An Amphiphile–Lipase Aggregate Bearing 1,2-Dialkylated Mannitol Ether as a New Type of Immobilized Enzyme in Organic Solvents

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Neutral amphiphiles of a new type corresponding to 1,2-dialkylated mannitol ethers (6—10) were synthesized. Amphiphile–lipase aggregates were found to function as an immobilized enzyme system for enantioselective acetylation or enantioselective hydrolysis of α-hydroxy esters or α-acetoxy esters, respectively, in organic solvents.

Key words enantioselective acetylation; enantioselective hydrolysis; α-acetoxy ester; thioindolmycin; amphiphile–lipase aggregate

In enzymatic reactions using water-insoluble substrates, the use of immobilized enzymes is often effective in organic media.[1)] Such immobilized enzymes are particularly easy to handle and can be easily recovered from the reaction medium by simple filtration. Furthermore, many immobilized enzymes can be used repeatedly without loss of the enzymatic activity, and sometimes exhibit enhanced stability. In the preceding paper,[2)] we reported that phospholipid–lipase aggregates with ether linkages function effectively as immobilized lipases in asymmetric hydrolysis or esterification reactions in water-saturated organic solvent. The phospholipid analogues A bearing an *O*-ω-cyclohexyltridecyl group as well as an *O*-hexadecyl group have a zwitterion structure and their synthesis requires multiple steps. In order to develop a new type of neutral amphiphile bearing both lipophilic and hydrophilic moieties, the 1,2-dialkylated mannitol ethers B were selected as synthetic targets. We describe here a simple synthesis of 1,2-dialkylated mannitol analogues as a new type of support for immobilized enzymes and their application to enzymatic reactions in organic media.

Synthesis of 1,2-Dialkylated Mannitol Analogues (6—10) i) The reaction of 1,2;3,4-di-*O*-isopropylidene-D-mannitol (**1**)[3)] and alkyl bromide (1-bromododecane, 1-bromotetradecane, 1-bromohexadecane, 1-bromooctadecane) in the presence of KOH at reflux in toluene gave the corresponding 5,6-dialkylated ethers (**2**; 47% yield, **3**; 98% yield, **4**; 87% yield, **5**; 82% yield), which were treated with 10% HCl in tetrahydrofuran (THF) to afford the corresponding 1,2-dialkylated mannitol analogues (**6**; 59% yield, **7**; 56% yield, **8**; 64% yield, **9**; 57% yield).

ii) The 12-bromododecanol derivative **11**[2c)] was reacted with the diol 1 under the same reaction conditions as in the previous cases to afford the tetrahydropyranyl ether **12** in 48% yield. Removal of the tetrahydropyranyl group of **12** by using camphorsulfonic acid (CSA) afforded a diol **13** in 81% yield. Pyridinium chlorochromate (PCC) oxidation of **13** gave a dialdehyde **14** in 82% yield. Wittig reaction of **14** and cyclohexylmethyltriphenylphosphonium salt, which was obtained by the reaction of cyclohexylmethyl bromide and Ph_3P, produced a mixture of *cis* and *trans* isomers **15** in 56% yield. Removal of the diacetonide in **15** followed by catalytic hydrogenation in the presence of 20% $Pd(OH)_2$–C gave the desired diol **10** in 75% overall yield.

Enantioselective Acetylation by Using Amphiphile–Lipase Aggregates In order to investigate enantioselective acetylation of α-hydroxy ester by using amphiphile–lipase aggregates, (±)-indolmycenic ester **16** was chosen as a suitable substrate, because its optically active congener, the (2*S*,3*R*)-α-acetoxy ester **17**, is an important chiral intermediate for the synthesis of pharmacologically and optically active indolmycin.[4)]

In the preceding paper,[2c)] it was shown that a 1 : 1 mixture of two racemates ((±)-**16** and (±)-**17**) was well separated by high-performance liquid chromatographic (HPLC) analysis with a chiral column (thiracel OD (4.6×250 mm)). Thus, it is feasible to determine the chemical and optical yields and absolute structure of

