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Chemoenzymatic Synthesis of the Macrolide Antibiotic (-)-A26771B

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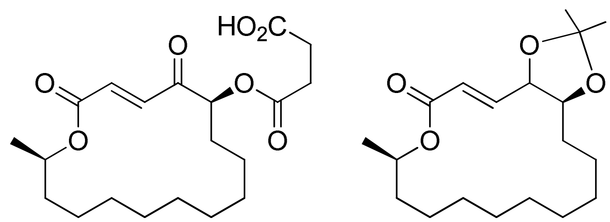
Abstract: The formal and total syntheses of the macrolide antibiotic (-)-A26771B have been developed wherein the stereochemistries at its C-5 and C-15 centres were installed using the lipase-catalyzed acylation of suitable MeCH(OH) and allylic secondary carbinol centres. A lipase-catalyzed chemoselective and hazard-free acrylation protocol, and a ring-closing metathesis reaction were used to construct the macrocylic skeleton.

Keywords: Natural products / Total synthesis / Enantioselectivity / Enzymatic catalysis / Macrocycles / Lactones

Introduction

The macrolides are of wide occurrence in various natural sources and several of these show impressive medicinal and other biological activities.[1a-c] The antimicrobial spectrum of macrolides is wider than that of penicillin, making them attractive substitutes for patients with a penicillin allergy. Several highly oxygenated, conformationally restricted marine macrolides possess outstanding cell growth antiproliferative properties, and some of these are under preclinical and/or clinical trials.[2] The 16-membered macrolide, (-)-A26771B (**1**), isolated from the fungus *Penicillium turbatum* showed moderate activity against the gram-positive bacteria, mycoplasma, and fungi.[3] Its macrolide skeleton also contains an additional keto moiety that may broaden the antimicrobial spectrum. All these have generated tremendous interest among organic chemists leading to several racemic[4] and enantioselective syntheses of **1**.[5] The first enantiomeric synthesis of **1** employed D-glucose and a chromatographic separation to instil the 5*S*- and 15*R*-stereochemistry respectively.[5a] Many of the other syntheses used (*R*)-(+)-methyloxirane to generate the required 15*R* stereocentre.[5b-e] In an interesting approach, both the stereogenic centres of **1** were introduced in a single Sharpless asymmetric dihydroxylation (AD) step,[5f] while the AD reaction in combination with a lipase-catalyzed trans-acylation were the key steps in its another synthesis.[5g] Sharpless' kinetic resolution[5c,d] or AD reaction[5e] of 2-furylcarbinols were instrumental in furnishing the required 5*S*-carbinol as well as the γ-keto-*E*-α,β-unsaturated carboxylic acid moieties of **1**. However, many of the reported syntheses of **1** were targeted to the lactone **1a** or followed the known procedure[5a] of converting **1a** to **1**. This prompted us to explore alternate strategies to develop a formal and a total synthesis of **1**. The additional motivation of the present work stems from our interest in anti-inflammatory, and anti-neoplastic agents.[6]

Compound **1** contains a methylcarbinol moiety, $CH_3CH(OH)$ that often contributes to the chirality of many biochemicals and pharmaceuticals.[7] Usually this moiety is obtained starting from the "chiral pool" compounds such as lactic acid or alanine. However, this approach provides only the (*S*)-methylcarbinol moiety, while its antipode is accessible only via circuitous routes. The biocatalytic reactions are now a viable option to develop low-waste asymmetric syntheses of pharmaceuticals, chiral intermediates, and complex target molecules.[8] Whole cell microorganisms such as bakers' yeast,[9a,b] *Rhizopus arrhizus*,[9c-e] *Geotrichum candidum*[9f,g] etc. have been effectively used for the bio-reduction of methyl ketones to the corresponding chiral methylcarbinols. Nevertheless, microbial reduction often proceeds with low enantioselectivity, and does not furnish both the alcohol enantiomers using a single biocatalyst. Use of commercially available alcohol dehydrogenases[9h] for asymmetric reduction of ketones is restricted due to the prohibitive cost of the enzymes and the cofactors. Instead, the lipase-catalyzed kinetic resolution of alcohols is more promising, as it provides the carbinol enantiomers, and when required, the efficiency of this resolution-based protocol can be improved by dynamic kinetic resolution[10a,b] or stereo-inversion under the Mitsunobu conditions.[10c-e] Lipases are commercially available, affordable, display good stereoselectivity, work in organic or aqueous media, and are easily handled by organic chemists.[11] We have used chemoenzymatic approaches involving lipase-catalyzed asymmetric reactions as the key steps for easy access to a diverse array of target compounds including the macrolides.[12] Here we present a new enantioselective synthesis of the macrolide core **1a** of the antibiotic using two lipase-catalyzed acylation reactions as the key steps for incorporating the stereogenic centres. In addition, a suitable extension of the method led to an operationally simple total synthesis of (-)-**1** using easily accessible materials/reagents.



1 **1a**

Results and Discussion

The synthetic plan of **1** was conceived in consideration of the efficacy of the inexpensive and robust lipase preparation, Novozym 435® in resolving methylcarbinols,[13a] and allylic alcohols.[13b,c] Novozym 435® is an immobilized preparation of lipase from *Candida antarctica* B (CAL-B) on acrylic resin. While the importance of the resolution of methylcarbinols is obvious, the chiral allylic alcohol moiety with a terminal alkene function was useful in constructing the macrolide structure via a ring-closing metathesis reaction (RCM).[14] The synthesis (Scheme 1) commenced from 11-bromo-1-undecene (**2**), which was converted to the corresponding Grignard reagent and subsequently reacted with acetaldehyde to furnish the alcohol (±)-**3**. The alcohol (±)-**3** was subjected to a *trans*-acetylation with vinyl acetate in hexane or diisopropyl ether in the presence of different commercial lipase preparations (porcine pancreatic lipase (PPL), *Candida rugosa* lipase (CRL), an immobilized-CRL (Sigma-Aldrich, 80841), and Novozym 435®). The results are shown in Table 1. PPL and the immobilized-CRL were ineffective in both the solvents, while CRL catalyzed the acetylation in diisopropyl ether without significant enantioselectivity. However, the Novozym 435®-catalyzed acetylation of the alcohol **3** in diisopropyl ether furnished the acetate (*R*)-**4** (97% ee, E = 126) and (*S*)-**3** (84% ee) after 40% conversion (*cf.* GC, 75 min). When the reaction was allowed to proceed up to 51% conversion (*cf.* GC, 2 h), (*S*)-**3** was obtained in 96% ee. Alternatively, the resolved alcohol (*S*)-**3** (obtained at 40% conversion) was enantiomerically enriched to 98% ee by a second Novozym 435®-

catalyzed acetylation (15% conversion) as above. The reaction was repeated several times at various scales with reproducible results.

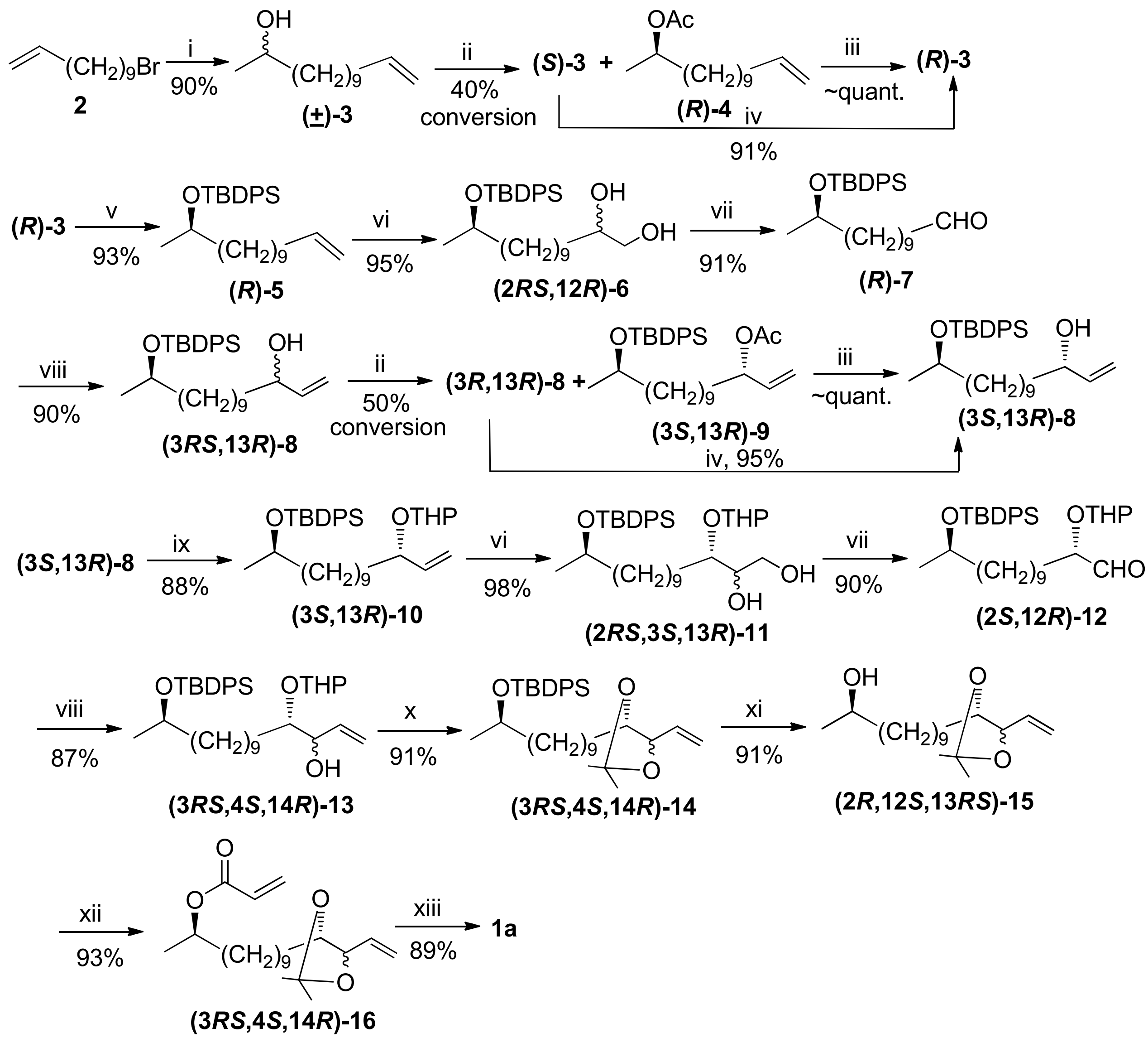
Table 1. Resolution of (±)-**3** with different lipases[a]

