Tetrahedron: Asymmetry Vol. 4, No. 5, pp. 903-906, 1993
Printed in Great Britain

0957-4166/93 $6.00+.00
Pergamon Press Ltd

Enzymatic Resolution of Aziridine-Carboxylates.

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(*Received* 9 *March* 1993)

Abstract: N-acyl-aziridine-2-carboxylates and N-acyl-aziridine-2,3-dicarboxylates have been resolved with good to excellent stereochemical purity by enzymatic hydrolysis catalyzed by lipase from *Candida cylindracea.*

In recent years, aziridine-2-carboxylates have played an interesting role as chiral synthons in the synthesis of α- and β-aminoacids[1] and β-lactams;[2] the development of new and easy ways of synthesizing optically active aziridine-carboxylates has therefore become a matter of great importance. There are relatively few generally-applicable methods available in the literature,[3] and it was in an attempt to find an alternative to chemical methods that we recently discovered[4] that hydrolases can be efficiently used to resolve three-membered ring heterocycles containing ester groups on the ring carbon atom, such as oxaziridines and N-chloroaziridines. Previous results showed that several hydrolases are quite active in aziridine enantiomer differentiation;[4] among these, α-chymotrypsin and lipases from *Rhizopus delemar* and *Candida cylindracea* display higher enantioselectivity towards N-H and N-substituted aziridine -carboxylates.[5]

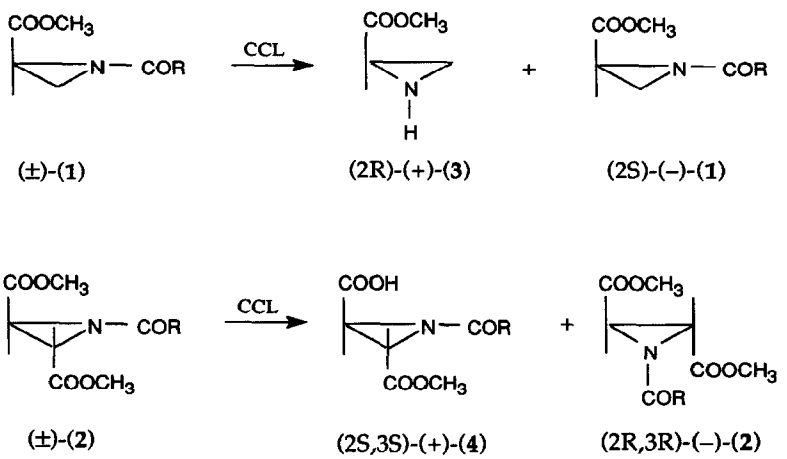
In this paper we report the resolution of racemic N-acyl-2-methoxycarbonylaziridines (**1**) and N-acyl-2,3-bismethoxycarbonylaziridines (**2**) by enantioselective enzymatic hydrolysis catalyzed by lipase from *Candida cylindracea* (CCL).

Racemic aziridines **1** and **2** were prepared by acylation of the corresponding N-H aziridines with acetyl or butyryl chloride, as described in the literature.[6] CCL was purchased from Sigma and used without purification.

Hydrolyses were performed in phosphate buffer (pH 7.5; 0.1 M), standard conditions which we have already used advantageously for the selective resolution of other substrates,[4] and transformations were usually stopped at 60% of conversion. Under these conditions, CCL catalyzed the hydrolysis of the aziridines **1** and **2**

with high enantioselectivity and chemoselectivity; surprisingly, however, it showed a very high esterolytic activity towards substrate **2**, but primarily catalyzed the hydrolysis of the amide bond in compounds **1**. Enzymatic hydrolysis of racemic derivatives **1**, up to 50% conversion, thus afforded the unhydrolyzed esters **1** and the corresponding aziridine **3** as enzymatic hydrolysis product, both with good to excellent stereochemical purity (Table 1, Scheme 1).

Scheme 1



a) R = CH_3; b) R = $n.C_3H_7$

The optically-active unreacted esters **1** and the aziridine **3** were isolated from the aqueous phase by simple extraction with CH_2Cl_2. Owing to its high volatility, aziridine **3** was recovered by conversion into the corresponding N-chloro derivative whose absolute configuration and optical purity are well established.[7] After extraction, the combined organic layers were therefore treated with t-butylhypochlorite[4] at –10 °C, then concentrated and the residue was chromatographed on silica gel (CH_2Cl_2/ethyl ether as eluant). The recovered unchanged aziridines **1a** and **1b** showed 63% and 90% enantiomeric excess (ee.), respectively. It is worth noting that, while higher conversion did not improve the optical purity of **1a**, stopping the reaction at 20% conversion afforded the aziridine **3** in 80% ee.