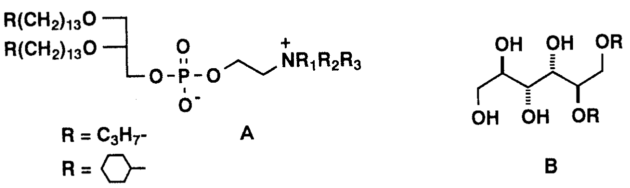

Chart 1

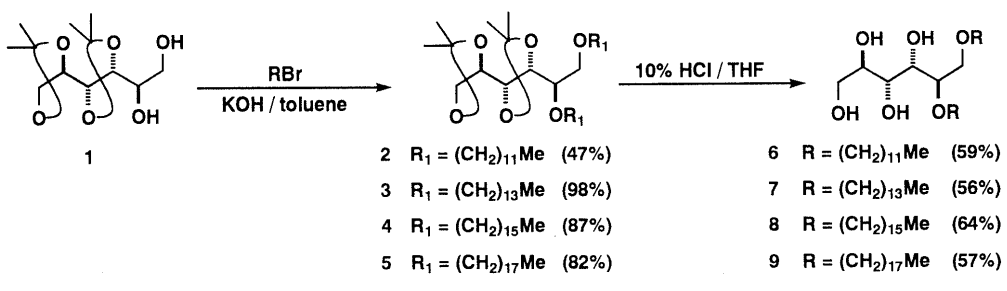

Chart 2

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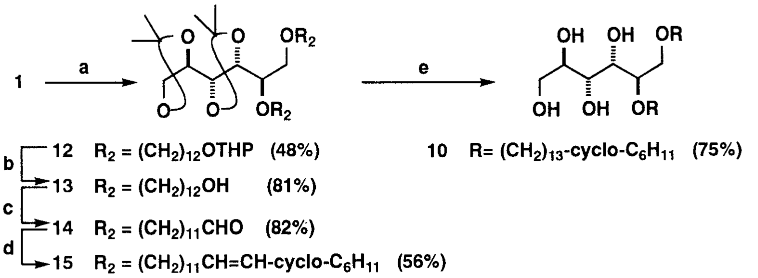
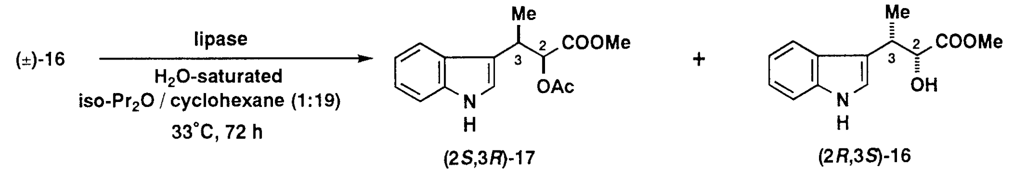


a; $THPO(CH_2)_{12}Br$(11) / KOH / toluene b; CSA / CH_2Cl_2 /MeOH c; PCC / CH_2Cl_2
d; cyclo-$C_6H_{11}CH_2P^+Ph_3Br^-$ / *n*-BuLi / THF e; 1) 10% HCl / MeOH / THF 2) H_2 / $Pd(OH)_2$ / AcOEt / MeOH

Chart 3

Table 1. Enantioselective Acetylation Using 10 mg of (±)-**16**



Entry	Lipase	Acylating reagent (mg)	Product (yield (%), Optical purity (% ee)) (2*S*,3*R*)-17	(2*R*,3*S*)-16
1	OF-360	A (20)	8.2 (>99)	83.7 (10)
2	OF-360	B (20)	8.3 (>99)	82.2 (11)
3	OF-360	C (20)	7.6 (>99)	78.5 (9)
4	OF-360/C (12)	A (20)	6.7 (>99)	85.8 (10)
5	OF-360/C (12)	B (20)	16.2 (>99)	81.0 (23)
6	OF-360/C (12)	C (20)	2.4 (>99)	92.0 (4)
7	OF-360/C (14)	A (20)	10.0 (>99)	88.1 (10)
8	OF-360/C (14)	B (20)	7.5 (>99)	90.2 (9)
9	OF-360/C (14)	C (20)	2.8 (>99)	96.1 (4)
10	OF-360/C (16)	A (20)	10.8 (>99)	87.2 (13)
11	OF-360/C (16)	B (20)	13.9 (>99)	85.6 (16)
12	OF-360/C (16)	C (20)	12.4 (99)	83.4 (16)
13	OF-360/C (18)	A (20)	12.4 (>99)	83.9 (16)
14	OF-360/C (18)	B (20)	19.7 (98)	78.4 (27)
15	OF-360/C (18)	C (20)	5.5 (92)	92.2 (3)

