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Note

Cancer oriented ketonucleosides: synthesis of 7-[6-*O*-(5-carboxypentyl)-3,4-dideoxy- and 3,4-dideoxy-6-*O*-(6-hydroxyhexyl)-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophyllines and their coupling with cancer-specific proteins

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A major problem in cancer chemotherapy is the lack of sufficient tumour selectivity of cytotoxic agents which might be circumvented by conjugation of drugs to antibodies to tumour-associated antigens[1]. Another approach[2] uses the ability of alpha-fetoprotein (AFP) to bind polyunsaturated fatty acids strongly and selectively, even in the presence of high concentrations of serum albumin, and the properties of various cancer cells which have specific AFP receptors that are absent from normal cells[3]. Uptake of the AFP–fatty acid complex delivers fatty acid to the cell, and the delipidated AFP released by the cell is able to transport and concentrate other fatty acid molecules inside the cell.

We have reported the synthesis and the significant antitumour activity of unsaturated ketonucleosides[4] and recently we have initiated the synthesis of derivatives of unsaturated ketonucleosides each carrying a spacer arm of various lengths which could be linked to tumour-specific antibodies or to AFP using polyunsaturated fatty acid derivatives as intermediate drug carriers[5].

We now describe the preparation of 7-[6-*O*-(5-carboxypentyl)-3,4-dideoxy-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline (**11**) and 7-[3,4-dideoxy-6-*O*-(6-hydroxyhexyl)-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline (**16**), and their coupling with proteins and polyenic fatty acids.

1,2:3,4-Di-*O*-isopropylidene-α-D-galactose was treated with sodium hydride and 1-bromo-6-pyranyloxyhexane to yield the 6-*O*-(6-tetrahydropyranyloxyhexyl) derivative **1**, hydrolysis of which with methanolic 0.1M hydrochloric acid afforded crystalline 6-*O*-(6-hydroxyhexyl)-α,β-D-galactose (**2**). Condensation of the syrupy tetra-acetate (**3**) of **2** with trimethylsilyltheophylline, using stannic chloride as catalyst[6], yielded 7-[6-*O*-(6-acetoxyhexyl)-2,3,4-tri-*O*-acetyl-β-D-galactopyranosyl]theophylline (**4**), which was deacetylated to give 7-[6-*O*-(6-hydroxyhexyl)-β-D-galactopyranosyl]theophylline (**5**). Treatment of **5** with 2,2-dimethoxypropane in acetone afforded the 3,4-*O*-isopropylidene derivative **6**, which was converted into the dibenzoate **7**. Deacetalation of **7** with formic acid in dichloromethane then af-

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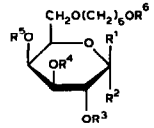
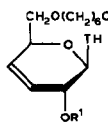
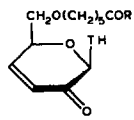
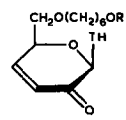








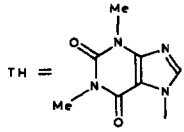
1 $R^1 = H, R^2, R^3 = R^4, R^5 = Me_2C, R^6 = THP$
2 $R^1, R^2 = H, OH, R^3 = R^4 = R^5 = R^6 = H$
3 $R^1, R^2 = H, OAc, R^3 = R^4 = R^5 = R^6 = Ac$
4 $R^1 = TH, R^2 = H, R^3 = R^4 = R^5 = R^6 = Ac$
5 $R^1 = TH, R^2 = R^3 = R^4 = R^5 = R^6 = H$
6 $R^1 = TH, R^2 = R^3 = R^6 = H, R^4, R^5 = Me_2C$
7 $R^1 = TH, R^2 = H, R^3 = R^6 = Bz, R^4, R^5 = Me_2C$
8 $R^1 = TH, R^2 = R^4 = R^5 = H, R^3 = R^6 = Bz$

9 $R^1 = R^2 = Bz$
10 $R^1 = R^2 = H$
14 $R^1 = H, R^2 = Tr$

11 R = OH
12 R = O–N
13 R = NHIgG

15 R = Tr
16 R = H
17 R = $OCC_{21}H_{31}$

TH =



THP =

forded 7-[2-*O*-benzoyl-6-*O*-(6-benzoyloxyhexyl)-β-D-galactopyranosyl]theophylline (**8**).

