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Synthesis of Deoxy Sugar Esters: A Chemoenzymatic Stereoselective Approach Affording Deoxy Sugar Derivatives Also in the Form of Aldehyde

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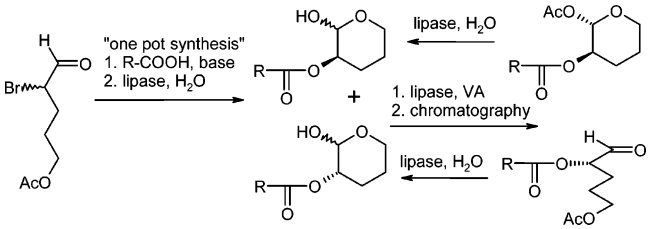
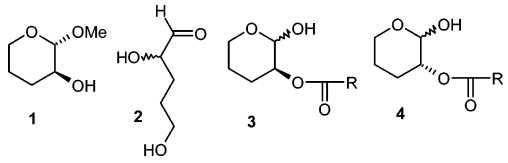


A chemoenzymatic synthesis of deoxy sugar esters is described. The synthesis is based on the *O*-alkylation of carboxylic acid with 2-bromo-5-acetoxypentanal. The method allows treatment of hydroxy carboxylic acids without protection of alcoholic hydroxyl groups. Several stereoisomeric deoxy sugar esters were resolved (up to ee or de > 98%) using a lipase-catalyzed acetylation of hemiacetals that in certain cases afforded deoxy sugar derivatives in the form of aldehydes. The stereochemistry of the reactions was determined by the NMR spectra of mandelic acid derivatives.

Deoxy sugars (DOS) play a significant role in many active compounds of medicines such as antibiotics, antiviral drugs,[1] glycosylation inhibitors,[2] etc. Some of the DOS derivatives have been used as chiral auxiliaries in organic synthesis.[3] Considering the above, the development of diverse strategies for the preparation of DOS derivatives[4] has become an important field of synthetic research.

Several researchers have used methyl glycoside **1** as a source for 3,4-dideoxy ribose (**2**) for inclusion in conjugates. Racemic *trans*-glycoside **1** has been synthesized by the epoxidation of



2,3-dihydropyran in methanol[5] and used in the synthesis of polycyclic ethers: (a) upon a Friedel−Crafts cyclization of 2-*O*-benzyl ethers,[6] (b) by means of a cation-mediated cyclization of the thioglycoside derived from glycoside **1** to afford a ketooxetane,[7] (c) for the preparation of 2,7-dioxabicyclo[4.4.0]-decane and 2,8-dioxabicyclo[5.4.0]undecane.[8]

Derivatives of **2** have been synthesized with high enantiomeric purity starting from L-glutamic acid or D- or L-arabinose.[1,12] The synthesis of (2*S*,3*S*)-2-methoxytetrahydropyran-3-ol[9] (**1**) by bromohydroxylation of 2,3-dihydropyran followed by treatment with LiOH in methanol has been described.[10] In this synthesis, the enantiomers of bromohemiacetal **5** were resolved by lipase-catalyzed acetylation.[11] 3,4-Dideoxy ribose in the form of a glycoside has been included in several conjugates to be used as a chiral auxiliary in an asymmetric modification of the parent structure.[12−14]

For the synthesis of sugar esters, several enzymatic processes have been developed.[15] For the synthesis of hydroxy carboxylic acid esters using routine acylation techniques, the alcoholic hydroxyl groups of the acid have to be protected prior to acylation.[16] For a chemoselective esterification of unprotected hydroxy carboxylic (phenolic) acids the Mitsunobu reaction has been used.[17]

The aim of the present work was to develop a synthetic approach for the preparation of stereochemically pure 3,4-

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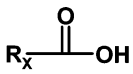
TABLE 1. Structures of the Carboxylic Acids Used



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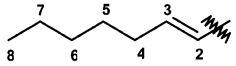
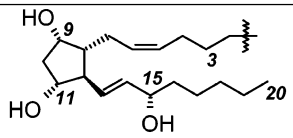
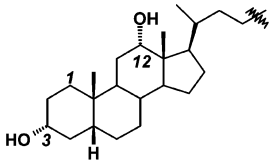
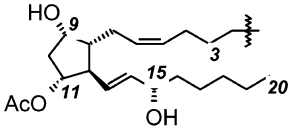
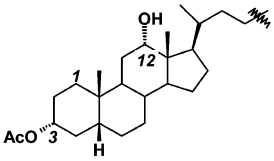
1	
2	
3	
4	
2a	
3a	

dideoxy ribose esters of general formulas **3** and **4** to be used as building blocks in the synthesis of putative medicines. The hemiacetal (and especially the aldehyde group) of the deoxy sugar enables the linking of the derivative to a proper functional group of another synthetic building block.