Entry	Lipase	Solvent	Time	% Conversion	% ee of **3**	% ee of **4**
1	PPL	hexane	48 h	<10	--	--
2	PPL	diisopropyl ether	48 h	<10	--	--
3	immobilized-CRL	hexane	48 h	Nil	--	--
4	immobilized-CRL	diisopropyl ether	48 h	Nil	--	--
5	CRL	diisopropyl ether	48 h	25[b]	--	--
6	Novozym 435®	diisopropyl ether	75 min	40	84	97
7	Novozym 435®	diisopropyl ether	2 h	51	96	92
8	Novozym 435®	diisopropyl ether	8-10 h	15	98[b]	--

[a]The experiments were carried out using (±)-**3** (2 mmol) and vinyl acetate (3 mmol) at 25 ^{o}C. [b]In this case, (*R*)-**3**, obtained from entry 6 was used.

Alkaline hydrolysis of the acetate (*R*)-**4** with K_2CO_3/aqueous MeOH furnished the alcohol (*R*)-**3**. The % ees of the enantiomeric alcohols (*R*)-**3** and (*S*)-**3** were determined from the relative intensities of the methoxyl resonances of the corresponding α-methoxytrifluoromethyl phenyl acetates (MTPA), prepared using (*R*)-MTPA chloride.[15] The configurations of the alcohols were assigned by converting a small aliquot of the respective alkenol enantiomers into tridecan-2-ol enantiomers and comparing their optical rotations with the reported values.[16] As per the requirement of the synthesis, (*S*)-**3** was converted to its enantiomer (*R*)-**3** under the Mitsunobu conditions (Ph_3P/DIAD/*p*-nitrobenzoic acid/THF/8 h; K_2CO_3/MeOH/25 ^oC/3 h, 91-95%).[10c,d] This made the synthesis enantioconvergent and improved its yield. The alcohol (*R*)-**3** was silylated with *tert*-butyldiphenylsilyl chloride (TBDPSCl) in the presence of imidazole as the base and 4-dimethylaminopyridine (DMAP) to furnish **5**. Dihydroxylation of the alkene function in compound **5** with OsO_4/*N*-methylmorpholine *N*-oxide (NMO) gave the diol **6**, which was converted to the aldehyde **7** by reacting with $NaIO_4$. Reaction of the aldehyde **7** with vinylmagnesium bromide gave the alcohol **8** as a mixture of C-3 epimers.

Next, the alcohol **8** was subjected to another Novozym 435®-catalyzed acetylation with vinyl acetate in diisopropyl ether to produce the acetate **9** (95% ee, E = 145) and (3*R*,13*R*)-**8** (98% ee) after 50% conversion (*cf*. GC, 6 h, E = 98). The stereochemical outcome of the transesterification was consistent with our previous results,[13] and followed Kazlauskas' empirical rule.[17] For determination of the % ees of the products, the acetate **9** was subjected to alkaline hydrolysis to obtain (3*S*)-**8**. Subsequently, both (3*S*)-**8** and (3*R*)-**8** were converted to their respective (*R*)-MTPA esters and analyzed by ^{1}H NMR spectra as above. Next, in order to increase the yield of the synthesis, the alcohol (3*R*,13*R*)-**8** was converted to the required alcohol (3*S*,13*R*)-**8** by a Mitsunobu inversion. The carbinol function in (3*S*,13*R*)-**8** was protected with 3,4-dihydropyran (DHP) in the presence of pyridinium *p*-toluenesulphonate (PPTS) in CH_2Cl_2 to furnish **10**. Conversion of **10** to the allylic alcohol **13** was straightforward and involved alkene dihydroxylation, $NaIO_4$ cleavage of the resultant diol **11**, followed by reaction of the aldehyde **12** with vinylmagnesium bromide. Since the C-3 carbinol function would be eventually converted to the keto group in the target compound, the synthesis was continued using the C-3 epimeric mixtures of **13**. Thus, compound **13** was depyranylated and the resultant diol reacted with 2,2-dimethoxypropane (2,2-DMP) in

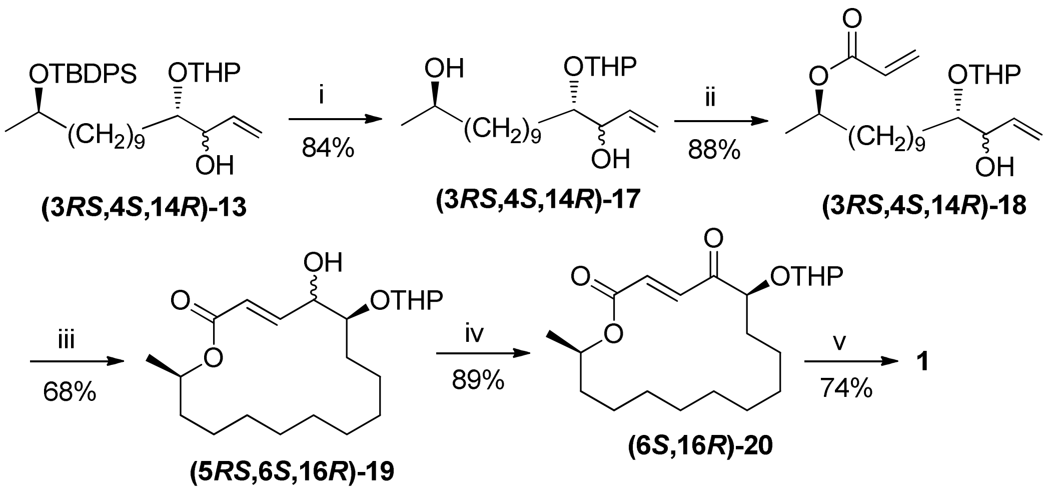


i) Mg/THF/25 °C/CH_3CHO/3 h, ii) Vinyl acetate/ diisopropyl ether/Novozym 435®/25 °C/75 min (for **(±)-3**); 6 h (for **(3*RS*,13*R*)-8**), iii) K_2CO_3/aqueous MeOH/25 °C/6 h, iv) DIAD/Ph_3P/*p*-$NO_2C_6H_4CO_2H$/THF/8 h; K_2CO_3/aqueous MeOH, v) TBDPSCl/imidazole/4-DMAP/CH_2Cl_2/0 to 25 °C/7 h, vi) OsO_4/NMO/acetone-H_2O (8:1)/*t*-BuOH/25 °C/10 h, vii) $NaIO_4$/MeCN-H_2O/0 °C/2 h, viii) CH_2=CHMgBr/THF/-78 °C/1 h, ix) DHP/PPTS /CH_2Cl_2/25 °C/4 h, x) PPTS/MeOH/25 °C/6 h; 2,2-DMP/PPTS/25 °C/12 h, xi) Bu_4NF/THF/0 °C/4 h, xii) CH_2=$CHCO_2Et$/ Novozym 435®/25 °C/24 h, xiii) Grubbs' II catalyst/CH_2Cl_2/Δ/8 h.

Scheme 1.

the presence of PPTS to furnish the acetonide **14**. This was desilylated with Bu_4NF in THF to furnish the alcohol **15**. This on reaction with ethyl acrylate in the presence of Novozym 435® as the catalyst afforded the acrylate ester **16**. Finally, an RCM reaction of **16** in the presence of Grubbs' II catalyst in refluxing CH_2Cl_2 furnished the desired macrolide **1a** in good (75%) yield. The spectral data of the synthesized **1a** were in conformity with its structure and corresponded well with the reported values.[5]

Synthesis of the macrolide **1** from **1a** requires hydrolysis of the acetonide function, followed by the regioselective installation of a succinic acid moiety onto the C-5 carbinol function and oxidation of the C-4 carbinol moiety. Previous attempts for selective oxidation of the unnecessary C-4-hydroxyl group in the presence of those at the C-5 and C-15 positions were unsuccessful. Hence, the synthesis of the macrolide **1** from **1a** was earlier achieved using multiple steps.[5a-c] In view of these, presently, the Scheme 1 was modified to formulate an improved total synthesis of **1**. For this (Scheme 2), the alcohol **13** was desilylated with Bu_4NF in THF to obtain the diol **17**. Its Novozym 435-catalyzed reaction with ethyl acrylate proceeded regioselectively at the methylcarbinol centre, without affecting the allylic carbinol function to furnish **18**.



i) Bu_4NF/THF/0 °C/4 h, ii) CH_2=$CHCO_2Et$/Novozym 435/25 °C/72 h, iii) Grubbs' II catalyst/CH_2Cl_2/ 50 °C/4 h, iv) PCC/NaOAc/CH_2Cl_2/2 h, v) TFA/moist THF/0 °C/3 h; succinic anhydride/DMAP (cat.)/ CH_2Cl_2/2 h.

Scheme 2.