Reaction of **8** with iodoform–triphenylphosphine–imidazole in a one-pot procedure[7] gave 70% of the olefinic compound **9**. Deoxygenation at C-3′,4′ *via* the cyclic thionocarbonate[8] or the 1-dimethylamino(methylene)acetal[9] derivatives could also be effected, but only moderate yields of **9** were obtained. The ^{1}H-n.m.r. spectrum of **9** contained, *inter alia*, a two-proton multiplet at δ 5.8–6.2 for the olefinic protons H-3′,4′. Debenzoylation of **9** gave 7-[3,4-dideoxy-6-*O*-(6-hydroxyhexyl)-β-D-*erythro*-hex-3-enopyranosyl]theophylline (**10**), the common precursor of ketonucleosides having different functional groups at the spacer arm.

Thus, simultaneous oxidation of the primary and allylic hydroxyl groups of **10** with pyridinium dichromate in *N*,*N*-dimethylformamide[10] afforded 71% of 7-[6-*O*-(5-carboxypentyl)-3,4-dideoxy-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline (**11**). In the ^{1}H-n.m.r. spectrum of **11**, the signals for H-3′,4′ appeared markedly downfield at δ 7.23 ($J_{3,4}$ 10, $J_{4,5}$ 1.5 Hz) and 6.4 ($J_{3,5}$ 2 Hz), those for H-1′ and H-5′ at δ 6.63 ($J_{1,5}$ 1.5 Hz) and 4.9, respectively. Irradiation of the H-5′ resonance caused the H-1′ doublet to collapse to a singlet, indicating a six-bond coupling between the two protons. Furthermore, the downfield shift of a methylene signal (δ 2.35) indicated it to be adjacent to the carboxyl group.

Compound **11** could be linked through its side-arm to tumour specific antibodies without affecting the unsaturated ketonucleoside structure. Studies of the covalent binding of methotrexate to immunoglobulins[1c] showed that the active ester method was the most effective and yielded conjugates with retention of both antibody activity and dihydrofolate reductase-inhibitory capacity. Treatment of **11** with *N*-hydroxysuccinimide and dicyclohexylcarbodi-imide in anhydrous 1,4-dioxane gave 65% of the active ester **12**, the structure of which was confirmed by ^{1}H-n.m.r. spectroscopy (signals for four additional methylene protons at δ 2.97), and which was used immediately for the coupling reaction. Human immunoglobulins (IgG) were treated with 10–240 mol of **12**, resulting in the introduction of 4–30 mol of

TABLE I

COVALENT BINDING OF **11** TO IgG

Active ester–IgG molar ratio	10	30	70	140	240
Bound **11**–IgG molar ratio	4	10	19	25	30
Protein recovery (%)	77	75	70	80	70

unsaturated ketonucleoside per mol of IgG with 70–80% recovery of the protein (Table I).

Tritylation of **10** gave **14**, which was oxidised with the pyridinium dichromate–molecular sieve system[11] in dichloromethane, yielding the unsaturated ketonucleoside **15** in its partly hydrated form. Detritylation of **15** with formic acid in ether[12] afforded 7-[3,4-dideoxy-6-*O*-(6-hydroxyhexyl)-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline (**16**), the ^{1}H-n.m.r. spectrum of which was essentially the same as that of **11** except for the absence of the methylene signal at δ 2.35.

Treatment of **16** with docosahexaenoic acid in the presence of dicyclohexylcarbodi-imide and dimethylaminopyridine[13] gave 7-[3,4-dideoxy-6-*O*-(6-docosahexa-4,7,10,13,16,19-enoyloxyhexyl)-β-D-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline (**17**).