A chemoenzymatic synthesis of the 3,4-dideoxy ribose esters of selected carboxylic acids (Table 1) was performed to test the chemoselectivity and stereoselectivity of the novel procedure.

We started from (*E*)-2-octenoic acid (OA) as a nonchiral compound to investigate a kinetic resolution of 3,4-dideoxy ribose ester enantiomers upon the lipase-catalyzed acetylation of the hemiacetal group. The inclusion of (*S*)-mandelic acid (MA) in the test set was necessary for NMR-based stereochemical studies.[16,18] Deoxycholic acid (DCA) was chosen to test the limitations of lipase-catalyzed kinetic resolution of the diastereomers of sterically demanding hemiacetal products. We also bore in mind that the glycoconjugates of bile acids are of interest in several cases.[19] Prostaglandin $F_{2\alpha}$($PGF_{2\alpha}$) is a carboxylic acid with a large flexible hydrocarbon skeleton. Despite their complex structure, several prostanoids have been smoothly esterified or acylated by lipases.[20] In general, prostaglandins enjoy diverse clinical applications, reflecting their wide-ranging physiological and pharmacological properties.

Bromoaldehyde **6** (Scheme 1) was synthesized by the incubation of racemic bromohemiacetal **5** with Novozym 435 (*Candida antarctica* lipase B (CALB) immobilized on an acrylic resin) in a chloroform/vinyl acetate (VA) mixture at rt.[10] Under these conditions, the conversion of the starting material was >98%, and two chemically different products were formed. After chromatography the yield of the *trans*-acetyl bromohemiacetal (acetic acid (2*S*,3*R*)-3-bromo-tetrahydropyran-2-yl ester) was 50%, and that of aldehyde **6**, 40%. Both products were, somewhat unexpectedly, gained with a low (<50%) enantiomeric excess upon the prolonged process, while the faster process reported earlier afforded the above *trans*-acetyl bromohemiacetal with a higher ee.[10] However, we found that starting from bromoaldehyde **6** of low ee and resolving the stereoisomers of target compounds are advantageous in some cases, allowing the preparation of both the individual enantiomers or diastereomers of the DOS esters.

The stereoisomeric mixtures of DOS esters were synthesized using *O*-alkylation of carboxylic acids with bromoaldehyde **6** followed by lipase-catalyzed deacetylation (Scheme 1).

The *O*-alkylation of OA afforded ester **7**, to the same reaction mixture were added water and the CALB, resulting in hemiacetals **8**. In this "one-pot synthesis" the latter step includes a selective lipase-catalyzed deacetylation of the terminal hydroxyl group, affording hydroxyaldehyde, which spontaneously transformed to a hemiacetal as a thermodynamically more stable form. The procedure afforded **8** as a mixture of four stereoisomers (because of the anomers) in a near quantitative yield.

The *O*-alkylation of the carboxyl group of $PGF_{2\alpha}$ resulted in the formation of diastereomeric esters **9**, actually existing as a complex mixture of products, very probably intramolecular macrocyclic hemiacetals (visible as four spots on TLC). The following lipase-catalyzed deacetylation selectively cleaved the acetyl group and afforded a mixture of four hemiacetal diastereomers **10** (inseparable on silica gel) in less than 18 h. The process triggered by the lipase-catalyzed deacetylation evidently involves also the decyclization of macrocyclic hemiacetals and the formation of more stable six-membered hemiacetals as cascade reactions. This five-step reaction sequence, including also the former reactions—the *O*-alkylation followed by spontaneous acetalizations, all taking place as a "one-pot synthesis"—is a reliable way to hemiacetals **10** (overall yield 60%).