With respect to compounds **2**, enzyme-catalyzed hydrolysis provided the optically-pure unreacted esters **2** and the corresponding N-acyl-2,3-dicarboxylic acid monomethylesters **4** (Scheme 1). The unhydrolyzed esters **2** were isolated from the reaction mixture by extraction with CH_2Cl_2 followed by column chromatography on

silica gel (CH_2Cl_2/ethyl ether as eluant). The enzymatic-hydrolysis products **4** were recovered from the aqueous phase by acidification with HCl 10% and extraction with ethyl ether. The optical purities of monoesters **4** were estimated through their conversion into the corresponding diesters **2** by esterification with diazomethane. The absolute configuration of aziridines **2** was determined by its conversion into the free NH-diacid by hydrolysis with lithium hydroxide.[8]

Table 1. Enzymatic hydrolysis of aziridines **1** (**a**,**b**) and **2** (**a**,**b**) catalyzed by lipase from *Candida cylindracea.*

Substrate	Reaction condition			unchanged aziridine[a]			
	t/h	E/S[b]	conversion%[c]	yield%	$[\alpha]_D$[d]	ee%[e]	conf.
1a	4	1/4	55	20	–46.1	63	2S[f]
1b	1	1/20	60	35	–74.3	90	2S[g]
2a	24	1/1	60	30	–56.4	≥95	2R,3R[h]
2b	3	1/1	70	25	–34.8	≥95	2R,3R[h]

a) Nmr properties and mass spectra are consistent with the structures considered. b) All hydrolyses were performed in phosphate buffer (pH 7.5; 0.1 M) at room temperature. Reaction mixture defined from the ratio (w/w) enzyme/aziridine (E/S). c) Conversion calculated from the relationship[11] $c = ee_s/ee_s+ee_p$ where ee_s (enantiomeric excess of substrate) refers to the recovered unchanged aziridines and ee_p (enantiomeric excess of product) refers to the hydrolyzed product. The hydrolyzed product **3** showed 40% and 60% ee, from **1a** and **1b** respectively, evaluated as N-chloro derivatives. The monoesters **4** showed 60% and 30% ee, from **2a** and **2b**, respectively. d) Data from chloroform solution. e) ±2%. Optical yield (ee%) determined from ^1H nmr spectra recorded in $CDCl_3$ solution and in presence of $Eu(hfc)_3$. f) from ref. 6. g) Assigned by acylation of the hydrolyzed product (2R)-(+)-**3**.[7] h) assigned by correlation with diethyl aziridine-2,3-carboxylate.[8]

The results reported in Table 1 show that the CCL-catalyzed hydrolysis of aziridines **1** and **2** affords good yield with excellent enantioselectivity. N-acetyl aziridines **1a** and **2a** were hydrolyzed more slowly than the corresponding butyryl **1b** and **2b**; moreover, the change of the N-substituent from acetyl to butyryl in aziridine-2-carboxylates significantly increased enantioselectivity. In our opinion, this is a very easy and efficient way of obtaining both enantiomers of aziridine-carboxylates in good optical and chemical yield. The synthetic potential of these results is strengthened by the nature of the chiral aziridines **1** and **2**: both are

activated towards nucleophilic ring opening reactions and therefore afford easy access to functionalized aminoacid-derivatives.[1,9,10]

Acknowledgements

The authors thank *Ministero dell'Università e della Ricerca Scientifica e Tecnologica*, Rome, for financial support.

References

1. J. Legters, J. G. H. Willems, L. Thijs and B. Zwanenburg, *Rec. Trav. Chim. Pays-Bas*, **1992**, *111*, 59; and references therein.
2. F. H. van der Steen and G. van Koten, *Tetrahedron*, **1991**, *47*, 7503.
3. J. Legters, L. Thijs and B. Zwanenburg, *Rec. Trav. Chim. Pays-Bas*, **1992**, *111*, 1.
4. M. Bucciarelli, A. Forni, I. Moretti and F. Prati, *Tetrahedron: Asymmetry*, **1990**, 5.
5. Results to be submitted for publication.
6. G. V. Shustov, F. D. Polyak, G. K. Kadorkina, I. A. Vosekalna, M. A. Shokhen, R. K. Alekperov, A. V. Eremeev and R. G. Kostyanovsky, *Khim. Geterot. Soedin.*, **1989**, 41.
7. G. V. Shustov, G. K. Kadorkina, R. G. Kostyanovsky and A. Rauk, *J. Am. Chem. Soc.*, **1988**, 1719.
8. J. Legters, L. Thijs and B. Zwanenburg, *Tetrahedron*, **1991**, 5287.
9. D. Tanner and C. Birgersson, *Tetrahedron Lett.*, **1991**, *32*, 2533.
10. J. E. Baldwin, R. M. Adlington, I. A. O'Neil, C. Shofield, A. C. Spivey and J. B. Sweeney, *J. Chem. Soc., Chem. Commun.*, **1989**, 1852 and references cited therein.
11. C. J. Sih and S. H. Wu, *Topics in Stereochemistry*, **1989**, *19*, 63.