As compared to the ^{1}H-n.m.r. spectrum of **16**, that of **17** contained, *inter alia*, signals for 12 additional olefinic protons centered at δ 5.42, as well as signals for 14 allylic protons centered at δ 2.85 and 2.38. The ester **17** was immediately complexed with delipidated AFP. The biological activity of these conjugated unsaturated ketonucleosides is under investigation.

EXPERIMENTAL

General methods. — U.v. spectra were recorded with a Varian UV-VIS M 635 spectrophotometer and ^{1}H-n.m.r. spectra (internal Me_4Si) with a Varian T-60 instrument. Optical rotations were determined with a Roussel–Jouan Quick polarimeter. Melting points are uncorrected. T.l.c. was performed on Silica Gel 60 F_{254} (Merck), and Silica Gel 60 (230–400 mesh, Merck) was used for flash chromatography[14], with ethyl acetate (*A*), and 7:3 (*B*) and 5:5 (*C*) ethyl acetate–hexane.

1,2:3,4-Di-O-isopropylidene-6-O-[6-(tetrahydropyran-2-yloxy)hexyl]-D-galactopyranose (**1**). — A mixture of 1,2:3,4-di-*O*-isopropylidene-α-D-galactopyranose (22 g, 84.6 mmol) and sodium hydride (6.14 g, 256 mmol) in dry *N,N*-dimethylformamide (245 mL) was stirred under nitrogen for 30 min at room temperature and then for 30 min at 80°. 2-(6-Chlorohexyloxy)tetrahydropyran (44 g, 198 mmol) was added dropwise and stirring was continued for 30 min at 80°. Excess of reagents was destroyed with methanol (30 mL), and the mixture was diluted with dichloromethane, washed with 1:1 aqueous 10% NH_4Cl and saturated NaCl, dried (Na_2SO_4), and concentrated. Flash chromatography (continuous gradient from

hexane to solvent *C*) of the residue yielded **1**, isolated as a syrup (26.4 g, 70%), $[\alpha]_D^{22}$ −39° (*c* 0.15, chloroform).

Anal. Calc. for $C_{23}H_{40}O_8$: C, 62.16; H, 9.01. Found: C, 61.66; H, 9.01.

6-O-(6-Hydroxyhexyl)-D-galactose (**2**). — A solution of **1** (24.42 g, 55 mmol) in methanol (60 mL) and 0.1M hydrochloric acid (600 mL) was stirred for 1 h at 90°, then cooled, neutralised with Amberlite IR-45 (HO^-) resin, filtered, and concentrated. The residue was recrystallised from methanol, to yield **2** (13.45 g, 87%), m.p. 119–121°, $[\alpha]_D^{22}$ +67° (*c* 0.15, methanol).

Anal. Calc. for $C_{12}H_{24}O_7$: C, 51.28; H, 8.89. Found: C, 51.34; H, 8.64.

6-O-(6-Acetoxyhexyl)-1,2,3,4-tetra-O-acetyl-α,β-D-galactose (**3**). — Treatment of **2** (13.2 g, 47.1 mmol) with acetic anhydride (50 mL) in pyridine (80 mL) and flash chromatography (solvent *C*) of the product gave **3**, isolated as a syrup (19.6 g, 40 mmol), $[\alpha]_D^{22}$ +26° (*c* 0.15, chloroform), α,β-ratio ~15:85.

Anal. Calc. for $C_{22}H_{34}O_{12}$: C, 53.88; H, 6.94. Found: C, 53.42; H, 6.91.