The *O*-alkylation of DCA afforded ester **11**, and the acetoxy group was selectively hydrolyzed by adding the CALB and H_2O to the same reaction mixture. This resulted in the formation of ester **12** consisting of four stereoisomers inseparable on silica.

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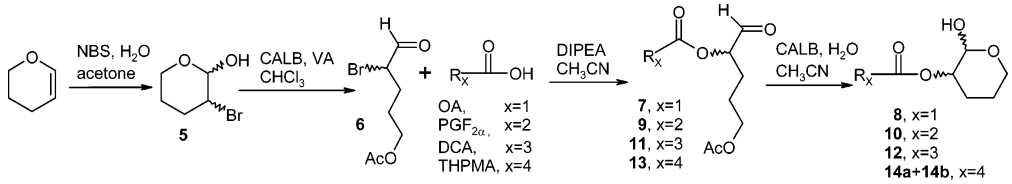
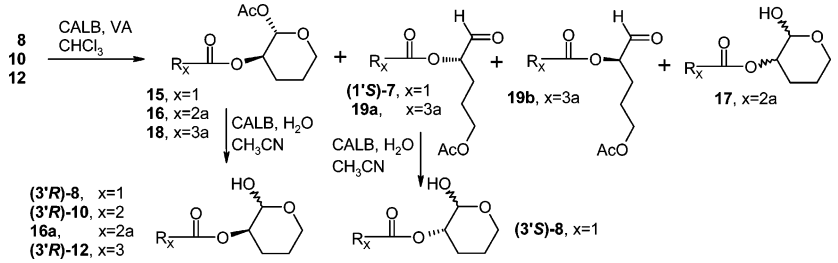
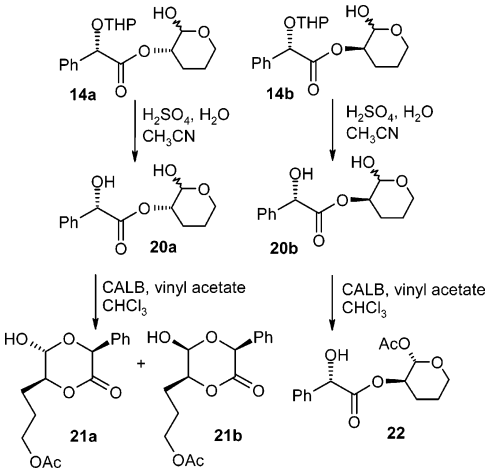
SCHEME 1. Synthesis of Esters 8, 10, 12, 14a and 14b



SCHEME 2. Synthesis of Deoxy Sugar Esters (3′S)-8, (3′R)-8, (3′R)-10, and (3′R)-12



SCHEME 3. Synthesis of Mandelic Acid 3,4-Dideoxy Ribose Esters



The THP-protected MA was *O*-alkylated to afford ester **13** followed by lipase-catalyzed deacetylation to afford hemiacetals **14a** and **14b** in a near quantitative yield.

The synthesis of stereochemically pure DOS esters of OA, $PGF_{2\alpha}$, and DCA using lipase-catalyzed resolution of the stereoisomeric hemiacetals is described in the Scheme 2.

A kinetic resolution of **8** by the lipase-catalyzed acetylation resulted in the simultaneous formation of two chemically different compounds—hemiacetal **8** with 3′*S* configuration afforded aldehyde **(1′*S*)-7**, whereas hemiacetal **8** with 3′*R* configuration afforded acetylated hemiacetal **15** that were separated by chromatography over silica. The stereochemistry of the CALB-catalyzed acetylation of hemiacetals was determined by the NMR assignment of diastereomeric MA 3,4-dideoxy ribose esters (Scheme 3). We found it to be in accordance with the Kazlauskas rule.[21] Also, the exclusive formation of only the *trans*-isomer of acetylated hemiacetal was observed. The subsequent deacetylation of individual products **(1′*S*)-7** and **15** catalyzed by CALB gave the desired, deoxy sugar esters of OA **(3′*S*)-8** and **(3′*R*)-8** in an almost quantitative yield. The enantiomeric excess of the target ester stereoisomers (both of them existing as an equilibrium mixture of anomers) was high.