Due to its potential application in chemical industry, the lipase-catalyzed acrylation has been extensively studied.[18] But till to date there is only report of using it for the kinetic resolution of alcohols.[19] It is noteworthy that we achieved the acrylation using commercially available ethyl acrylate instead of vinyl acrylate that was used previously and needed to be synthesized separately.[19] Despite being a slow reaction, we found several advantages in the enzymatic acrylation, compared to the conventional base-catalyzed reaction with acryloyl chloride. The enzymatic reaction could be carried out with **15** (*vide supra*) and **17** avoiding the hygroscopic, hazardous and toxic acryloyl chloride. With both the compounds, the reaction proceeded without any side reaction or formation of any colored products, and the acrylate esters **16** and **18** were conveniently isolated by filtering the reaction mixture, column chromatography followed by solvent removal. In particular, the Novozym 435®-catalyzed acrylation of **17** was very interesting. Given that Novozym 435® is known to acylate both 2-alkanols and 3-alkenols,[13] the exclusive formation of **18** suggested that the chosen lipase can also discriminate between the designated carbinol functionalities. Further, the 14*R*-stereochemistry of the alcohols **15** and **17** also matched with the inherent enantioselectivity of the chosen lipase. Hence this strategy may be useful in asymmetric syntheses of compounds, possessing a chiral methylcarbinol moiety. At present we don't have any explanation for the observed chemo-selectivity of the reaction. Nevertheless, our results are valuable in organic synthesis, and unprecedented to the best of our knowledge. Finally, an RCM reaction of **18** in the presence of Grubbs' II catalyst furnished the macrolide **19** in good (68%) yield. This on oxidation with buffered pyridinium chlorochromate (PCC) gave the ketone **20** uneventfully. Depyranylation of **20** with aqueous trifluoroacetic acid (TFA), followed by a base-catalyzed succinoylation produced the target compound **1**.

Conclusions

The macrolide antibiotic (-)-A26771B has been synthesized using a chemoenzymatic approach. The required stereogenic centres of the target molecules were instilled using the biocatalytic reactions as the key steps. We also used Mitsunobu inversion after the lipase-catalyzed acylation steps to offset the limitation of a resolution-based synthesis by making it enantio-convergent. This improved the yield of the synthesis. Since our methodology gives access to all possible stereoisomers of the key intermediate **8**, it would be possible to access all the stereomers of the macrolide based on the described methodology. This strategy also provided easy access to the enantiopure methylcarbinol $CH_3CH(OH)$ and secondary allylic alcohol moieties that are very useful for the syntheses of many bioactive compounds. Further, the unprecedented Novozym 435®-catalyzed protocol for the chemo-selective acrylation using a non-traditional acyl donor (ethyl acrylate) is particularly noteworthy and elevates the significance of the work. This strategy may be useful in kinetic resolution of a racemic $CH_3CH(OH)$ moiety to furnish the corresponding acrylates for their subsequent conversion to a diverse array of natural products. Use of inexpensive reagents/materials, and application of operationally simple reactions were the other attractive features of the flexible, efficient and scalable syntheses.

Experimental Section

General methods

The chemicals (Fluka and Lancaster) were used as received. Other reagents were of AR grade. All anhydrous reactions were carried out under an Ar atmosphere, using freshly dried solvents. The organic extracts were dried over anhydrous Na_2SO_4. The IR spectra as thin films were scanned with a Jasco model A-202 FT-IR spectrometer. The ^1H NMR (200 MHz) and ^{13}C NMR (50 MHz) spectra were recorded with a Bruker AC-200 spectrometer. The optical rotations were recorded with a Jasco DIP 360 digital polarimeter.

(±)-Tridec-12-en-2-ol (3). To a stirred solution of the Grignard reagent prepared from **2** (10.0 g, 43.1 mmol) and Mg (1.25 g, 51.8 mmol) in THF (170 mL) was added acetaldehyde (3.61 mL, 64.7 mmol) in THF (20 mL). After stirring for 3 h, the mixture was treated with aqueous saturated NH_4Cl, the organic layer separated, and the aqueous portion extracted with Et_2O (3 × 80 mL). The combined organic extracts were washed with H_2O (3 × 15 mL) and brine (1 × 5 mL), dried and concentrated in vacuo. The residue was purified by column chromatography (silica gel, 0-10% Et_2O/hexane) to afford pure (±)-**3** (7.7 g, 90%). colorless oil; IR ν (cm^{-1}) 3374, 1640; ^1H NMR (200 MHz) δ 1.17 (d, J = 6.2 Hz, 3H), 1.28-1.52 (m containing a s at δ 1.29, 17H), 1.97-2.04 (m, 2H), 3.73-3.82 (m, 1H), 4.89-5.01 (m, 2H), 5.73-5.87 (m, 1H). ^{13}C NMR (50 MHz) δ 23.2, 25.7, 28.8, 29.0, 29.3, 29.4, 29.5, 33.7, 39.2, 67.7, 113.9, 138.9. Anal. Calcd. for $C_{13}H_{26}O$: C, 78.72; H, 13.21%. Found: C, 78.58; H, 12.93%.

Optimization of the lipase-catalyzed acetylation of (±)-3. A mixture (±)-**3** (2 mmol), vinyl acetate (3 mmol) and different lipases in hexane or diisopropyl ether (3 mL) was agitated on an orbital shaker at 110 rpm at 25 ^{o}C for different periods (Table 1). The extent of conversion was determined by analyzing an aliquot of the reaction mixture by GC. The GC analyses were carried out with a Shimadzu GC–2010 Plus instrument (Shimadzu Corporation, Kyoto, Japan) equipped with a split/splitless injector, FID detector using a DB-5 (5%-phenyl)-methylpolysiloxane, J&W Scientific, Folsom, CA, USA) capillary column (length, 30 m; i.d., 0.25 mm and film thickness, 0.25 µm). The operating conditions were: column temperature programmed from 80 to 200 ºC at the rate of 4 ºC/min, held at initial temperature for 5 min and at 200 ºC for 2 min and further to 280 ºC at the rate of 10 ºC/min, held at final temperature for 10 min; injection port temperature: 210 ºC; carrier gas He (flow rate, 1.0 mL/min). Samples (0.1 µL) were injected in the splitless mode.

(*R*)-12-Acetoxytridec-1-ene (4). A mixture of (±)-**3** (3.5 g, 17.7 mmol), vinyl acetate (2.4 mL, 26.4 mmol) and Novozyme 435® (0.175 g) in diisopropyl ether (25 mL) was agitated on an orbital shaker at 110 rpm for (75 min). The reaction mixture was filtered, and the solution concentrated in vacuo to get a residue, which on column chromatography (silica gel, 0-10% EtOAc/hexane) gave pure (*S*)-**3** (1.9 g, 54%) and (*R*)-**4** (1.5 g, 35%). (*S*)-**3**: colorless oil; $[\alpha]_D^{24}$ +5.7 (*c* 1.15, $CHCl_3$). (*R*)-**4**: colorless oil; $[\alpha]_D^{24}$ -1.7 (*c* 1.06, $CHCl_3$); IR ν (cm^{-1}) 1738, 1243; ^{1}H NMR (200 MHz) δ 1.18 (d, J = 6.2 Hz, 3H), 1.22-1.63 (m containing a s at δ 1.25, 16H), 1.98-2.10 (m containing a s at δ 2.01, 5H), 4.82-5.01 (m, 3H), 5.77-5.81 (m, 1H). ^{13}C NMR (50 MHz) δ 19.5, 20.8, 25.1, 28.6, 28.8, 29.1, 29.2, 33.5, 35.6, 70.4, 113.8, 138.5, 169.9. Anal. Calcd. for $C_{15}H_{28}O_2$: C, 74.95; H, 11.74%. Found: C, 74.71; H, 12.03%.

(*S*)-Tridec-12-en-2-ol ((*S*)-3). Following the same procedure, (*S*)-**3** (1.9 g, 9.60 mmol) (obtained from the above experiment) was acetylated with vinyl acetate till 15% conversion, and the product purified by column chromatography to obtain enantiomerically pure (*S*)-**3** (1.5 g, 80%).

colorless oil; $[\alpha]_D^{26}$ +6.7 (*c* 1.50, $CHCl_3$); IR ν (cm^{-1}) 3349, 1641, 988; ^1H NMR (200 MHz) δ 1.17 (d, J = 6.2 Hz, 3H), 1.26-1.53 (m containing a s at δ 1.36, 17H), 1.97-2.08 (m, 2H), 3.71-3.83 (m, 1H), 4.89-5.03 (m, 2H), 5.70-5.90 (m, 1H). ^{13}C NMR (50 MHz) δ 23.3, 25.7, 28.8, 29.0, 29.4, 29.5, 29.6, 33.7, 39.2, 67.9, 114.0, 139.0.

(*R*)-Tridec-12-en-2-ol ((*R*)-3). A mixture of (*R*)-**4** (2.18 g, 9.10 mmol) and 2M K_2CO_3 in 10% aqueous MeOH (20 mL) was stirred at room temperature for 6 h. The mixture was filtered, concentrated in vacuo, H_2O (30 mL) added into it, and extracted with EtOAc (2 × 20 mL). The organic layer was washed with H_2O (2 × 10 mL) and brine (1 × 5 mL), and dried. Removal of solvent in vacuo followed by column chromatography of the residue (silica gel, 0-10% EtOAc/hexane) afforded pure (*R*)-**3** (1.7 g, ~quant.). colorless oil; $[\alpha]_D^{25}$ -6.3 (*c* 1.15, $CHCl_3$); IR ν (cm^{-1}) 3371, 1640, 991; ^1H NMR (200 MHz) δ 1.18 (d, J = 6.2 Hz, 3H), 1.23-1.54 (m containing a s at δ 1.28, 17H), 1.99-2.06 (m, 2H), 3.75-3.81 (m, 1H), 4.90-5.04 (m, 2H), 5.75-5.88 (m, 1H). ^{13}C NMR (50 MHz) δ 23.4, 25.7, 28.9, 29.1, 29.4, 29.5, 29.6, 33.8, 39.3, 68.1, 114.0, 139.2. Anal. Calcd. for $C_{13}H_{26}O$: C, 78.72; H, 13.21%. Found: C, 78.35; H, 13.56%.

