Pergamon

Tetrahedron: Asymmetry Vol. 5, No. 1, pp. 9-12, 1994
Elsevier Science Ltd
Printed in Great Britain
0957-4166/94 $6.00+0.00

0957-4166(93)E0031-Q

Enzymatic Asymmetrization of Some Prochiral and Meso Diols through Monoacetylation with Pig Pancreatic Lipase (PPL)

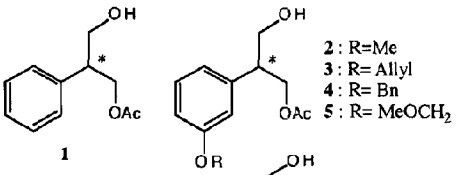
Giuseppe Guanti,* Luca Banfi, and Renata Riva

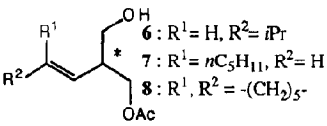
Istituto di Chimica Organica e C.N.R., Centro di Studio per la Chimica dei Composti Cicloalifatici ed Aromatici, corso Europa 26, I-16132 GENOVA (Italy)

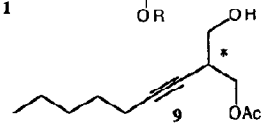
Abstract: Monoacetates **1-8** and **10-11**, derived from asymmetric monoesterification of prochiral or meso diols, have been obtained in good to excellent enantiomeric excess by using inexpensive crude PPL supported on celite, and vinyl acetate as both solvent and acylating agent. Under these conditions reactions are fast and reproducible, and the enzyme can be recycled

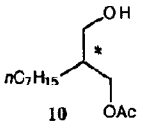
During the last seven years, our group has been interested[1] in the synthetic applications of optically active monoacetates, like **1-11**, obtained through enzyme-mediated hydrolysis of the corresponding prochiral or meso diacetates. Recently many papers have highlighted the usefulness of enzyme-catalyzed transesterification reactions in organic solvents.[2] Thus, in order to find a complementary methodology for the production of **1-11**, we decided to explore the enzyme-catalyzed monoacetylation of the corresponding diols. The main advantage of this approach would be the possibility to obtain both enantiomers of a given monoacetate, by using the hydrolysis or the esterification reactions. Moreover, esterification in organic solvents are often more easily worked-up, and recycling of the enzyme is more simple. On the other hand, the need to use large amounts of catalyst or expensive, highly purified, enzyme preparations, may sometimes hamper the practical application of this methodology. We present here the results of an optimization study which had allowed the obtainment of most of monoacetates 1-11 in high enantiomeric excess, using only small amounts of suitably modified inexpensive crude pig pancreatic lipase (PPL).[3,4]

Our preliminary tests, employing crude PPL, gave unsatisfactory results: the reactions were sluggish, especially for compounds **2-5**, the ees low or not reproducible, and large amounts of enzyme were needed. Therefore we turned our attention to other lipases, mainly those from *Pseudomonas*, which are known to be particularly efficient for enzymatic esterifications in organic solvents.[2,7a] The collected data, shown in Table 1, were rather disappointing. While monoacetate (1R,6S) **11** was obtained in good e.e. using lipase SAM-II, as already reported by Schneider,[6] for the other tested substrates the ees were unsatisfactory. In particular, although there are some literature examples of the efficient use of lipases from *Pseudomonas* in the asym-









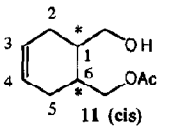


Table 1. Monoacetylation of prochiral or meso diols with various enzymes[a]

