Available online at www.sciencedirect.com
SCIENCE DIRECT
Tetrahedron
Tetrahedron 61 (2005) 3065–3073

Regioselective enzymatic acylation of vicinal diols of steroids

M. Manuel Cruz Silva,[a] Sergio Riva[b] and M. Luisa Sá e Melo[a,*]

[a]*Centro de Estudos Farmacêuticos, Lab. Química Farmacêutica, Faculdade de Farmácia, Universidade de Coimbra, Rua do Norte 3000-295 Coimbra, Portugal*
[b]*Istituto di Chimica del Riconoscimento Molecolare, C.N.R., Via Mario Bianco 9, 20131 Milano, Italy*

Received 28 October 2004; revised 29 November 2004; accepted 25 January 2005

Abstract—Monoacylated derivatives of a complete set of 2,3- and 3,4-vicinal diols of steroids were prepared by regioselective lipase-catalysed transesterification reactions. The enzymes displayed different selectivities towards the vicinal diols depending on the configuration of the hydroxyl groups.
© 2005 Elsevier Ltd. All rights reserved.

1. Introduction

Polyhydroxylated steroids bearing vicinal diols on the A-ring are frequently found in Nature and some of them have relevant biological activities. For instance, the *trans*-diaxial 2β,3α-di-hydroxy pattern is present in natural sulphated sterols with antiviral[1] or anti-angiogenic[2] action, while 2α,3α-diols isolated from marine sources hold cytotoxic activity.[3] Steroidal saponins displaying *trans*-diequatorial 2α,3β-vicinal diols are quite frequent as gitogenin derivatives with antitumor properties.[4] In turn, the 3β,4β-diol functionality is present in a variety of steroids like in the agosterols, which induce reversal of multidrug resistance[5] and proteasome inhibition,[6] as well as in formestane metabolites[7] and in volkendousins, which are potent antitumor agents.[8] Finally, the 3α,4β-vicinal diol pattern has been identified in contignasterol, a natural anti-inflammatory compound[9] and, recently, in a steroid possessing chemotaxis activity.[10]

The discovery that lipases and proteases are able to act in organic solvents opened the way to an intensive synthetic exploitation of these biocatalysts, which, as shown in hundreds of papers and several industrial applications, display remarkable chemo-, regio- and stereoselectivity.[11]

Specifically in the steroids field, enzyme catalysis can play an important role for the mild and selective interconversion of functional groups via regioselective transformations.[12–19] Studies on the transesterification of polyfunctionalyzed steroids have shown that hydrolases can have access to substituents either on the A-ring or on the D-ring and/or on the side-chain of steroids. Several lipases showed a preference for C-3 hydroxyl groups,[13,14] whereas the protease subtilisin Carlsberg catalysed the acylation of C-17 OH.[14] Moreover, stereoselective resolutions of epimeric alcohols located on the steroid side-chain have been carried out by lipase PS[15] and, more recently, by subtilisin.[16]

Concerning the modifications of A-ring substituents, the selectivity of lipases for 3-hydroxysteroids has been applied to the chemoenzymatic synthesis of pharmacologically relevant tibolone metabolites.[17] Quite recently, we have reported a highly selective lipase-catalysed preparation of epimerically pure 5α,6α- and 5β,6β-epoxysteroids through acylation or deacylation reactions at the C-3 OH.[18] Finally, the ability to discriminate among different hydroxyl groups on the A-ring has been demonstrated in the esterification of ecdysteroids catalysed by *Candida antarctica* lipase, which afforded the 2β-monoacyl derivatives in good yields.[19]

In this context, to further explore the enzymatic transformations of steroids, we endeavoured a systematic study on the selectivity of commercially available lipases towards a complete set of stereoisomeric 2,3- and 3,4-vicinal diols, to provide a new tool for the selective transformation of these molecules and of related natural compounds.

2. Results and discussion

Different synthetic strategies were used to afford the

Keywords: Lipases; Steroids; Diols; Regioselectivity; Enzymatic esterification.
* Corresponding author. Tel.: +351 239 859990; fax: +351 239 827030; e-mail: samelo@ci.uc.pt

0040–4020/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2005.01.104

Table 1. Lipase-catalysed monoacylation of vicinal diols[a]

Substrate	Novozym 435	*C. rugosa* lipase	Lipase AY	Lipase PS	Lipase AK	*C. viscosum* lipase	Porcine pancreatic lipase	Lipozyme IM 20	Lipase CE
1	−	++	+	+	+	+	−	−	−
2	−	+	+	++	+	+	−	−	−
3	++	−	−	−	−	−	−	−	−
4	++	+[b]	+[b]	++	−	−	−	−	−
5	+	++	++	+	+	+	−	−	−
6	+	+	+	+	+	++	−	−	−
7	−	+	−	++	+	+	−	−	−
8	−	−	−	−	−	−		−	−