(*R*)-12-*tert*-Butyldiphenylsilyloxytridec-1-ene (5). To a stirred and cooled (0 °C) solution of the mixture of (*R*)-**3** (1.6 g, 8.08 mmol), imidazole (0.82 g, 12.12 mmol) and DMAP (catalytic) in CH_2Cl_2 (20 mL) was dropwise added TBDPSCl (2.67 g, 9.70 mmol). After stirring the mixture for 7 h at room temperature, it was poured into ice-cold H_2O (20 mL), the organic layer separated and the aqueous portion extracted with $CHCl_3$ (3 × 10 mL). The combined organic extracts were washed with H_2O (2 × 10 mL) and brine (1 × 5 mL), and dried. Removal of solvent in vacuo followed by purification of the residue by column chromatography (silica gel, 0-5% EtOAc/hexane) afforded pure **5** (3.3 g, 93%). colorless oil; $[\alpha]_D^{26}$ +16.3 (*c* 1.16, $CHCl_3$); IR ν (cm^{-1}) 997, 910; ^1H NMR (200 MHz) δ 1.07 (merged s and d, J = 6.0 Hz, 12H), 1.22-1.65 (m, 16H), 2.01-2.09 (m, 2H), 3.78-3.90 (m, 1H), 4.90-5.10 (m, 2H), 5.74-5.94 (m, 1H), 7.38-7.48 (m, 6H), 7.66-7.74 (m, 4H). ^{13}C NMR (50 MHz) δ 19.3, 23.2, 25.2, 27.0, 28.9, 29.1, 29.5, 29.6, 33.8, 39.5, 69.6, 114.1, 127.3, 127.4, 129.4, 134.7, 135.0, 135.9, 139.2. Anal. Calcd. for $C_{29}H_{44}OSi$: C, 79.75; H, 10.15%. Found: C, 79.56; H, 10.47%.

(2*RS*,12*R*)-12-*tert*-Butyldiphenylsilyloxytridecane-1,2-diol (6). To a stirred solution of *(R)*-**5** (3.54 g, 8.12 mmol) and NMO (2.19 g, 16.24 mmol) in acetone-H_2O (8:1, 20 mL) was added OsO_4 (0.103 g, 0.41 mmol) in *tert*-BuOH (4 mL). After consumption of **5** (*cf.* TLC, 10 h), the reaction mixture was treated with aqueous saturated Na_2SO_3 and stirred for 1 h. The organic layer was separated and the aqueous portion extracted with EtOAc (3 × 100 mL). The combined organic extracts were washed with H_2O (2 × 30 mL) and brine (1 × 10 mL), dried and concentrated in vacuo. The residue was purified by column chromatography (silica gel, 0-40% EtOAc/hexane) to afford pure **6** (3.6 g, 95%). colorless oil; $[\alpha]_D^{27}$ +12.0 (*c* 1.05, $CHCl_3$); IR ν (cm^{-1}) 3375; ^{1}H NMR (200 MHz) δ 1.08 (merged s and d, J = 6.0 Hz, 12H), 1.15-1.47 (m, 18H), 1.69 (broad s, 2H), 3.43-3.52 (m, 1H), 3.65-3.88 (m, 3H), 7.35-7.48 (m, 6H), 7.67-7.78 (m, 4H). ^{13}C NMR (50 MHz) δ 19.2, 23.2, 25.2, 25.5, 27.0, 29.5, 29.6, 33.2, 39.4, 66.8, 69.6, 72.3, 127.3, 127.4, 129.3, 134.7, 135.0, 135.8. Anal. Calcd. for $C_{29}H_{46}O_3Si$: C, 73.99; H, 9.85%. Found: C, 73.78; H, 9.75%.

(*R*)-11-*tert*-Butyldiphenylsilyloxydodecanal (7). To a cooled (0 ^{o}C) and stirred solution of **6** (3.86 g, 8.21 mmol) in MeCN-H_2O (3:2, 15 mL) was added $NaIO_4$ (3.52 g, 16.43 mmol). After stirring for 2 h, the mixture was concentrated in vacuo, the residue taken in EtOAc (30 mL) and washed successively with H_2O (1 × 10 mL), aqueous 10% $NaHSO_3$ (1 × 10 mL), H_2O (2 × 10 mL) and brine (1 × 5 mL), and dried. Solvent removal furnished the pure aldehyde **7** (3.2 g, 91%). colorless oil; $[\alpha]_D^{27}$ +14.9 (*c* 1.03, $CHCl_3$); IR ν (cm^{-1}) 2712, 1727; ^{1}H NMR (200 MHz) δ

1.09 (merged s and d, J = 6.2 Hz, 12H), 1.18-1.40 (m, 12H), 1.60-1.80 (m, 4H), 2.46 (dt, J = 1.8, 7.2 Hz, 2H), 3.79-3.95 (m, 1H), 7.35-7.48 (m, 6H), 7.72-7.83 (m, 4H), 9.80 (t, J = 1.8 Hz, 1H). ^{13}C NMR (50 MHz) δ 19.0, 21.9, 23.0, 24.5, 25.0, 26.8, 28.8, 29.0, 29.1, 29.2, 29.3, 33.8, 39.2, 43.7, 69.4, 127.1, 127.2, 129.2, 134.5, 134.8, 135.7, 179.6. Anal. Calcd. for $C_{28}H_{42}O_2Si$: C, 76.66; H, 9.65%. Found: C, 76.33; H, 9.84%.

(3*RS*,13*R*)-13-*tert*-Butyldiphenylsilyloxytetradec-1-en-3-ol (3*RS*,13*R*)-(8). To a cooled (-40 ^{o}C) and stirred solution of **7** (3.0 g, 6.84 mmol) in THF (20 mL) was added CH_2=CHMgBr (13.7 mL, 1M in THF, 13.7 mmol). After stirring for 1 h, H_2O (15 mL) was added to the mixture, the organic layer separated, and the aqueous layer extracted with EtOAc (2 × 10 mL). The combined organic extracts were washed with H_2O (1 × 10 mL) and brine (1 × 5 mL), and dried. Solvent removal followed by column chromatography (silica gel, 0-15% EtOAc-hexane) of the residue gave pure (3*RS*,13*R*)-**8** (2.9 g, 90%). colorless oil; $[\alpha]_D^{27}$ +14.2 (*c* 1.08, $CHCl_3$); IR ν (cm^{-1}) 3359, 996, 920 ; ^{1}H NMR (200 MHz) δ 1.03 (merged s and d, J = 6.0 Hz, 12H), 1.15-1.35 (m, 12H), 1.45-1.68 (m containing a s at 1.56 Hz, 7H), 3.73-3.85 (m, 1H), 4.03-4.13 (m, 1H), 5.07-5.25 (m, 2H), 5.76-5.96 (m, 1H), 7.29-7.41 (m, 6H), 7.63-7.78 (m, 4H). ^{13}C NMR (50 MHz) δ 19.2, 23.2, 25.2, 25.3, 27.0, 29.5, 37.0, 39.4, 69.6, 73.2, 114.5, 127.3, 127.4, 129.3, 129.4, 134.6, 134.9, 135.8, 141.3. Anal. Calcd. for $C_{30}H_{46}O_2Si$: C, 77.19; H, 9.93%. Found: C, 77.44; H, 10.21%.

(3*S*,13*R*)-3-Acetoxy-13-*tert*-butyldiphenylsilyloxytetradec-1-ene (9). Acetylation of (3*RS*,13*R*)-**8** (2.1 g, 4.51 mmol) with vinyl acetate (0.58 g, 6.76 mmol) in diisopropyl ether (25 mL) and Novozyme 435® (0.035 g) for 6 h, followed by usual work up, isolation and purification by column chromatography (silica gel, 0-10% EtOAc/hexane) gave pure **9** (0.860 g, 45%) and (*3R,13R*)-**8** (0.860 g, 41%). (*3R,13R*)-**8**: colorless oil; $[\alpha]_D^{22}$ +11.9 (*c* 1.05, $CHCl_3$); IR ν (cm^{-1}) 3367, 991, 927; ^{1}H NMR (200 MHz) *δ* 1.01 (merged s and d, *J* = 5.8 Hz, 12H), 1.15-1.26 (m, 14H), 1.44-1.55 (m, 4H), 2.01 (s, 1H), 3.78-3.81 (m, 1H), 4.05-4.11 (m, 1H), 5.04-5.24 (m, 2H), 5.76-5.84 (m, 1H), 7.32-7.37 (m, 6H), 7.62-7.67 (m, 4H). ^{13}C NMR (50 MHz) *δ* 19.2, 23.2, 25.2, 26.9, 29.6, 37.1, 39.3, 69.5, 73.3, 114.8, 127.6, 129.4, 134.9, 135.8, 141.3. Anal. Calcd. for $C_{30}H_{46}O_2Si$: C, 77.19; H, 9.93%. Found: C, 77.29; H, 9.71%. **9**: colorless oil; $[\alpha]_D^{22}$ +9.1 (*c* 1.02, $CHCl_3$); IR ν (cm^{-1}) 1732, 1246, 997; ^{1}H NMR (500 MHz) *δ* 1.05 (s, 9H), 1.20-1.28 (m, 17H), 1.42-1.66 (m, 4H), 2.07 (s, 3H), 3.81-3.84 (m, 1H), 5.15-5.17 (m, 1H), 5.22-5.26 (m, 2H), 5.75-5.80 (m, 1H), 7.35-7.46 (m, 6H), 7.70-7.78 (m, 4H). ^{13}C NMR (125 MHz) *δ* 19.3, 21.3, 23.3, 25.1, 25.2, 27.1, 29.4, 29.5, 29.6, 29.7, 32.6, 34.2, 39.5, 69.6, 74.9, 116.5, 127.4, 127.5, 127.6, 129.4, 134.7, 135.0, 135.6, 135.9, 136.7, 170.4. Anal. Calcd. for $C_{32}H_{48}O_3Si$: C, 75.74; H, 9.51%. Found: C, 75.82; H, 9.86%.