Entry	Product	Lipase[b]	Cat. amount (mg/mmol)	Solvent	Temp.	Time	Conversion[c]	Isolated yield	E.e.[d]	Conf.
1	**1**	PS	162	CH_2Cl_2	reflux	21h	58.5%	49%	36.5%	R
2	**1**	PS	161	VA	r.t.	22h	55.6%	56%	22%	R
3	**1**	SAM-II	15.2	$CHCl_3$	r.t.	64h	48%	70%	37%	R
4	**1**	SAM-II	23	MeO*t*Bu	r.t.	24h	55%	70%	36%	R
5	**1**	SAM-II	23	iPr_2O	r.t.	24h	53%	68%	40%	R
6	**1**	SAM-II	24	VA	r.t.	28h	51%	59%	27.5%	R
7	**1**	PSL	3	MeO*t*Bu	r.t.	24h	32%	52%	30%	R
8	**1**	AY	160	VA	r.t.	24h	72.4%	44%	43.2%	R
9	**1**	LCA	42	*n*hexane	0°C	30h	68.6%	43%	36.7%	S
10	**2**	PS	189	CH_2Cl_2	reflux	21h	55.2%	65%	35%	n.d.
11	**3**	LCA	56	*n*hexane	r.t.	1.5h	69.2%	41%	28.7%	n.d.e
12	**6**	SAM-II	18	$CHCl_3$	r.t.	30h	31%	50%	75%	R
13	**9**	SAM-II	10	$CHCl_3$	r.t.	71h	52%	40%	0%	-
14[f]	**11**	SAM-II	56	MeO*t*Bu	r.t.	24h	66%	68%	88%	1R,6S

a) All reactions were carried out on 0.5-1.0 mmol scale using vinyl acetate (VA) (4 equivalents) as acyl donor, with the exception of entries 9 and 11, where a large excess of isopropenyl acetate was used (25% of the overall solvent mixture). b) See note 4. c) See note 5. d) Determined by ^1H n.m.r. in the presence of $Eu(hfc)_3$. e) Configuration was opposite to that of entry 10. f) This reaction was already reported by Schneider (see ref. 6).

metrization of 2-substituted-1,3-propanediols,[7] they do not appear to be good catalysts for the obtainment of our monoacetates (entries 1-7,10,12-13). It is worth noting that lipase from *Candida antarctica*[4] afforded opposite enantioselectivity compared to other lipases (entries 9 and 11).

Thus we return to study the PPL catalyzed reactions, changing the experimental conditions with the aim of improving both rate and enantioselectivity of the processes. Table 2 shows a comparative study carried out on 2-phenyl-1,3-propanediol as model substrate. While reaction in AcOMe under the conditions reported by Didier[3b] was sluggish and required long reaction times (entry 1), the rate was drastically increased using vinyl acetate both as acyl donor and solvent (entry 2),[9] although with a slight decrease of enantioselection. Use of thoroughly dried PPL[10] (entry 3) brought about a slight increase in rate and selectivity. In order to further improve the reaction we then examined PPL supported on celite.[3a,c,e,11] While the catalyst prepared according to Ramos Tombo *et al.*[3a] was found to be by far less efficient than crude PPL itself for this reaction, both in AcOMe and in vinyl acetate (entries 4,5), we found that a 2.5 to 4 fold increase in rate (compare entries 2,3 with entry 7) was achieved by simply liophylising a mixture of crude PPL and celite in pH 7 phosphate buffer[8] according to a similar procedure reported by Carrea *et al.*[11a] Most important, a substantial increase of enantioselection was found. Entries 6 and 9 show that for achieving highest rate and enantioselection it is important not only to use this supported PPL, but also to employ vinyl acetate as the only solvent. The good reaction rate allows to carry out the monoacetylation even at 0°C with moderate catalyst amounts (entries 10-11). At this temperature the yields have been higher, and also a very slight increase in enantioselection was observed. The e.e. is even better than the one achieved in PPL catalyzed hydrolysis of the corresponding diacetate (92%).[1a] Finally it must be stressed that the supported catalyst can be easily recovered and reutilized with minimum loss of activity as demostrated by entry 12.