[a] Conversion and product(s) formation was evaluated by TLC.
[b] The formation of two products was observed.

requested stereoisomeric vicinal diols **1**–**8**. Woodward's *cis*-dihydroxylation method, which is known to be selective for the more hindered β-face of 5α-steroids,[20] was applied to Δ^2- or Δ^3-unsaturated precursors affording the *cis*-β diols **1** and **5** (Scheme 2). Complementarily, the *cis*-α diols **3** and **7** were accessed through osmium tetroxide-mediated dihydroxylation on the same olefins.[21] *Trans*-diequatorial diols **2** and **6** were obtained by two different approaches. Starting from cholestan-3-one, α-acetoxylation by lead tetracetate,[22] followed by stereoselective reduction by $NaBH_4/CeCl_3$ and deacetylation rendered the diol **2**. On the other hand, the 3β,4α-diol **6** was directly accessed by hydroboration of the Δ^4-3-one precursor.[23] Finally, *trans*-diaxial diols **4** and **8** were obtained by epoxide opening reactions.

As shown in the formula, the 2,3-diols were prepared by modifying the cholestane skeleton, whereas the difficulties found on separating the 5α- and 5β-epimers of Δ^3-cholestane by sequential crystallizations led us to prepare the set of 3,4-diols in the androstane series. The cholestane derivative **9** was also synthesised to confirm that lipases selectivity towards the A-ring OH was not affected by different substituents on the D-ring.

The performances of a panel of 9 commercial lipases were evaluated for the esterification of the vicinal diols **1**–**9**, using vinyl acetate as the acyl donor and toluene or acetone/THF as solvents for the cholestane or the androstane derivatives, respectively. TLC monitoring allowed the identification, for each substrate, of the lipase(s) able to promote the monoacylation of the substrates. As shown in Table 1, all the stereoisomeric vicinal diols were accepted as substrates by some of the enzymes tested with the exception of compound **8**.

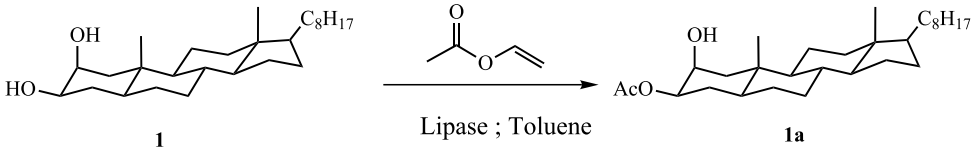
Lipases from different sources (*Candida antarctica*, column 2; *Candida rugosa*, columns 3 and 4; *Pseudomonas* strains, columns 5 and 6; *Chromobacterium viscosum*, column 7) were able to acylate the target compounds. Usually more than one enzyme was acting on the same substrate, with the notable exception of compound **3** (2α,3α-diol) acylated only by Novozym 435 (immobilized lipase B from *Candida antarctica*). Enzymatic acylations were highly regioselective, showing the formation of only one product by TLC, with the exception of *Candida rugosa* lipase (from Sigma or Amano) acting on compound **4**. Noteworthy, lipase PS and Novozym 435 showed complementary regioselectivity towards compound **4**.

For each substrate the best performing lipase (evaluated by TLC) was chosen for scale-up reactions, allowing the isolation of the corresponding monoester in good yields (Scheme 1). Products identification was easily done by NMR analysis (downfield shift of the signals due to the proton geminal to the acylated OH) and, when possible, by comparison with literature data.

The general preference of lipases for the C-3 OH[13,14] was observed with most of the substrates (Scheme 2). Specifically, the diequatorial vicinal diols **2** and **6** were converted into the corresponding 3β-acetate, showing a common preference of different lipases toward a 3β-equatorial OH in the presence of 2α-equatorial OH (substrate **2**) or of 4α-equatorial OH (substrate **6**).

Concerning the diaxial 2β,3α- and 3α,4β-diols (substrates **4** and **8**), different outcomes were noticed. The diaxial 2β,3α-diol was differently accepted by the lipases tested. Whereas Novozym 435 converted this diol exclusively into the 3α-acetate **4a**, lipase PS showed opposite selectivity rendering the 2β-acyl derivative **4b** as the only product. Moreover, acylations catalysed by *Candida rugosa* lipases were not regioselective with this substrate. Conversely, the diaxial 3α,4β diol (**8**) was not accepted by any of the enzymes tested.

Finally, the equatorial/axial 2α,3α-diol (**3**), only accepted by Novozym 435, was acylated at the axial 3α-position, while, at variance, the 3α,4α-dihydroxy steroid (**7**) displaying axial/equatorial configuration, was acylated by different lipases at its equatorial 4α-OH.



1 **1a**

Scheme 1.

Download English Version:

https://daneshyari.com/en/article/5234178

Download Persian Version:

https://daneshyari.com/article/5234178

Daneshyari.com