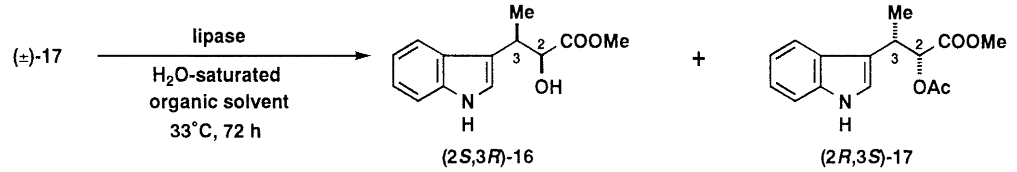
A, isopropenyl acetate. B, phenyl thioacetate. C, *o*-nitrophenyl acetate.

each reaction product. For the purpose of finding a suitable acetylating reagent, a screening experiment using 10 mg of substrate (±)-**16** and lipase "OF-360" itself (50 mg) was carried out in a mixed solvent (iso-Pr_2O/cyclohexane = 1/19). In every case, optically pure (2*S*)-acetate **17** was obtained in *ca.* 8% yield and it was concluded that the acylating reagent has little or no influence on the enantioselective esterification (Table 1, entries 1—3). On the other hand, the chemical yield of (2*S*)-**17** was found to be influenced by the amphiphile–lipase aggregate and acylating reagent used. The use of phenylthioacetate and C(18)[5]-aggregate gave the best result ((2*S*)-**17**; 19.7% yield, 98% ee) was obtained (Table 1, entry 14).

Enantioselective Hydrolysis by Using Amphiphile–Lipase Aggregates The influence of solvents on enantioselective hydrolysis was studied by using 100 mg of substrate (±)-**17** and lipase "OF-360" itself (50 mg), and a mixed solvent system (iso-Pr_2O/cyclohexane = 1/19) was found to provide the best conversion yield ((2*S*)-**16**; 42% yield, Table 2, entry 3). When amphiphile–lipase aggregates were employed instead of native lipase "OF-360", the conversion yield of the (2*S*)-hydroxy ester **16** slightly decreased, while the optical purity of (2*S*)-**16** was maintained at more than 97% ee in every case (Table 2, entries 4—14). When the recovered aggregates were used repeatedly, the yield and optical purity of the reaction products were maintained at the same levels (Table 2, entries 5, 7, 9, 10, 12, 14) as the first cases (Table 2, entries 4, 6, 8, 11, 13). These observations indicate that the amphiphile–lipase aggregates can function as an immobilized enzyme system.

Finally, (2*S*,3*R*)-indolmycenate (**16**) obtained in 97% ee was converted into the (−)-(5*S*)-((1*R*)-indol-3-ylethyl)-2-methylamino-Δ^2-thiazolin-4-one ((1*R*,5*S*)-thioindolmycin) (**18**) *via* the (2*S*)-2-chloro ester (**19**). Treatment of (2*S*,3*R*)-**16** with MsCl in pyridine gave (2*S*)-**19** in 60% yield with complete retention of C_2-stereochemistry because of double inversion along with neighbouring-group participation involving the electron-rich C-3 of the indole ring.[6] The reaction of (2*S*)-**19** and methyl thiourea followed by treatment with 1 N NaOH in MeOH afforded the (1*R*,5*S*)-thioindolmycin (**18**) (mp 233—235 °C, $[\alpha]_D$

Table 2. Enantioselective Hydrolysis Using 100 mg of (±)-**17**



Entry	Lipase (mg)	Organic solvent	Product (yield (%), Optical purity (% ee)) (2*S*,3*R*)-16	(2*R*,3*S*)-17
1	OF-360 (50)	A	25 (95)	73 (30)
2	OF-360 (50)	B	20 (97)	79 (24)
3	OF-360 (50)	C	42 (93)	57 (69)
4	OF-360/C (12) (30)	C	19 (97)	80 (22)
5[a]	OF-360/C (12)	C	20 (97)	76 (27)
6	OF-360/C (14) (74)	C	39 (97)	57 (72)
7[a]	OF-360/C (14)	C	27 (98)	71 (38)
8	OF-360/C (16) (74)	C	37 (98)	59 (66)
9[a]	OF-360/C (16)	C	41 (97)	56 (71)
10[a]	OF-360/C (16)	C	33 (98)	65 (50)
11	OF-360/C (18) (71)	C	29 (97)	69 (42)
12[a]	OF-360/C (18)	C	22 (98)	77 (29)
13	OF-360/Cy (52)	C	28 (97)	69 (40)
14[a]	OF-360/Cy	C	26 (97)	71 (37)