Having found an efficient procedure for the obtainment of **1**, we decided to apply the same protocol for the production of the other monoacetates **2-11** (Table 3). For comparison, the last column shows the results of PPL catalyzed hydrolysis of the corresponding diacetates.[1] As shown in the table, most of monoacetates **2-11** could be prepared in high e.e. using small amounts of PPL, usually less than the quantity needed for the diacetate hydrolyses. Moreover, the yields were usually better, and,with the exception of compounds **8** and **9**, the ees were comparable or in some cases even superior (see entry 6). In all cases the configuration was op-

Table 2: Monoacetylation of 2-Phenyl-1,3-propanediol with PPL under various conditions[a]

Entry	PPL type[b]	Crude PPL amount[c]	Solvent[d]	Acyl donor[d]	Temp.	Time	Conversion[e]	Isolated yield	E.e.[f]
1	A	762	AcOMe	AcOMe	r.t.	24.5h	46.0%	73%	90.9%
2	A	119	VA	VA	r.t.	5.6h	54.6%	61%	87.9%
3	B	122	VA	VA	r.t.	3.7h	53.5%	76%	90.9%
4	C	152	AcOMe	AcOMe	r.t.	120h	<15%	n.d.	n.d.
5	C	152	VA	VA	r.t.	71h	51.8%	66%	77.5%
6	D	120	CH_2Cl_2	VA	reflux	24h	38.5%	51%	85.1%
7	D	120	VA	VA	r.t.	1.5h	54.6%	69%	94.9%
8	D	120	VA	VA	r.t.	9h	73.4%	43.4%	95.2%
9	D	120	VA/iPr$_2$O 1:1	VA	r.t.	1.1h	53.9%	75%	90.1%
10	D	120	VA	VA	0°C	14.8h	54.3%	72%	**96.6%**
11[g]	D	120	VA	VA	0°C	14.8h	58.4%	78%	**94.2%**
12[g]	E	120	VA	VA	0°C	14.8h	50.2%	89%	**94.3%**

a) All reactions were carried out on 0.4 mmol scale using 5 ml of solvent, in the presence of powdered 3 Å mol. sieves. When vinyl acetate was not used as solvent, 4 eq. were employed. b) A: PPL "straight from the jar". B: PPL dried over P_2O_5 to constant weight. C: PPL fractionated and supported on celite according to note 12 of ref. 3a. D: PPL supported on celite according to note 8; E: the supported enzyme recovered from entry 10 was used. c) mg of starting crude PPL (before supportation or drying) per mmol substrate d) VA = vinyl acetate, IPA = isopropenyl acetate. e) see note 5. f) Determined by ^{1}H n.m.r. in the presence of Eu(hfc)$_3$. Major enantiomer was always R. g) Performed on 2.00 mmol scale.

posite to that produced in PPL catalyzed hydrolysis. The effect of an unsaturation adjacent to the chiral centre is noteworthy (see for examples entries 5 and 6) and parallels the behaviour found out for the corresponding hydrolyses.[12]

From a synthetic point of view the results of entries 1-4,5, 6 and 10 are of particular relevance. Actually **2-5** are useful building blocks for the synthesis of Aklavinone AB ring system,[1b] **6** is a synthetic equivalent of *tris*(hydroxymethyl)methane (THYM*) and has been utilized by us in several synthetic applications,[1a,13] compounds similar to **7** have been utilized in diastereoselective epoxidation reactions,[14] and, finally, **11** was used in alkaloid synthesis.[15]

We wish to thank prof. C. J. Sih and Dr. S. Riva for helpful suggestions. This work was financially as-

Table 3: Acetylation of various diols with supported PPL in vinyl acetate[a]

Entry	Product	Supported PPL amount[b]	PPL amount[c]	Temp.	Time	Conversion[d]	Isolated yield	e.e.[e]	Conf.	ee (yields) of PPL cat. hydrolysis
1	**2**	913	203	0°C	24h	47.6%	71%	96.3%	n.d.	93% (57%)
2	**3**	1041	231	0°C	24h	46.7%	69%	89.5%	n.d.	95% (66%)
3	**4**	1714	381	0°C	24h	52.9%	85%	87.0%	n.d.	96% (43%)
4	**5**	998	222	0°C	24h	52.8%	64%	88.8%	n.d.	50% (46%)
5[f]	**6**	173	38.5	r.t.	2h	54.7%	82%	96.0%	R	97% (75%)
6	**7**	206	45.7	0°C	15h	63.4%	70%	80.0%	S	55% (44%)
7	**8**	211	46.8	0°C	5.6h	55.6%	83%	48%	R	67% (29%)
8	**9**	198	44.0	0°C	22h	55.3%	82%	51%	R	82% (61%)
9	**10**	211.5	47.0	0°C	6h	46.7%	74%	78.0%	R	70% (56%)
10	**11**	211.5	47.0	0°C	23h	50.8%	76%	93.6%	1R,6S	98% (90%)