(3*S*,13*R*)-13-*tert*-Butyldiphenylsilyloxytetradec-1-en-3-ol ((3*S*,13*R*)-8). Hydrolysis of **9** (0.80 g, 1.57 mmol) with 2M K_2CO_3 in aqueous MeOH (25 mL) followed by work-up and column chromatography (silica gel, 0-10% EtOAc/hexane) afforded pure (3*S*,13*R*)-**8** (0.725 g, ~quant.). colorless oil; $[\alpha]_D^{23}$ +14.7 (*c* 1.01, $CHCl_3$); IR ν (cm^{-1}) 3367, 1006, 927; ^{1}H NMR (200 MHz) *δ* 1.03 (merged s and d, *J* = 6.0 Hz, 12H), 1.15-1.26 (m, 14H), 1.47-1.56 (m, 4H), 3.75-3.82 (m, 1H), 4.05-4.11 (m, 1H), 5.04-5.24 (m, 2H), 5.76-5.88 (m, 1H), 7.36-7.39 (m, 6H), 7.62-7.67 (m, 4H). ^{13}C NMR (125 MHz) *δ* 19.3, 23.2, 25.2, 25.4, 27.1, 29.6, 37.1, 39.5, 69.6, 73.3, 114.5, 127.4, 127.6, 129.4, 134.7, 135.0, 135.6, 135.9, 141.4. Anal. Calcd. for $C_{30}H_{46}O_2Si$: C, 77.19; H, 9.93%. Found: C, 77.37; H, 10.28%.

(3*S*,13*R*)-13-*tert*-Butyldiphenylsilyloxy-3-tetrahydropyranyloxytetradec-1-ene (10). A mixture of (3*S*,13*R*)-**8** (0.7 g, 1.5 mmol), DHP (0.2 mL, 2.25 mmol) and PPTS (catalytic) in CH_2Cl_2 (10 mL) was stirred for 4 h at room temperature. The mixture was poured into ice-cold aqueous 10% $NaHCO_3$ (20 mL), the organic layer separated and the aqueous portion extracted with $CHCl_3$ (3 × 10 mL). The combined organic extracts were washed with H_2O (2 × 10 mL) and brine (1 × 10 mL), and dried. Removal of solvent in vacuo followed by purification of the residue by column chromatography (silica gel, 0-5% EtOAc/hexane) afforded pure **10** (0.73 g, 88%). colorless oil; $[\alpha]_D^{29}$ +8.9 (*c* 1.07, $CHCl_3$); IR ν (cm^{-1}) 1320, 1259, 1077; ^{1}H NMR (200 MHz) δ 1.04 (merged s and d, J = 6.0 Hz, 12H), 1.16-1.25 (m, 14H), 1.49-1.65 (m, 10H), 3.42-3.52 (m, 1H), 3.76-3.89 (m, 2H), 4.03-4.07 (m, 1H), 4.63-4.68 (m, 1H), 5.11-5.24 (m, 2H), 5.56-5.86 (m, 1H), 7.34-7.38 (m, 6H), 7.64-7.69 (m, 4H). ^{13}C NMR (50 MHz) δ 19.2, 19.6, 23.2, 25.0, 25.2, 25.5, 27.0, 29.5, 29.6, 30.8, 35.6, 39.4, 62.3, 69.6, 95.0, 97.7, 114.7, 117.2, 127.3, 127.4, 129.3, 129.4, 134.6, 135.0, 135.9, 138.7, 139.8. Anal. Calcd. for $C_{35}H_{54}O_3Si$: C, 76.31; H, 9.88%. Found: C, 76.71; H, 10.28%.

(2*RS*,3*S*,13*R*)-13-*tert*-Butyldiphenylsilyloxy-3-tetrahydropyranyloxytetradecane-1,2-diol (11). Dihydroxylation of **10** (0.7 g, 1.27 mmol) with NMO (0.171 g, 2.54 mmol) and OsO_4 (0.016 g, 0.06 mmol) in acetone-H_2O (8:1, 10 mL) followed by usual isolation and column chromatography (silica gel, 0-40% EtOAc/hexane) afforded pure **11** (0.730 g, 98%). colorless oil; $[\alpha]_D^{26}$ +4.5 (*c* 1.03, $CHCl_3$); IR ν (cm^{-1}) 3410, 1389, 1183; ^{1}H NMR (200 MHz) δ 1.03 (merged s and d, J = 6.0 Hz, 12H), 1.17-1.24 (m, 14H), 1.44-1.86 (m, 10H), 1.89-2.19 (m, 2H), 3.47-3.57 (m, 2H), 3.63-3.76 (m, 4H), 3.79-3.88 (m, 1H), 4.06-4.10, 4.29-4.39 and 4.72-4.75 (three m, 1H), 7.34-7.41 (m, 6H), 7.64-7.68 (m, 4H). ^{13}C NMR (50 MHz) δ 19.1, 19.9, 21.6, 23.1, 24.8, 25.1, 25.2, 25.4, 25.9, 26.9, 29.4, 29.7, 30.8, 31.2, 31.3, 32.2, 39.3, 63.0, 63.2, 65.8, 69.5, 72.7, 73.2, 79.2, 83.1, 99.1, 102.4, 127.2, 127.3, 129.2, 129.3, 134.5, 134.8, 135.8. Anal. Calcd. for $C_{35}H_{56}O_5Si$: C, 71.87; H, 9.65%. Found: C, 71.48; H, 9.77%.

(2*S*,12*R*)-12-*tert*-Butyldiphenylsilyloxy-2-tetrahydropyranyloxytridecanal (12). Reaction of **11** (0.73 g, 1.25 mmol) with $NaIO_4$ (0.535 g, 2.50 mmol) in MeCN-H_2O (3:2, 15 mL) at 0 °C, followed by usual work-up furnished the pure aldehyde **12** (0.620 g, 90%). colorless oil; $[\alpha]_D^{25}$ -16.7 (*c* 1.01, $CHCl_3$); IR ν (cm^{-1}) 2707, 1734; ^{1}H NMR (200 MHz) δ 1.04 (merged s and d, J = 6.2 Hz, 12H), 1.17-1.25 (m, 12H), 1.31-1.88 (m, 12H), 3.46-3.56 (m, 1H), 3.74-3.92 (m, 2H), 4.17 (dt, J = 1.4, 7.2 Hz, 1H), 4.53-4.56 and 4.67-4.70 (two m, 1H), 7.35-7.41 (m, 6H), 7.62-7.70 (m, 4H), 9.63 (d, J = 1.6 Hz, 1H). ^{13}C NMR (50 MHz) δ 19.2, 19.3, 23.2, 25.1, 25.2, 25.3, 27.0, 29.3, 29.4, 29.5, 30.0, 30.5, 39.4, 62.6, 69.5, 79.8, 83.6, 97.6, 100.9, 127.3, 127.4, 129.3, 129.4, 134.6, 134.9, 135.8, 203.4, 203.9. Anal. Calcd. for $C_{34}H_{52}O_4Si$: C, 73.86; H, 9.48%. Found: C, 73.75; H, 9.88%.

(3*RS*,4*S*,14*R*)-14-*tert*-Butyldiphenylsilyloxy-4-tetrahydropyranyloxypentadec-1-en-3-ol (13). As described above, reaction of **12** (0.600 g, 1.09 mmol) with CH_2=CHMgBr (2.2 mL, 1M in THF, 2.20 mmol) in THF (10 mL) at -78 °C, followed by usual work up, and column chromatographic purification (silica gel, 0-20% EtOAc/hexane) afforded pure **13** (0.547 g, 87%). colorless oil; $[\alpha]_D^{27}$ -6.4 (*c* 1.09, $CHCl_3$); IR ν (cm^{-1}) 3433, 996, 921; ^{1}H NMR (200 MHz) δ 1.02 (merged s and d, J = 6.2 Hz, 12H), 1.06-1.29 (m, 15H), 1.31-1.66 (m, 8H), 1.77-1.82 (m, 2H), 3.44-3.66 (m, 2H), 3.71-4.04 (m, 3H), 4.18-4.45 and 4.62-4.78 (two m, 1H), 5.10-5.33 (m, 2H), 5.76-5.93 (m, 1H), 7.21-7.41 (m, 6H), 7.60-7.68 (m, 4H); ^{13}C NMR (50 MHz) δ 13.9, 19.0, 19.7, 21.0, 23.1, 24.8, 25.0, 25.2, 25.7, 25.8, 26.9, 29.3, 29.5, 30.7, 31.1, 31.8, 36.9, 39.2, 62.7, 63.0, 64.9, 69.4, 72.8, 73.5, 74.7, 79.1, 85.2, 97.3, 99.1, 102.1, 114.0, 116.0, 116.3, 127.2, 127.3, 129.1, 129.2, 134.4, 134.7, 135.6, 136.6, 136.8, 138.0, 141.4. Anal. Calcd. for $C_{36}H_{56}O_4Si$: C, 74.43; H, 9.72%. Found: C, 74.19; H, 9.97%.