A, iso-Pr$_2$O. B, CCl$_4$. C, iso-Pr$_2$O–cyclohexane (1 : 19). [a] Repeated experiment using the recovered aggregate.

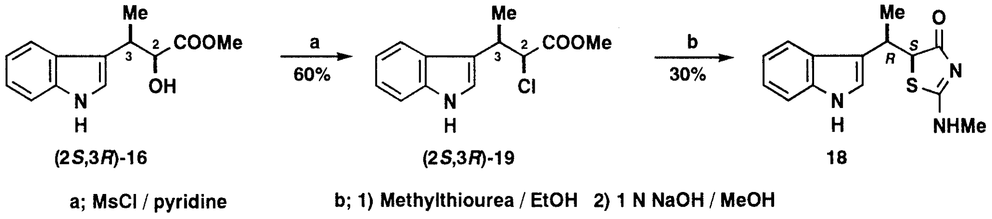


Chart 4

−101.6° ($c=0.3$, MeOH)) in 30% overall yield. This product appeared to be identical with the reported (1*R*,5*S*)-**18**[6] (mp 225—227 °C, $[\alpha]_D$ −100.7° ($c=0.3$, MeOH)).

In conclusion, the present 1,2-alkylated mannitol analogues were found to function as supports for enzyme immobilization and the newly prepared amphiphile–lipase aggregates were ascertained to be useful as immobilized lipase. Further investigations toward on other enzymatic reactions, such as glycosidation in water media, are warranted.

Experimental

All melting points were measured on a Yanaco MP-S3 micro melting point apparatus and are uncorrected. NMR spectra were measured on a JEOL GX-4000 spectrometer and spectra were taken as 5—10% (w/v) solutions in CDCl$_3$ with Me$_4$Si as an internal reference. IR spectra were measured on a JASCO A-3 spectrometer. FAB-MS were obtained with a JEOL JMS-HS 100 instrument. Optical rotations were measured on a Perkin-Elmer model 241 MC polarimeter. The HPLC system was composed of two SSC instruments (ultraviolet (UV) detector 3000B and flow system 3100). All the reactions were carried out in an atmosphere of argon. All evaporations were performed under reduced pressure.

The Synthesis of 1,2-Dialkylmannitol Analogues. (1) Synthesis of 1,2-Didodecylmannitol (6) i) A mixture of **1** (4 mmol, 1.048 g), dodecyl bromide (12 mmol, 3.0 g) and KOH (40 mmol, 2.24 g) in toluene (50 ml) was refluxed for 3 h with stirring. The reaction mixture was diluted with brine and extracted with AcOEt. The organic layer was dried over MgSO$_4$ and evaporated to give an oil, which was chromatographed on silica gel (60 g) to afford a homogeneous oil **2** (1.125 g, 47% yield) from the *n*-hexane–ethyl acetate (19 : 1) eluate. ii) A mixture of **2** (1.7 mmol, 1.023 g) and 10% aqueous HCl (5 ml) in THF (5 ml) was heated at 80 °C for 2.5 h with stirring. The reaction mixture was poured into saturated aqueous NaHCO$_3$, and extracted with CHCl$_3$. The organic layer was washed with brine and dried over MgSO$_4$. Removal of the organic solvent gave a semicrystalline compound, which was chromatographed on silica gel (50 g) to provide crystalline **6** from the ethyl acetate eluate. Recrystallization from CHCl$_3$ provided **6** as colorless plates (521 mg, 59% yield). **6**: mp 80.5—82 °C. *Anal.* Calcd for C$_{30}$H$_{62}$O$_6$: C, 69.45; H,12.05. Found: C, 69.53; H,12.15. NMR δ: 0.88 (6H, t, $J=6.8$ Hz, 2×Me), 1.20—1.60 (40H, m, methylene), 3.42—3.92 (12H, m, 4×CH$_2$–O, 4×CH–O).