7-[6-O-(6-Acetoxyhexyl)-2,3,4-tri-O-acetyl-β-D-galactopyranosyl]theophylline (**4**). — A mixture of **3** (13 g, 26.53 mmol), theophylline (4.78 g, 26.53 mmol), hexamethyldisilazane (2.23 mL, 10.61 mmol), and chlorotrimethylsilane (1.35 mL, 10.61 mmol) in acetonitrile (260 mL) was treated with $SnCl_4$ (2 mL) for 4 h at 80°. The mixture was then diluted with dichloromethane, neutralised with saturated aqueous $NaHCO_3$, filtered, washed with water, dried, and concentrated. Flash chromatography (solvent *C*) of the residue yielded **4** (9 g, 56%), isolated as a syrup, $[\alpha]_D^{22}$ +7° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 279 nm (ε 7400). ^{1}H-N.m.r. data [$(CD_3)_2CO$]: δ 8.16 (s, 1 H, H-8), 6.33 (d, 1 H, *J* 8.5 Hz, H-1′), 5.80 (t, 1 H, *J* 10 Hz, H-2′), 5.56 (dd, 1 H, *J* 1.5 Hz, H-4′), 5.43 (dd, 1 H *J* 3.3 Hz, H-3′), 4.48 (m, 1 H, *J* 6 Hz, H-5′), 4.0 [m, 2 H, $(CH_2)_5CH_2OAc$], 3.8–3.4 [m, 4 H, H-6′,6′ and $CH_2(CH_2)_5OAc$], 3.51 and 3.33 (2 s, 6 H, NMe-1,3), 2.2–1.85 (4 s, 12 H, 4 OAc), 1.7–1.1 [m, 8 H, $CH_2(CH_2)_4CH_2$].

Anal. Calc. for $C_{27}H_{38}N_4O_{12}$: C, 53.11; H, 6.23; N, 9.18. Found: C, 53.05; H, 6.46; N, 8.96.

7-[6-O-(6-Hydroxyhexyl)-β-D-galactopyranosyl]theophylline (**5**). — A solution of **4** (8.8 g, 14.43 mmol) in dry methanol (1 L) was treated with methanolic 1.6M sodium methoxide (50 mL) for 1 h, then neutralised with Amberlite IR-120 (H^+) resin, filtered, and concentrated. The residue was crystallised from methanol–ethyl acetate to give **5** (5.67 g, 89%), m.p. 168–170°, $[\alpha]_D^{22}$ +19° (*c* 0.15, methanol); λ_{max}^{MeOH} 275 nm (ε 8624).

Anal. Calc. for $C_{19}H_{30}N_4O_8$: C, 51.58; H, 6.78; N, 12.69. Found: C, 51.11; H, 6.81; N, 12.64.

7-[6-O-(6-Hydroxyhexyl)-3,4-O-isopropylidene-β-D-galactopyranosyl]theophylline (**6**). — A solution of **5** (3.527 g, 7.8 mmol) in anhydrous acetone (135 mL) was stirred with 2,2-dimethoxypropane (6.7 mL) and toluene-*p*-sulfonic acid (260 mg) for 2 h at room temperature, then neutralised with saturated, aqueous $NaHCO_3$, and concentrated. A solution of the residue in dichloromethane was washed with half-saturated aqueous NaCl, dried, and concentrated. Semi-crystal-

line **6** (3.645 g, 95%) was obtained from ethyl acetate–di-isopropyl ether; $[\alpha]_D^{22}$ +61° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 279 nm (ε 11,300).

Anal. Calc. for $C_{22}H_{34}N_4O_8$: C, 54.80; H, 7.05; N, 11.61. Found: C, 54.83; H, 7.17; N, 11.60.

*7-[2-O-Benzoyl-6-O-(6-benzoyloxyhexyl)-3,4-O-isopropylidene-β-*D*-galactopyranosyl]theophylline* (**7**). — Conventional treatment of **6** (3.61 g, 7.49 mmol) with benzoyl chloride (3.15 mL, 27.1 mmol) in pyridine (30 mL) and crystallisation of the product from ethanol gave **7** (4.435 g, 86%), m.p. 150–151°, $[\alpha]_D^{22}$ +97° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 277 nm (ε 13,700). ^{1}H-N.m.r. data ($CDCl_3$): δ 8.2–7.17 (m, 11 H, H-8 and 2 Ph), 6.28 (d, 1 H, *J* 8.5 Hz, H-1′), 5.61 (dd, 1 H, *J* 6.2 Hz, H-2′), 4.58 (dd, 1 H, *J* 1.5 Hz, H-3′), 4.5–3.5 (m, 8 H, H-4′,5′,6′,6′, 2 CH_2O), 3.53 and 3.28 (2 s, 6 H, NMe-1,3), 2.1–1.2 (m, 14 H, CMe_2 and 4 CH_2).