a) Reactions carried out on 0.4 mmol scale (entries 1-5) or on 0.6 mmol scale (entries 5-10) in 5 ml of vinyl acetate, in the presence of powdered 3 Å mol. sieves. b) mg/mmol substrate. c) calculated mg of purchased crude PPL/mmol substrate. d) see note 5. e) Determined by ^{1}H n.m.r. in the presence of Eu(hfc)$_3$. f) In this case no advantage was observed carrying out the reaction at 0°C. Also in this case the reaction was 2-3 fold faster than with not supported PPL.

sisted by C.N.R., Progetto Finalizzato Chimica Fine, and M.U.R.S.T.

REFERENCES AND NOTES

1. **a)** Guanti, G.; Narisano, E.; Podgorski T.; Thea, S.; Williams, A. *Tetrahedron* **1990**, *46*, 7081-7092 (compound **1**). **b)**. Guanti, G.; Banfi, L.; Brusco, S.; Riva R. *Tetrahedron Lett.* in the press (compounds **2-5**). **c)** Guanti, G.; Banfi, L.; Narisano, E. *J. Org. Chem.* **1992**, *57*, 1540-1554 (compounds **6-10**). **d)** Guanti, G.; Banfi, L.; Narisano, E.; Riva, R.; Thea, S. *Tetrahedron Lett.* **1986**, *27*, 4639-4642 (compound **11**).
2) Recent reviews on enzymatic esterifications in organic solvents: **a)** Chen, C.-S.; Sih, C. J. *Angew. Chem., Int. Ed. Engl.* **1989**, *28*, 695-707. **b)** Klibanov, A. M. *Acc. Chem. Res.* **1990**, *23*, 114-120. **c)** Boland, W.; Frössl, C.; Lorenz, M. *Synthesis* **1991**, 1049-1072. **d)** Xie, Z.-F. *Tetrahedron: Asymm.* **1991**, *2*, 733-750. **e)** Faber, K.; Riva, S. *Synthesis* **1992**, 895-910.
3) Previous works on PPL catalyzed monoacylation of prochiral or meso diols: **a)** Ramos Tombo, G. M.; Schär, H.-P.; Busquets, X. F. I.; Ghisalba, O. *Tetrahedron Lett.* **1986**, *27*, 5707-5710. **b)** Didier, E.; Lubinoux, B.; Ramos Tombo, G. M.; Rihs, G. *Tetrahedron* **1991**, *47*, 4941-4958. **c)** Barnett, C. J.; Wilson, T. M. *Tetrahedron Lett.* **1989**, *46*, 6291-6294. **d)** Mori, K.; Chiba, N. *Justus Liebigs Ann. Chem.* **1989**, 957-962. **e)** Gais, H.-J.; Hemmerle, H.; Kossek, S. *Synthesis* **1992**, 169-173.
4) Crude PPL was purchased from Sigma (50 U/mg of powder, measured using olive oil as standard). Lipase SAM-II (from *Pseudomonas fluorescens*) (42 U/mg) and PSL (lipoprotein lipase from *Pseudomonas sp.*)(2100 U/mg) were purchased from Fluka. Lipases Amano PS (from *Pseudomonas cepacia*)(30 U/mg) and Amano AY (from *Candida*)(30 U/mg) were kindly provided to us by Mitsubishi Italia. LCA (lipase Novozym 435, from *Candida antarctica*)(7 U/mg) was kindly provided by Novo Nordisk.
5) With conversion we mean the percentage of initial hydroxy groups which have been acetylated.
6) Ader, U.; Breitgoff, D.; Klein, P.; Laumen, K. E.; Schneider, M. P. *Tetrahedron Lett.* **1989**, *30*, 1793-1796.