(2) Synthesis of 1,2-Ditetradecylmannitol (7) i) A mixture of **1** (26 mmol, 6.9 g), tetradecyl bromide (78 mmol, 21.9 g) and KOH (260 mmol, 14.8 g) in toluene (100 ml) was refluxed for 3 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **2** to afford a homogeneous oil **3** (16.82 g, 98% yield). ii) A mixture of **3** (25.7 mmol, 16.82 g) and concentrated HCl (10 ml) in THF (10 ml) was heated at 80 °C for 1 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **6** to provide crystalline **7**. Recrystallization from CHCl$_3$ provided **7** as colorless plates (8.3 g, 56% yield). **7**: mp 75—81 °C. *Anal.* Calcd for C$_{34}$H$_{70}$O$_6$: C, 71.03; H, 12.27. Found: C, 70.53; H, 12.32. NMR δ: 0.88 (6H, t, $J=6.8$ Hz, 2×Me), 1.20—1.60 (48H, m, methylene), 3.4—3.94 (12H, m, 4×CH$_2$–O, 4×CH–O).

(3) Synthesis of 1,2-Dihexadecylmannitol (8) i) A mixture of **1** (30.6 mmol, 8.03 g), hexadecyl bromide (92 mmol, 28.1 g) and KOH (306 mmol, 17.2 g) in toluene (150 ml) was refluxed for 4.5 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **2** to afford a homogeneous oil **4** (18.86 g, 87% yield). ii) A mixture of **4** (26.5 mmol, 18.86 g) and concentrated HCl (10 ml) in

THF (10 ml) was heated at 80 °C for 1 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **7** to provide crystalline **8**. Recrystallization from $CHCl_3$ provided **8** as colorless plates (10.68 g, 64% yield). **8**: mp 83—87 °C. *Anal.* Calcd for $C_{38}H_{78}O_6$: C, 72.33; H, 12.46. Found: C, 71.79; H, 12.52. NMR δ: 0.88 (6H, t, $J=6.8$ Hz, 2 × Me), 1.20—1.60 (56H, m, methylene), 3.48—3.92 (12H, m, 4 × CH_2–O, 4 × CH–O).

(4) Synthesis of 1,2-Dioctadecylmannitol (9) i) A mixture of **1** (19.2 mmol, 5.04 g), octadecyl bromide (57 mmol, 19.2 g) and KOH (192 mmol, 10.8 g) in toluene (80 ml) was refluxed for 3 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **2** to afford a homogeneous oil **5** (12.11 g, 82% yield). ii) A mixture of **5** (15.8 mmol, 12.11 g) and concentrated HCl (10 ml) in THF (10 ml) was heated at 80 °C for 1 h with stirring. The reaction mixture was worked up in the same way as in the case of the preparation of **8** to provide crystalline **9**. Recrystallization from $CHCl_3$ provided **9** as colorless plates (6.15 g, 57% yield). **9**: mp 89.5—92 °C. *Anal.* Calcd for $C_{42}H_{86}O_6$: C, 73.41; H, 12.62. Found: C, 73.12; H, 12.72. NMR δ: 0.88 (6H, t, $J=6.8$ Hz, 2 × Me), 1.20—1.60 (64H, m, methylene), 3.42—3.92 (12H, m, 4 × CH_2–O, 4 × CH–O).