(3*RS*,4*S*,14*R*)-14-*tert*-Butyldiphenylsilyloxy-3,4-isopropylidenedioxypentadec-1-ene (14). A solution of **13** (0.23 g, 0.40 mmol) and PPTS (20 mol%) in MeOH (5 mL) was stirred for 6 h at room temperature. Concentration of the mixture in vacuo gave the crude product, which was diluted with 2,2-DMP (1 mL) and stirred for 12 h at room temperature. The mixture was concentrated in vacuo, H_2O (15 mL) added into it, and extracted with EtOAc (2 × 15 mL). The organic layer was washed with H_2O (2 × 10 mL) and brine (1 × 5 mL), and dried. Removal of solvent in vacuo followed by column chromatography of the residue (silica gel, 0-10% EtOAc/hexane) afforded pure **14** (0.190 g, 91%). colorless oil; $[\alpha]_D^{21}$ +28.8 (*c* 1.04, $CHCl_3$); IR ν (cm^{-1}) 3399, 992, 926; ^{1}H NMR (500 MHz) δ 1.04 (merged s and d, 12H), 1.21-1.29 (m, 10H), 1.37-1.66 (m containing two s at δ 1.37 and 1.42, 14H), 3.62-3.70, 3.81-3.84, 3.95-3.98, 4.13-4.14, and 4.46-4.48 (five m, 3H), 5.22-5.37 (m, 2H), 5.79-5.84 (m, 1H), 7.35-7.45 (m, 6H), 7.65-7.75 (m, 4H). ^{13}C NMR (125 MHz) δ 19.3, 23.2, 25.2, 25.7, 26.2, 27.1, 28.3, 29.5, 29.6, 29.7, 30.4, 39.5, 69.6, 78.3, 79.9, 80.7, 82.8, 108.0, 108.5, 118.1, 118.7, 127.4, 127.6, 129.3, 129.4, 134.7, 135.0, 135.6, 135.9. Anal. Calcd. for $C_{34}H_{52}O_3Si$: C, 76.07; H, 9.76%. Found: C, 75.91; H, 9.79%.

(2*R*,12*S*,13*RS*)-12,13-Isopropylidenedioxypentadec-14-en-2-ol (15). To a cooled (0 ^{o}C) and stirred solution of **14** (0.26 g, 0.49 mmol) in THF (5 mL) was added Bu_4NF (0.97 mL, 0.97 mmol, 1 M in THF). After stirring for 4 h, the mixture was concentrated in vacuo, the residue taken in EtOAc (10 mL) and the combined organic extracts washed with H_2O (1 × 5 mL) and brine (1 × 5 mL), and dried. The residue was purified by column chromatography (silica gel, 0-30% EtOAc/hexane) to afford pure **15** (0.131 g, 91%). colorless oil; $[\alpha]_D^{24}$ -3.3 (*c* 1.10, $CHCl_3$); IR ν (cm^{-1}) 3399, 992, 926; ^{1}H NMR (200 MHz) δ 1.15-1.55 (m containing a d at δ 1.17, *J* = 6.2 Hz, and two s at δ 1.39 and δ 1.47, 27H), 3.59-3.83, 3.93-4.17 and 4.42-4.49 (three m, 3H), 5.19-

5.39 (m, 2H), 5.71-5.90 (m, 1H). ^{13}C NMR (50 MHz) δ 23.2, 25.5, 25.6, 25.9, 26.0, 26.8, 27.2, 28.1, 29.3, 29.4, 29.5, 30.2, 31.7, 39.1, 68.0, 78.2, 79.7, 80.5, 82.6, 107.9, 108.3, 118.0, 118.6, 134.4, 135.4. Anal. Calcd. for $C_{18}H_{34}O_3$: C, 72.44; H, 11.48%. Found: C, 72.46; H, 11.61%.

(3*RS*,4*S*,14*R*)-14-Acryloxy-3,4-isopropylidenedioxypentadec-1-ene (16). A mixture of **15** (0.12 g, 0.40 mmol), ethyl acrylate (0.33 mL, 3.20 mmol) and Novozyme 435® (0.10 g) was agitated on an orbital shaker at 110 rpm for 24 h. The reaction mixture was filtered, and concentrated in vacuo to get a residue, which on column chromatography (silica gel, 0-30% EtOAc/hexane) gave pure **16** (0.100 g, 93% based on conversion) along with unreacted **15** (16%). colorless oil; $[\alpha]_D^{23}$ -4.9 (*c* 1.03, $CHCl_3$); IR ν (cm^{-1}) 1723, 986; ^{1}H NMR (200 MHz) δ 1.20-1.67 (m containing a d at δ 1.22, *J* = 6.2 Hz, and two s at δ 1.35 and 1.47, 27H), 3.65-3.82, 3.93-4.00, 4.07-4.16, and 4.42-4.49 (four m, 2H), 4.91-5.00 (m, 1H), 5.18-5.38 (m, 2H), 5.71-5.89 (m, 2H), 6.01-6.15 (m, 1H), 6.35 (dd, *J* = 1.8, 17.2 Hz, 1H). ^{13}C NMR (50 MHz) δ 19.9, 25.3, 25.7, 26.0, 26.8, 27.3, 28.2, 29.3, 29.5, 30.2, 31.8, 35.9, 71.2, 78.2, 79.9, 80.7, 82.7, 107.9, 108.4, 118.0, 118.7, 129.0, 130.1, 134.6, 135.5, 165.9. Anal. Calcd. for $C_{21}H_{36}O_4$: C, 71.55; H, 10.29%. Found: C, 71.46; H, 10.61%.

(4*RS*,5*S*,15*R*)-4,5-Dihydroxyhexadec-2-en-15-olide 4,5-acetonide (1a). A mixture of **16** (0.05 g, 0.14 mmol) and Grubbs' II catalyst (5 mol%) in CH_2Cl_2 (10 mL) was refluxed for 8 h. The reaction mixture was concentrated in vacuo and the residue subjected to column chromatography (silica gel, 0-40% EtOAc/hexane) to afford pure **1a** (0.037 g, 81%). viscous gum; $[\alpha]_D^{24}$ -20.9 (*c* 1.10, $CHCl_3$); IR ν (cm^{-1}) 1713, 982; ^{1}H NMR (200 MHz) δ 1.12-1.42 (m containing a d at δ 1.27, *J* = 6.4 Hz and two s at δ 1.36 and δ 1.50, 20H), 1.50-1.60 (m, 7H), 4.15-4.24 (m, 1H), 4.55-4.62 (m, 1H), 4.98-5.10 (m, 1H), 6.00 (d, *J* = 15.6 Hz, 1H), 6.83 (dd, *J* = 7.8, 15.6 Hz, 1H). ^{13}C NMR (50 MHz) δ 20.4, 23.1, 23.5, 25.4, 26.6, 26.7, 27.0, 27.2, 28.0, 28.3, 29.7, 35.0, 71.0, 76.3, 78.6, 108.7, 124.9, 142.2, 165.4. Anal. Calcd. for $C_{19}H_{32}O_4$: C, 70.33; H, 9.94%. Found: C, 70.57; H, 9.83%.

(3*RS*,4*S*,14*R*)-4-Tetrahydropyranyloxypentadec-1-ene-3,14-diol (17). Desilylation of **13** (0.73 g, 1.26 mmol) with Bu_4NF (2.5 mL, 1 M in THF, 2.5 mmol) in THF (10 mL) at 0 ^{o}C, followed by usual isolation and purification by column chromatography (silica gel, 0-30% EtOAc/hexane) afforded pure **17** (0.360 g, 84%). colorless oil; $[\alpha]_D^{25}$ -17.8 (*c* 1.08, $CHCl_3$); IR ν (cm^{-1}) 3399, 992, 925; ^{1}H NMR (200 MHz) δ 1.18 (d, *J* = 6.2 Hz, 3H), 1.21-1.87 (m, 26H), 3.45-3.56 (m, 1H), 3.59-3.84 (m, 2H), 3.85-4.03 (m, 2H), 4.20-4.54 and 4.65-4.88 (two m, 1H), 5.16-5.46 (m, 2H), 5.82-5.99 (m, 1H). ^{13}C NMR (50 MHz) δ 19.5, 19.7, 20.9, 23.1, 24.7, 25.1, 25.5, 25.6, 29.2, 29.3, 29.4, 29.6, 30.5, 31.0, 31.6, 36.8, 39.0, 62.5, 64.8, 67.4, 72.6, 73.4, 74.5, 79.1, 84.9, 97.3, 101.9, 113.8, 115.8, 116.2, 136.5, 136.8, 137.8, 141.3. Anal. Calcd. for $C_{20}H_{38}O_4$: C, 70.13; H, 11.18%. Found: C, 69.74; H, 11.55%.

(3*RS*,4*S*,14*R*)-14-Acryloxy-4-tetrahydropyranyloxypentadec-1-en-3-ol (18). A mixture of **17** (0.30 g, 0.88 mmol) and Novozyme 435® (0.20 g) in ethyl acrylate (0.80 mL, 7.04 mmol) was agitated on an orbital shaker at 110 rpm for 72 h. The reaction mixture was concentrated in vacuo to get a residue, which on column chromatography (silica gel, 0-30% EtOAc/hexane) gave pure **18** (0.236 g, 88% based on conversion) and unreacted **17** (23%). colorless oil; $[\alpha]_D^{25}$ -5.1 (*c* 1.18, $CHCl_3$); IR ν (cm^{-1}) 3429, 1723, 1638, 1618, 986, 921; ^{1}H NMR (200 MHz) δ 1.19-1.69 (m, 27H), 1.76-1.88 (m, 1H), 3.35-3.70 (m, 2H), 3.82-4.12 (m, 2H), 4.37-4.46 (m, 1H), 4.90-5.12 (m, 1H), 5.15-5.45 (m, 2H), 5.75-5.94 (m, 2H), 6.01-6.14 (m, 1H), 6.32-6.50 (m, 1H). ^{13}C NMR (50 MHz) δ 14.0, 19.8, 21.2, 22.6, 24.8, 25.2, 25.9, 29.3, 29.6, 31.2, 32.0, 35.8, 36.9, 65.2, 71.1, 73.0, 74.8, 85.4, 102.0, 102.3, 114.2, 116.5, 117.2, 129.0, 130.0, 136.7, 137.0, 141.3, 165.8. Anal. Calcd. for $C_{23}H_{40}O_5$: C, 69.66; H, 10.17%. Found: C, 69.35; H, 10.53%.