7) **a)** Review: ref. 2d. **b)** Santaniello, E.; Ferraboschi, P.; Grisenti, P. *Tetrahedron Lett.* **1990**, *31*, 5657-5660. **c)** Atsumi, S.; Nakano, M.; Koike, Y.; Tanaka, S.; Ohkubo, M.; Yonezawa, T.; Funabashi, H.; Hashimoto, J.; Morishima, H. *Tetrahedron Lett.* **1990**, *31*, 1601-1604. **d)** Wang, Y.-F.; Wong, C.-H. *J. Org. Chem.* **1988**, *53*, 3129-3130. **e)** Terao, Y.; Akamatsu, M.; Achiwa, K. *Chem. Pharm. Bull.* **1991**, *39*, 823-825. **f)** Terao, Y.; Murata, M.; Achiwa, K.; Nishio, T.; Akamatsu, M.; Kamimura, M. *Tetrahedron Lett.* **1988**, *29*, 5173-5176. **g)** Tsuji, K.; Terao, Y.; Achiwa, K. *Tetrahedron Lett.* **1989**, *30*, 6189-6192. **h)** Ghisalba, O.; Lattmann, R.; Gygax, D. *Recl. Trav. Chim. Pays-Bas* **1991**, *110*, 263-264.
8) 15 g of crude PPL were suspended in 0.067 M pH 7 phosphate buffer (KH_2PO_4-Na_2HPO_4) (250 ml), stirred for 15 min, treated with 50 g of Hyflo Supercel (Fluka), stirred for 15 min., allowed to stand at r.t. for 9h, and lyophilized. The residual solid was triturated in a mortar and further dried under vacuum over P_2O_5 to constant weight. 0.222 g of crude PPL gave 1 g of supported enzyme. This procedure is similar to that reported in ref. 11a, although in that case purified PPL was used.
9) This increase in rate is not obvious, since in other cases the use of vinyl acetate as solvent was not beneficial (see ref. 6 and Herradón, B. *Synlett* **1993**, 108-110).
10) **a)** Therisod, M.; Klibanov, A. M. *J. Am. Chem. Soc.* **1986**, *108*, 5638-5640. **b)** Stokes, T. M.; Oehlschlager, A. C. *Tetrahedron Lett.* **1987**, *28*, 2091-2094. **c)** Janssen, A. J. M.; Klunder, A. J. H.; Zwanenburg, B. *Tetrahedron* **1991**, *47*, 7645-7662.
11) **a)** Secundo, F.; Riva, S.; Carrea, G. *Tetrahedron: Asymm.* **1992**, *3*, 267-280. **b)** Kvittingen, L.; Sjursnes, B.; Halling, P.; Anthonsen, T. *Tetrahedron* **1992**, *48*, 5259-5264; however, in this last paper, no details are given on how the enzyme was supported.
12) A possible explanation for this effect was proposed in ref. 1c. For a recent report regarding the influence of π systems on PPL enantioselectivity see Morgan, B.; Oehlschlager, A. C.; Stokes, T. M. *J. Org. Chem.* **1992**, *57*, 3231-3236.
13) Guanti, G.; Banfi, L.; Riva, R.; Zannetti, M. T. *Tetrahedron Lett.* **1993**, *34*, 5483-5486, and refs. therein.
14) Guanti, G.; Banfi, L.; Merlo, V.; Narisano, E.; Thea, S. *Tetrahedron* **1993**, *49*, 9501-9516.
15) **a)** Riva, R.; Banfi, L.; Danieli, B.; Guanti, G.; Lesma, G.; Palmisano, G. *J. Chem. Soc., Chem. Commun.* **1987**, 299-300; **b)** Danieli, B.; Lesma, G.; Mauro, M.; Palmisano, G.; Passarella, D. *Tetrahedron: Asymm.* **1990**, *1*, 793-800.

(*Received in UK* 11 *October* 1993; *accepted* 2 *December* 1993)