(5) Synthesis of 1,2-di-*O*-ω-Cyclohexyltridecylmannitol (10) i) A mixture of **1** (27.5 mmol, 7.2 g), the 12-bromododecanol derivative (**11**) (69 mmol, 24 g) and KOH (553 mmol, 31 g) in toluene (300 ml) was refluxed for 24 h with stirring. The reaction mixture was diluted with brine and extracted with AcOEt. The organic layer was dried over $MgSO_4$ and evaporated to give an oil, which was chromatographed on silica gel (200 g) to afford **12** as a homogeneous oil (10.6 g, 48% yield) from the *n*-hexane–ethyl acetate (20 : 1) eluate. **12**: FAB-MS *m/z*: 799 ($M^+ + 1$). NMR δ: 4.58 (2H, t, $J=3.4$ Hz, 2 × anomeric-H). ii) A mixture of **12** (2.9 mmol, 2.3 g) and CSA (0.39 mmol, 91 mg) in a mixed solvent (CH_2Cl_2 (30 ml) and MeOH (30 ml)) was stirred for 11 h at room temperature. The reaction mixture was diluted with saturated aqueous $NaHCO_3$ and extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$. Removal of the solvent gave an oily product **12**, which was treated with acetone (250 ml) in the presence of 60% $HClO_4$ (0.5 ml) for 10 min at room temperature because partial deketalization of **12** had occurred as judged from NMR analysis. The reaction mixture was treated with powdered $NaHCO_3$ (3 g), diluted with brine and extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$. Removal of the solvent gave an oily product, which was chromatographed on silica gel (60 g) to afford **13** as a homogeneous oil (1.45 g, 81% yield) from the *n*-hexane–ethyl acetate (3 : 1) eluate. **13**: FAB-MS *m/z*: 631 ($M^+ + 1$). IR: 3430, 3367 cm^{-1}. NMR: No characteristic signals. iii) A mixture of **13** (9.7 mmol, 6.15 g) and PPC (29 mmol, 6.3 g) in CH_2Cl_2 (150 ml) was stirred for 1 h at room temperature. The reaction mixture was directly chromatographed on Fluorisil (150 g) to afford **14** as a homogeneous oil (5 g, 82% yield) from the CH_2Cl_2 eluate. **14**: FAB-MS *m/z*: 627 ($M^+ + 1$). IR: 1728 cm^{-1}. NMR δ: 9.76 (2H, t, $J=2$ Hz, CHO). vi) *n*-BuLi/*n*-hexane solution (1.5 M; 2.3 ml) was added to a solution of cyclohexylmethyl phosphonium bromide[2c)] (3.4 mmol, 1.5 g) in dry THF (10 ml) with stirring at 0 °C. The mixture was stirred for 30 min, then a solution of **14** (1.1 mmol, 0.71 g) in dry THF (2 ml) was added. The reaction mixture was stirred for 30 min at 0 °C, then diluted with water and AcOEt. The organic layer was washed with brine and dried over $MgSO_4$. Evaporation of the solvent gave a crude product, which was chromatographed on silica gel (50 g) to provide **15** as a homogeneous oil (0.5 g, 56% yield) from the *n*-hexane : AcOEt (400 : 14) eluate. **14**: FAB-MS *m/z*: 787 ($M^+ + 1$). IR: 1645 cm^{-1}. NMR δ: 5.16—5.34 (4H, m, olefinic-H). v) A mixture of **15** (3.5 mmol, 2.8 g) and 10% aqueous HCl (6 ml) in a mixed solvent (MeOH (15 ml) and THF (20 ml)) was heated at 80 °C for 6 h with stirring. The reaction mixture was poured into saturated aqueous $NaHCO_3$, and extracted with $CHCl_3$. The organic layer was washed with brine and dried over $MgSO_4$. Removal of the organic solvent gave a crude product. A mixture of this crude compound and 20% $Pd(OH)_2$–C (200 mg) in a mixed solvent (AcOEt (20 ml) and MeOH (20 ml)) was subjected to catalytic hydrogenation at ordinary temperature and the reaction mixture was filtered with the aid of Celite. The filtrate was evaporated to give a crude product, which was chromatographed on silica gel (60 g) to provide crystalline **10** (1.9 g, 75% overall yield) from the $CHCl_3$–MeOH (15 : 1) eluate. **10**: mp 82—83 °C. *Anal.* Calcd for $C_{44}H_{86}O_6 \cdot 1/4H_2O$: C, 73.84; H, 12.18. Found: C, 73.83; H, 12.34. FAB-MS *m/z*: 711 ($M^+ + 1$). IR (KBr): 3367, 3249 cm^{-1}. NMR δ: 0.8—1.80 (70H, m, methylene and methine), 3.4—3.92 (12H, m, 4 × CH_2–O, 4 × CH–O).