(5*RS*,6*S*,16*R*,3*E*)-5-Hydroxy-16-methyl-6-(tetrahydropyranyloxy)oxacyclohexadec-3-en-2-one (19). A mixture of **18** (0.10 g , 0.25 mmol) and Grubbs' II catalyst (20 mol%) in CH_2Cl_2 (10 mL) was refluxed for 4 h. Usual work-up, and purification by column chromatography (silica gel, 0-40% EtOAc/hexane) afforded pure **19** (0.063 g, 68%). colorless oil; $[\alpha]_D^{27}$ -17.0 (*c* 1.00, $CHCl_3$); IR ν (cm^{-1}) 3404, 1711, 985; ^1H NMR (200 MHz) δ 1.18-1.46 (m, 28H), 3.49-3.54, 3.70-3.76, 3.91-3.94 and 4.18-4.22 (four m, 3H), 4.59-4.62 (m, 1H), 4.72-4.81 (m, 1H), 4.88-5.10 (m, 1H), 6.04-6.18 (m, 1H), 6.82-6.95 (m, 1H). ^{13}C NMR (50 MHz) δ 20.3, 20.6, 22.3, 22.7, 23.4, 23.8, 25.2, 26.1, 27.0, 27.4, 27.7, 27.8, 28.9, 29.3, 29.5, 29.7, 30.3, 31.0, 31.1, 31.9, 35.1, 35.6, 63.0, 70.9, 71.3, 78.3, 82.5, 96.1, 100.4, 122.1, 123.0, 145.1, 145.8, 165.6, 166.1. Anal. Calcd. for $C_{21}H_{36}O_5$: C, 68.44; H, 9.85%. Found: C, 68.62; H, 9.52%.

(6*S*,16*R*,3*E*)-16-Methyl-6-(tetrahydropyranyloxy)oxacyclohexadec-3-ene-2,5-dione (20). To a cooled (0 °C) and stirred suspension of PCC (0.027 g, 0.12 mmol) and NaOAc (10 mol%) in CH_2Cl_2 (5 mL) was added **19** (0.03 g, 0.08 mmol) in one lot. After stirring for 2 h, the reaction mixture was diluted with Et_2O (15 mL) and the supernatant passed through a pad of silica gel (2" × 1"). Removal of solvent in vacuo followed by column chromatography of the residue (silica gel, 0-15% EtOAc/hexane) furnished pure **20** (0.027 g, 89%). colorless oil; $[\alpha]_D^{25}$ -53.0 (*c* 1.00, $CHCl_3$); IR ν (cm^{-1}) 1725, 980; ^1H NMR (200 MHz) δ 1.14-1.88 (m, 27H), 3.46-3.54 (m, 1H), 3.80-3.88 (m, 1H), 4.40-4.58 (m, 1H), 4.86-5.12 (m, 1H), 6.78 (d, J = 15.8 Hz, 1H), 7.28 (d, J = 15.8 Hz, 1H). ^{13}C NMR (50 MHz) δ 19.4, 20.1, 22.0, 22.7, 23.6, 25.3, 26.6, 27.5, 27.6, 28.9, 29.1, 29.3, 29.7, 30.5, 30.6, 31.9, 33.8, 34.7, 62.9, 72.6, 80.0, 97.7, 132.1, 134.9, 165.0, 199.7. Anal. Calcd. for $C_{21}H_{34}O_5$: C, 68.82; H, 9.35%. Found: C, 68.40; H, 9.49%.

Antibiotic (-)-A26771B (1). To a cooled (0 ^{o}C) and stirred solution of **20** (0.025 g, 0.07 mmol) in moist THF (5 mL) was added TFA (0.27 mL). After stirring for 3 h, the mixture was concentrated in vacuo to afford the corresponding depyranylated product (0.02 g). To a solution of the above crude product in CH_2Cl_2 (5 mL) was added DMAP (catalytic), followed by succinic anhydride (0.011 g, 0.11 mmol). After stirring for 2 h, the reaction mixture was concentrated and the residue subjected to preparative TLC (8% MeOH/$CHCl_3$) to afford pure **1** (0.019 g, 74%). white powder; mp: 122 °C (lit.[20] mp: 121-123 °C); $[\alpha]_D^{25}$ -12.2 (*c* 0.5, MeOH), (lit.[20] $[\alpha]_D^{12}$ -13 (*c* 0.2, MeOH)); IR ν (cm^{-1}) 3420, 1748, 1713, 1701; ^{1}H NMR (200 MHz) *δ* 1.24-1.43 (m containing a d at δ 1.28, *J* = 6.5 Hz, 15H), 1.56-2.01 (m, 6H), 2.29 (t, *J* = 7.0 Hz, 2H), 2.65-2.69 (m, 2H), 5.10-5.15 (m, 1H), 5.34 (t, *J* = 5.4 Hz, 1H), 6.67 (d, *J* = 15.2 Hz, 1H), 7.63 (d, *J* = 15.2 Hz, 1H), 8.16 (broad s, 1H). ^{13}C NMR (50 MHz) *δ* 19.5, 22.1, 23.5, 26.5, 26.9, 27.2, 27.4, 27.7, 28.2, 28.4, 28.7, 34.6, 72.7, 78.8, 122.8, 135.7, 165.3, 171.8, 177.1, 196.0. Anal. Calcd. for $C_{20}H_{30}O_7$: C, 62.81; H, 7.91%. Found: C, 63.18; H, 8.03%.

References

1 (a) S. Õmura, In *Macrolide Antibiotics. Chemistry, Biology and Practice*, S. Õmura, Ed.; Academic Press, Orlando 1984, pp. 509–552; (b) I. Paterson and M. M. Mansuri, *Tetrahedron*, 1985, **41**, 3569–3624; (c) N. Tadashi, In *Macrolide Antibiotics: Chemistry, Biology and Practice*, 2nd ed.; S. Omura, Ed.; Academic Press: San Diego, 2002; pp. 181–284.

2 For reviews on biology and synthesis: (a) J. G. Napolitano, A. H. Daranas, M. Norte and J. J. Fernandez, *Anticancer Agents Med. Chem*., 2009, **9**, 122–137; (b) K. J. Hale and S. Manaviazar, *Chem. Asian J*., 2010, **5**, 704–754.

3 K. H. Michel, P. V. Demarco and R. Nagarajan, *J. Antibiot*., 1977, **30**, 571–575.

4 (a) T. A. Hase and E. -L. Nylund, *Tetrahedron Lett.*, 1979, **20**, 2633–2636; (b) M. Asaoka, N. Yanagida and H. Takai, *Tetrahedron Lett*., 1980, **21**, 4611–4614; (c) M. Asaoka, T. Mukuta and H. Takai, *Chem. Lett.*, 1982, 215–218; (d) B. M. Trost and S. J. Brickner, *J. Am. Chem. Soc.*, 1983, **105**, 568–575; (e) T. Fujisawa, N. Okada, M. Takeuchi and T. Sato, *Chem. Lett*., 1983, 1271–1272; (f) H. J. Bestmann and R. Schobert, *Angew. Chem. Int. Ed. Engl.*, 1986, **24**, 791–792; (g) S. Bienz and M. Hew, *Helv. Chim. Acta*, 1987, **70**, 1333–1340; (h) J. E. Baldwin, R. M. Adlington and S. H. Ramcharitar, *J. Chem. Soc. Chem. Commun.*, 1991, 940–942.

5 (a) K. Tatsuta, Y. Amemiya, Y. Kanemura and M. Kinoshita, *Bull. Chem. Soc. Jpn.*, 1982, **55**, 3248–3253; (b) G. Quinkert, F. Kueber, W. Knauf, M. Wacker, U. Koch, H. Becker, H. P. Nestler, G. Dürner, G. Zimmermann, J. W. Bats and E. Egert, *Helv. Chim. Acta*, 1991, **74**, 1853–1923; (c) Y. Kobayashi and H. Okui, *J. Org. Chem.*, 2000, **65**,

612–615; (d) W. Lee, H. J. Shin and S. Chang, *Tetrahedron: Asymmetry*, 2001, **12**, 29–31; (e) J. Gebauer and S. Blechert, *J. Org. Chem.*, 2006, **71**, 2021–2025; (f) S. C. Sinha, A. Sinha-Bagchi and E. Keinan, *J. Org. Chem.*, 1993, **58**, 7789–7796; (g) M. Nagarajan, *Tetrahedron Lett.*, 1999, **40**, 1207–1210.

