a**, 93%; **2b**, 78%; **2c**, 82%; **2d**, 97%; **2e**, 86%).

3.3. Synthesis of (±)-organofluoro acetates **3a–e**

Alcohols (**2a**, 2.19 mmol; **2b**, 0.13 mmol; **2c**, 0.13 mmol; **2d**, 0.22 mmol; **2e**, 0.0005 mmol), pyridine (1.0 mL, 12.41 mmol) and acetic anhydride (Ac_2O) (1.0 mL, 10.49 mmol) were mixed in a 25 mL flask equipped with a magnetic stirrer. The mixture was stirred for 24 h at room temperature (Scheme 1). The reactions were stopped by the addition of 10% HCl (2 mL) and the acetate produced was extracted with ethyl acetate (3× 20 mL). The combined organic phases were dried over Na_2SO_4 and then filtered. The organic solvent was evaporated under reduced pressure and the residue purified by silica gel column chroma-



$R_1 = R_2 = R_3 = R_4 = H$, $R_5 = CF_3$ (**1a**)
$R_1 = R_3 = R_4 = H$, $R_2 = Br$, $R_5 = CF_3$ (**1b**)
$R_1 = R_2 = R_4 = H$, $R_3 = Br$, $R_5 = CF_3$ (**1c**)
$R_1 = R_3 = R_4 = F$, $R_2 = H$, $R_5 = CH_3$ (**1d**)
$R_2 = R_3 = R_4 = H$, $R_1 = CF_3$, $R_5 = CH_3$ (**1e**)

2a (93%)
2b (78%)
2c (82%)
2d (97%)
2e (86%)

3a (69%)
3b(94%)
3c (91%)
3d (82%)
3e (69%)

Scheme 1. Syntheses of racemic alcohols **2a–e** and respective acetates **3a–e**.

tography, using hexane and ethyl acetate as eluents, to give racemic acetates **3a–e** in good to high yields (**3a**, 69%; **3b**, 94%; **3c**, 91%; **3d**, 82%; **3e**, 69%).

3.4. Kinetic resolution of organofluoro alcohols *2a–e*

3.4.1. Lipase-catalyzed acylation under conventional conditions (orbital shaking)

Vinyl acetate (0.5 mL, 5.4 mmol), immobilized lipase (160 mg, 10,000 propyl laurate units per gram) and the alcohols (**2a**, 40 μL, 0.29 mmol; **2b**, 40 μL, 0.26 mmol; **2c**, 40 μL, 0.26 mmol; **2d**, 40 μL, 0.30 mmol; **2e**, 40 μL, 0.26 mmol) were added to 10 mL solvent (toluene, dichloromethane, methanol or hexane) in 50 mL Erlenmeyer flasks (Table 1). The reaction mixture was shaken in a rotary orbital shaker at 32 °C and 130 rpm. The progress of the reaction was monitored by collecting samples (0.1 mL) at to the times indicated in Tables 1–3. The enzymatic kinetic resolution was carried out in triplicate and the mean results presented in Tables 1–3 were obtained by GC–FID analysis. A similar procedure was repeated to obtain the isolated yield of the compounds. In this case, the 50 mL Erlenmeyer reaction flask contained 10 mL of toluene, 0.5 mL of vinyl acetate, 300 mg of lipase CALB and 200 μL of racemic alcohols **2a–e**. After the reaction was completed, the immobilized lipase was filtered off. The toluene was evaporated under reduced pressure. The residue was purified by column chromatography on silica gel, using 8:2 hexane/ethyl acetate as eluent. The enzymatic KR was carried out in duplicate and the mean results presented in Tables 1–3 were obtained by GC–FID analysis of the final mixture.

3.4.2. Lipase-catalyzed acylation under microwave irradiation

The microwave irradiation experiment was performed in a Discover System from CEM Corporation. Toluene (10 mL), vinyl acetate (0.5 mL, 5.4 mmol), lipase (160 mg CALB with 10,000 propyl laurate units per gram) and the alcohols (**2a**, 40 μL, 0.29 mmol; **2b**, 40 μL, 0.26 mmol; **2c**, 40 μL, 0.26 mmol; **2d**, 40 μL, 0.30 mmol; **2e**, 40 μL, 0.26 mmol) were mixed in a 50 mL round-bottom flask. The whole reaction mixture was placed in the microwave apparatus and irradiated for 10 s at 2.45 GHz frequency, and about 200 power. The reaction was monitored by collecting 0.1 mL aliquots at various times, which were analyzed by GC–FID. After the irradiation, the reaction flask was quickly removed and the temperature of the reaction mixture was 45, 65 or 80 °C (Tables 1–3).

3.4.3. Lipase-catalyzed acylation with oil-bath heating

To compare the results obtained by microwave heating with the outcome of a conventionally heated oil bath, the temperature of the reaction was monitored and controlled at 45 °C. Vinyl acetate (0.5 mL, 5.4 mmol), lipase (160 mg, 10,000 propyl laurate units per gram) and the alcohol (**2a**, 40 μL, 0.29 mmol), was added to a 50 mL round-bottom flask. The reaction mixture was stirred by a magnetic and progress was monitored by collecting samples (0.1 mL) at 2, 4, 24 and 48 h (Table 1).

3.5. Absolute configuration

The optical rotation of the purified alcohols **2a–d** and their acetates **3a–d** were measured with a Perkin-Elmer 241 polarimeter (Waltham, MA, USA) and data were collected at the sodium D line (589 nm), with a 1 dm cuvette. The absolute configurations of the alcohols **2a–d** were determined by comparing the measured specific rotations with those reported in the literature (Table 5) [21–25]. Assignments of absolute configurations of organofluoro compounds (**2a–d** and **3a–d**) were confirmed by the empirical Kazlauskas rule [20].

4. Conclusion

In conclusion, in this study the efficient kinetic resolution of organofluoro compounds **2a–d** by immobilized lipase from *C. antarctica* was performed under conventional conditions and microwave irradiation for the first time, producing the enriched fluoroalcohols **2a–d** and fluoroacetates **3a–d** with high selectivity (up to 99% *ee*). It was also observed that electronic and steric effects of substituents, on the aromatic ring of the substrates influenced the performance of the reactions. The reactions under MW occurred in a short time and in good yields and selectivities.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jfluchem.2013.06.014.

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