For the resolution of $PGF_{2\alpha}$ deoxy sugar ester diastereomers **10** the lipase-catalyzed acetylation afforded acetylated hemiacetal **16** as a major product separated by column chromatography on silica. It was gained as an individual compound, whereas **17** (15% of the material gained) was a mixture of several isomers. In esters **16** and **17** the hydroxyl group at the C_{11} of the $PGF_{2\alpha}$ skeleton was acetylated as well. The lipase-catalyzed deacetylation of **16** gave (under the nonoptimized reaction conditions) stereochemically pure deoxy sugar esters **(3′*R*)-10** and **16a** with the ratio of 1:1.4.

The enzymatic acetylation of DCA ester **12** afforded acetylated hemiacetal **18** (38%) as a pure stereoisomer after flash chromatography. The formation of aldehyde **19** in a significant quantity (46% of the separated material) as a mixture of diastereomers was observed. The low stereoselectivity of the lipase-catalyzed decyclization and/or racemization of the α-substituted aldehyde could be assumed. Nevertheless, the result revealed an interesting ability of lipase to produce 3,4-dideoxy ribose bile acid conjugates in the form of a chemically rather active aldehyde starting from less active hemiacetals. In esters **18** and **19**, the hydroxyl group at the C_3 of the DCA skeleton was acetylated as well. The enzymatic deacetylation of acetylated hemiacetal **18** afforded the desired homogeneous deoxy sugar ester **(3′*R*)-12**.

The separation of MA deoxy sugar esters **14a** and **14b** (Scheme 3), both being mixtures of four stereoisomers, proceeded in accordance with an absolute configuration at the C_2 of the deoxy sugar moiety; the diastereomers related to the stereogenic center of the THP protecting group were inseparable on silica gel. Afterward, the THP protecting group was cleaved with dilute H_2SO_4 to afford, after flash chromatography of the products, pure mandelates **20a** and **20b**, respectively. The absolute configurational assignment of mandelates **20a** and **20b** was based on the differential shielding effects caused by the phenyl group in NMR spectra. The assignment was confirmed by the results of their acetylation catalyzed by CALB, which occurred in accordance with the Kazlauskas rule. A separate acetylation of individual hemiacetals **20a** and **20b** exclusively afforded chemically different products. Diastereomer **20b** afforded a pure (>99%) *trans* acetylated hemiacetal **22** when diastereomer **20a,** unfavored in the CALB configuration,

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TABLE 2. Yields and Stereoisomeric Purities of the Products

cmpd	yield (%)	ee or de	cmpd	yield (%)	ee or de	cmpd	yield (%)	ee or de
5	90	rac	**12**	66	low	**18**	38	>98
6	40	<50	**(3′*R*)-12**	71	>98	**19a**+**19b**	46	low
(1′*S*)-7	32	>98	**14a**	43	mix[a]	**20a**	85	>98
8	94	nd[b]	**14b**	37	mix[a]	**20b**	86	>98
(3′*R*)-8	92	>92	**15**	48	>98	**21a**	92	nd
(3′*S*)-8	94	>86	**16**	52	>98	**22**	85	>98
10	60	low	**16a**	49	>98			
(3′*R*)-10	36	>98	**17**	15	low			

[a] A complex mixture of diastereomers. [b] Not determined.

underwent a novel three-step cascade of the reactions. The cascade started with the lipase-catalyzed decyclization of the hemiacetal followed immediately by the acetylation of the terminal hydroxyl group. Thereafter, the MA ester of the extended-chain deoxy sugar acetate underwent a spontaneous cyclization, affording thermodynamically preferable cyclic hemiacetal **21a** together with a low amount of isomeric **21b**. The cascade process consuming **20a** was at least 10 times slower than that consuming mandelate **20b**, thus offering a possibility for a kinetic resolution as well (Table 2).