6 (a) B. Maity and S. Chattopadhyay, *Curr. Bioact. Comp.*, 2008, **4**, 225–244; (b) P. Guha, A. Dey, B. Sarkar, M. V. Dhyani, S. Chattopadhyay and S. K. Bandyopadhyay, *J. Exp. Pharmacol. Ther.*, 2009, **328**, 1–10; (c) B. S. Patro, B. Maity and S. Chattopadhyay, *Antiox. Redox. Signal.*, 2010, **12**, 945–960; (d) P. Guha, A. Dey, A. Chatterjee, S. Chattopadhyay and S. K. Bandyopadhyay, *Br. J. Pharmacol.*, 2010, **159**, 726–734; (e) B. S. Patro, M. Tyagi, J. Saha and S. Chattopadhyay, *Bioorg. Med. Chem.*, 2010, **18**, 7043–7051; (f) P. Guha, A. Dey, R. Sen, M. Chatterjee, S. Chattopadhyay and S. K. Bandyopadhyay, *J. Pharmacol. Exp. Ther.*, 2010, **334**, 381–394; (g) P. Guha, A. Dey, R. Sen, M. Chatterjee, S. Chattopadhyay and S. K. Bandyopadhyay, *J. Pharmacol. Exp. Ther.*, 2011, **336**, 206–214; (h) S. K. Yadav, B. Adhikary, S. Chand, B. Maity, S. K. Bandyopadhyay and S. Chattopadhyay, *Free Radic. Biol. Med.*, 2012, **52**, 1175–1187; (i) B. Maity, S. K. Yadav, B. S. Patro, S. K. Bandyopadhyay and S. Chattopadhyay, *Free Radic. Biol. Med.*, 2012, **52**, 1680–1691.

7 (a) S. Masamune, G. S. Bates and J. W. Corcoran, *Angew. Chem. Int. Ed. Engl.*, 1977, **16**, 585–607; (b) K. C. Nicolaou, *Tetrahedron*, 1977, **33**, 683–710; (c) T. G. Back, *Tetrahedron*, 1977, **33**, 3041–3059; (d) K. Mori, *Tetrahedron*, 1989, **45**, 3233–3298; (e) T. J. Simpson, *Nat. Prod. Rep.*, 1987, **4**, 339–376.

8 For some reviews: (a) P. C. Michels, Y. L. Khmelnitsky, J. S. Dordick and D. S. Clark, *Trends Biotechnol.*, 1998, **16**, 210–215; (b) E. Garcia-Urdiales, I. Alfonso and V. Gotor,

Chem. Rev., 2005, **105**, 313–354; (c) F. van Rantwijk and R. Sheldon, *Chem. Rev*., 2007, **107**, 2757–2785; (d) T. Hudlicky and J. W. Reed, *Chem. Soc. Rev*., 2009, **38**, 3117–3132; (e) C. M. Clouthierzab and J. N. Pelletier, *Chem. Soc. Rev*., 2012, **41**, 1585–1605.

9 (a) J. B. Jones and J. F. Beck, In Application of Biochemical Systems in Organic Chemistry. Part 1; J. B. Jones, C. J. Sih, D. Perlman, Eds.; Wiley: New York, 1976; Vol. X, pp 236–401; (b) S. M. Roberts, A. J. Turner, A. J. Willetts and M. K. Turner, Introduction to Biocatalysis Using Enzymes and Microorganisms; Cambridge University Press: New York, 1995; (c) N. A. Salvi and S. Chattopadhyay, *Tetrahedron*, 2001, **57**, 2833–2839; (d) N. A. Salvi and S. Chattopadhyay, *Bioorg. Med. Chem*., 2006, **14**, 4918–4922; (e) N. A. Salvi and S. Chattopadhyay, *Tetrahedron: Asymmetry*, 2008, **19**, 1992–1997; (f) B. N. Zhou, A. S. Gopalan, F. Van Middlesworth, W. Ru Shieh and C. J. Sih, *J. Am. Chem. Soc*., 1983, **105**, 5925-5926; (g) A. Manzocchi, A. Fiecchi and E. Santaniello, *J. Org. Chem*., 1988, **53**, 4405–4407 and references cited therein; (h) E. Keinan, S. C. Sinha and A. Sinha-Bagchi, *J. Org. Chem*., 1992, **57**, 3631–3637.

10 (a) J. M. Keith, J. F. Larrow and E. N. Jacobsen, *Adv. Synth. Catal*., 2001, **343**, 5–26; (b) J. H. Lee, K. Han, M. -J. Kim and J. Park, *Eur. J. Org. Chem*., 2010, 999–1015; (c) O. Mitsunobu, *Synthesis*, 1981, 1–28; (d) R. Dembinski, *Eur. J. Org. Chem*., 2004, 2763–2772; (e) K. C. Swamy, N. N. Kumar, E. Balaraman and K. V. Kumar, *Chem. Rev*., 2009, **109**, 2551–2651.

11 (a) M. T. Reetz, *Curr. Opin. Chem. Biol*., 2002, **6**, 145–150; (b) K. Faber, *Biotransformations in Organic Chemistry*; Springer: Heidelberg, Germany, 5th Ed. 2004; (c) U. T. Bornscheuer and R. J. Kazlauskas, *Hydrolases in Organic Synthesis*; Wiley-

VCH: Weinheim. Germany, 2nd Ed. 2005; (d) V. Gotor-Fernández, E. Busto and V. Gotor, *Adv. Synth. Catal*., 2006, 797–812.

12 (a) S. Sankaranarayanan, A. Sharma, B. A. Kulkarni and S. Chattopadhyay, *J. Org. Chem.*, 1995, **60**, 4251–4254; (b) A. Sharma, S. Sankaranarayanan and S. Chattopadhyay, *J. Org. Chem.*, 1996, **61**, 1814–1816; (c) A. Sharma and S. Chattopadhyay, *J. Org. Chem*., 1998, **63**, 6128–6131; (d) A. Sharma and S. Chattopadhyay, *J. Org. Chem*., 1999, **64**, 8059–8062; (e) S. Ghadigaonkar, M. R. Koli, S. S. Gamre, M. K. Choudhary, S. Chattopadhyay and A. Sharma, *Tetrahedron: Asymmetry*, 2012, **23**, 1093–1099.

13 (a) A. Sharma, S. Gamre and S. Chattopadhyay, *Tetrahedron: Asymmetry*, 2009, **20**, 1164–1167; (b) A. Sharma, S. Mahato and S. Chattopadhyay, *Tetrahedron Lett*., 2009, **50**, 4986–4988; (c) S. Biswas, S. Chattopadhyay and A. Sharma, *Tetrahedron: Asymmetry*, 2010, **21**, 27–32.

14 For some leading references on olefin metathesis, see: (a) S. K. Armstrong, *J. Chem. Soc. Perkin Trans. 1*, 1998, 371–388; (b) R. R. Schrock, *Tetrahedron*, 1999, **55**, 8141–8153; (c) A. Fürstner, *Angew. Chem. Int. Ed*., 2000, **39**, 3012–3043; (d) T. M. Trnka and R. H. Grubbs, *Acc. Chem. Res*., 2001, **34**, 18–29; (e) H. E. Blackwell, D. J. O'Leary, A. K. Chatterjee, R. A. Washenfelder, D. A. Bussmann and R. H. Grubbs, *J. Am. Chem. Soc.*, 2000, **122**, 58–71.

15 J. A. Dale and S. H. Mosher, *J. Am. Chem. Soc*., 1973, **95**, 512–519.

16 A. Sharma, A. S. Pawar and S. Chattopadhyay, *Synth. Commun*., 1996, **26**, 19–25.

17 R. J. Kazlauskas, A. N. E. Weissfloch, A. T. Rappaport and L. A. Cuccia, *J. Org. Chem.*, 1991, **56***,* 2656–2665.

18 (a) B. D. Martin, S. A. Ampofo, R. J. Linhardt and J. S. Dordick, *Macromol*. 1992, **25**, 7081–7085; (b) L. Haughton, J. M. J. Williams and J. A. Zimmermann, *Tetrahedron: Asymmetry*, 2000, **11**, 1697–1701; (c) V. Athawale and N. Manjrekar, *J. Mol. Catal. B: Enz*., 2000, **10**, 551–554; (d) A. E. V. Petersson, P. Adlercreutz and B. Mattiasson, *Biotechnol. Bioeng*., 2007, **97**, 235–241; (e) M. Nordblad and P. Adlercreutz, *Biotechnol. Bioeng*., 2008, **99**, 1518–1524.

19 E. Sundby, L. Perk, T. Anthonsen, A. J. Aasen and T. V. Hansen, *Tetrahedron*, 2004, **60**, 521–524.

20 K. Arai, B. J. Rawlings, Y. Yoshizawa and J. C. Vederas, *J. Am. Chem. Soc.*, 1989, **111**, 3391–3399.

Table of Contents

Chemoenzymatic Synthesis of the Macrolide Antibiotic (-)-A26771B

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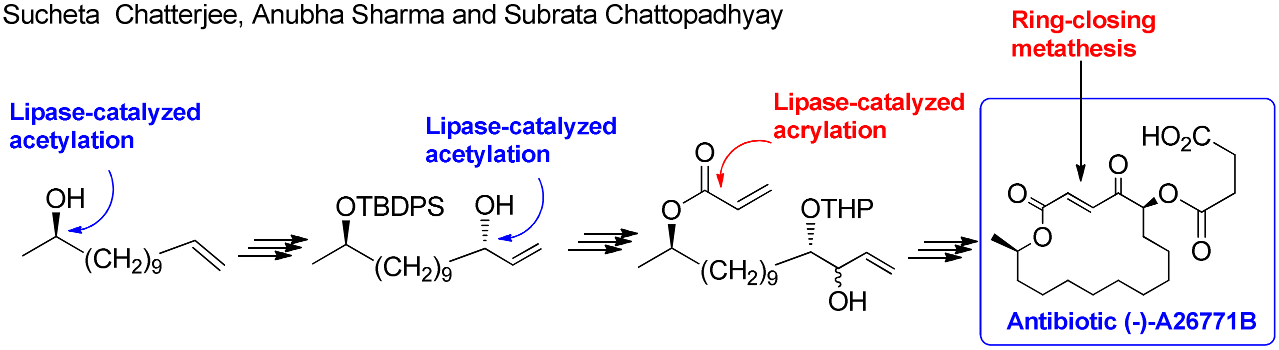

The macrolide antibiotic (-)-A26771B is synthesized using lipase-catalyzed reactions as the key steps.