Preparation of Amphiphile–Lipase Aggregates Bearing 1,2-Dialkylated Mannitol Analogues (6—10) A mixture of 50 mg of lipase "OF-360" from *Candida rugosa* in water (2.5 ml) and 50 mg of a 1,2-dialkylated mannitol analogue in benzene (40 ml) was sonicated for 30 min at 0 °C. The resulting precipate was centifugated at 3000 × *g* and the solvent was decanted off. The residual precipate was lyophilized to provide an amorphous powder (OF-360/C(12)[5)]; 30 mg, OF-360/C(14)[5)]; 74 mg, OF-360/C(16)[5)]; 74 mg, OF-360/C(18)[5)]; 71 mg and OF-360/Cy[5)]; 52 mg).

General Procedure of Enantioselective Acetylation A mixture of (±)-**16** (10 mg), an acetylating reagent (see Table 1) and amphiphile–lipase aggregates (10 mg) in a mixed solvent (iso-Pr_2O (0.25 ml) and cyclohexane (4.75 ml)) was shaken at 33 °C for 3 d. The reaction mixture was dried over $MgSO_4$ and evaporated to afford a crude product, which was analyzed by HPLC. The results are shown in Table 1.

General Procedure of Enantioselective Hydrolysis A mixture of (±)-**17** (100 mg) and amphiphile–lipase aggregates (see Table 2) in water-saturated mixed solvent (iso-Pr_2O (2.5 ml) and cyclohexane (47.5 ml)) was stirred at 33 °C for 3 d. The reaction mixture was dried over $MgSO_4$ and evaporated to afford a crude product, which was subjected to silica gel (30 g) column chromatography. The fraction with *n*-hexane : AcOEt (9 : 1, V/V) gave (2*R*,3*S*)-**17**. The second fraction eluted with *n*-hexane : AcOEt (7 : 1, V/V) provided (2*S*,3*R*)-**16**. Both fractions were analyzed by HPLC and the results are given in Table 2.

(−)-(5*S*)-((1*R*)-Indol-3-ylethyl)-2-methylamino-Δ^2-thiazolin-4-one (18) A solution of (2*S*,3*R*)-**16** (1.4 mmol, 330 mg) in pyridine (2 ml) was treated with MsCl (2.8 mmol, 0.22 ml) at 0 °C and the reaction mixture was allowed to stand for 3 d at 5 °C. The reaction mixture was diluted with water and extracted with ether. The ether layer was washed with 10% aqueous HCl and brine, and dried over $MgSO_4$. Evaporation of the solvent gave crude (2*S*,3*R*)-**19**. A solution of crude **19** in EtOH (30 ml) was treated with methylthiourea (1.4 mmol, 129 mg) and the mixture was stirred for 8 d at room temperature. After evaporation of EtOH, a mixture of a residue and 1 N NaOH (0.2 ml) in MeOH (30 ml) was refluxed for 13 h. After evaporation of the solvent, the residue was directly chromatographed on silica gel (20 g) to afford crystalline **18** (116 mg, 30% yield) from the AcOEt–MeOH (10 : 1) eluate. Recrystallization from AcOEt gave **18** as colorless needles. **18**: mp 233—235 °C. *Anal.* Calcd for $C_{14}H_{15}N_3OS$: C, 61.5; H, 5.5; N, 15.4 Found: C, 61.63; H, 5.45; N, 15.11. FAB-MS *m/z*: 274 ($M^+ + 1$). IR (KBr): 3250, 1680, 1608 cm^{-1}. $[\alpha]_D^{24}$ −101.6° ($c=0.33$, MeOH). NMR (DMSO-d_6) δ: 1.21 (3H, d, $J=6.8$ Hz, C-Me), 2.86 (0.6H, s, C=NMe), 2.95 (2.4H, d, $J=4.4$ Hz, $NH\underline{Me}$), 3.88 (1H, m, $C\underline{H}$-Me), 4.79 (1H, d, $J=3.4$ Hz, CH-S), 7.01 (1H, t, $J=7$ Hz, aromatic-H), 7.09 (1H, t, $J=7$ Hz, aromatic-H), 7.12 (1H, d, $J=2$ Hz, aromatic-H), 7.36 (1H, d, $J=8$ Hz, aromatic-H), 7.58 (1H, d, $J=8$ Hz, aromatic-H), 9.12 (0.8H, q, $J=4.4$ Hz, $N\underline{H}Me$), 9.46 (0.2H, br s, NH), 10.92 (1H, s, indol NH).

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References and Notes

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