In summary, a novel approach for the synthesis of stereochemically pure deoxy sugar esters has been developed. The hydroxy carboxylic acids of complex structure ($PGF_{2\alpha}$, DCA) were treated without protection of alcoholic hydroxyl groups. The synthesis began with *O*-alkylation of the carboxylic acid with bromoaldehyde affording deoxy sugar esters in the form of an aldehyde. The latter was deacetylated selectively by CALB, and DOS esters, in the form of hemiacetals, were gained. Integration of the *O*-alkylation and CALB-catalyzed deacetylation into a "one-pot synthesis" is justified because of the lability of aldehydes. However, the lipase-catalyzed hydrolytic deacetylation should be carefully performed because an uncontrolled incubation of the DOS esters under basic conditions may lead to partial hydrolysis of the product. The resolution of stereoisomeric DOS esters has been performed upon lipase-catalyzed acetylation of the hemiacetals. In all cases, the derivatives corresponding to (2′*R*)-3,4-dideoxy ribose afforded individual acetylated hemiacetals. Depending on structure, the CALB-catalyzed acetylation of (2′*S*)-3,4-dideoxy ribose esters led to a stereochemically pure aldehyde (OA ester), a diastereomeric mixture of the aldehyde (DCA ester), or novel 1,4-dioxane hemiacetal products (MA ester).

Experimental Section

General Procedure A: The *O*-Alkylation of a Carboxylic Acid Followed by a Lipase-Catalyzed Deacetylation. Carboxylic acid (1 mmol) was dissolved in CH_3CN (8 mL), 4 equiv of DIPEA was added, followed by 0.7 mmol of bromoaldehyde **6** dissolved in 2 mL of CH_3CN. After stirring the mixture for 24 h Novozym 435 (600 mg) and H_2O (5 mmol) were added, the mixture was shaken at rt for 18−48 h, and the reaction was monitored by TLC. The solution was diluted with Et_2O, the enzyme was filtered off, and the solution was washed with water, 1 M $NaHSO_4$, water, and brine and dried over anhydrous Na_2SO_4. The solvent was evaporated, and the product was purified by column chromatography over silica.

General Procedure B: The Lipase-Catalyzed Acetylation of Hemiacetal Compounds (8, 10, 12, 20a, 20b). To the solution of 0.2 mmol of hemiacetal in 4 mL of $CHCl_3$ were added 1 mL of vinyl acetate and 200 mg of Novozym 435. The reaction mixture was shaken at rt for 24−96 h, and the reaction was monitored by TLC. After the process had been completed, the enzyme was filtered off and the solution evaporated. The products were separated by column chromatography over silica.

General Procedure C: The Lipase Catalyzed Deacetylation of (1′*S*)-7, 15, 16 and 18. To the solution of 0.5 mmol of an acetylated compound in 8 mL of CH_3CN (containing 2% of H_2O) was added 350 mg of Novozym 435. The reaction mixture was shaken at rt, and the process was monitored by TLC. The enzyme was filtered off, the reaction mixture was evaporated, and the products were purified by column chromatography over silica.

(*E*)-Oct-2-enoic Acid (2′*S*,3′*R*)-2′-acetoxy-tetrahydropyran-3′-yl Ester (15). The synthesis was carried out following General Procedure B to yield 126 mg (48%) of **15**. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.0, 5.87, 5.82, 4.79, 3.88, 3.70, 2.20, 2.10, 2.04, 1.93, 1.82, 1.53, 1.46, 1.31, 1.29, 0.89. ^{13}C NMR (500 MHz, $CDCl_3$) δ 169.1, 165.5, 150.7, 120.7, 91.5, 67.3, 62.8, 32.2, 31.3, 27.5, 24.3, 22.4, 20.9, 13.9. MS (*m*/*z*): 225.05, 168.25, 142.30, 125.15, 100.10. IR (neat, cm^{-1}): 1198, 1264, 1362, 1440, 1468, 1654, 1723, 1757. $[\alpha]^{20}_{546} = -63$ (*c* 1.8, EtOAc). Anal. Calcd for $C_{15}H_{24}O_5$ (284.39): C, 63.35; H, 8.52. Found: C, 63.21; H, 8.54. TLC: R_f = 0.46 (eluent: 20% EtOAc/hexane). Flash chromatography eluent: 15% EtOAc/hexane.

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Supporting Information Available: General experimental methods and compound characterization data including copies of ^{1}H and ^{13}C NMR spectra. This material is available free of charge via the Internet at http://pubs.acs.org.

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