Anal. Calc. for $C_{36}H_{42}N_4O_{10}$: C, 62.61; H, 6.08; N, 8.11. Found: C, 62.82; H, 6.16; N, 8.24.

*7-[2-O-Benzoyl-6-O-(6-benzoyloxyhexyl)-β-*D*-galactopyranosyl]theophylline* (**8**). — A solution of **7** (4.435 g, 6.43 mmol) in dichloromethane (23 mL) and aqueous 90% formic acid (23 mL) was stirred overnight at room temperature, then diluted with dichloromethane, washed with half-saturated aqueous NaCl, saturated $NaHCO_3$, and water, dried, and concentrated. Flash chromatography (solvent *B*) of the residue gave **8** isolated as an amorphous powder (3.96 g, 95%), $[\alpha]_D^{22}$ −39° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 277 nm (ε 8000).

Anal. Calc. for $C_{33}H_{38}N_4O_{10}$: C, 60.92; H, 5.85; N, 8.62. Found: C, 60.80; H, 5.73; N, 8.53.

*7-[2-O-Benzoyl-6-O-(6-benzoyloxyhexyl)-3,4-dideoxy-β-*D*-*erythro*-hex-3-enopyranosyl]theophylline* (**9**). — To a solution of **8** (3.92 g, 6.03 mmol) in toluene (40 mL) and *N,N*-dimethylformamide (10 mL) were added iodoform (4.91 g, 12.48 mmol) and triphenylphosphine (6.5 g, 24.78 mmol). The mixture was kept for 1 h at 100°, then diluted with ethyl acetate, washed with aqueous $NaHCO_3$, $Na_2S_2O_3$, and water, dried, and concentrated. Flash chromatography (solvent *C* followed by ether) of the residue gave **9** (2.56 g, 69%), isolated as a syrup, $[\alpha]_D^{22}$ −108° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 277 nm (ε 9140). ^{1}H-N.m.r. data ($CDCl_3$): δ 8.2–7.2 (m, 11 H, H-8 and 2 Ph), 6.43 (d, 1 H, *J* 8 Hz, H-1′), 6.17–5.85 (m, 3 H, H-2′,3′,4′), 4.85 (m, 1 H, H-5′), 4.35 [m, 2 H, $OCH_2(CH_2)_4CH_2OBz$], 3.7–3.3 [m, 4 H, H-6′,6′ and $OCH_2(CH_2)_4OBz$], 3.56 and 3.36 (2 s, 6 H, NMe-1,3), 2.1–1.1 [m, 8 H, $OCH_2(CH_2)_4CH_2O$].

Anal. Calc. for $C_{33}H_{36}N_4O_8$: C, 64.28; H, 5.84; N, 9.09. Found: C, 64.32; H, 5.85; N, 8.70.

*7-[3,4-Dideoxy-6-O-(6-hydroxyhexyl)-β-*D*-*erythro*-hex-3-enopyranosyl]theophylline* (**10**). — A solution of **9** (2.3 g, 3.73 mmol) in anhydrous methanol (90 mL) was treated with methanolic 2M sodium methoxide (1.5 mL) for 18 h at room temperature. The mixture was neutralised with Amberlite IR-120 (H^+) resin, filtered, and concentrated to dryness. Column chromatography (ethyl acetate–methanol, 9:1) of the residue afforded **10** (1.395 g, 92%), isolated as a syrup, $[\alpha]_D^{22}$ −22° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 279 nm (ε 8220).

Anal. Calc. for $C_{19}H_{28}N_4O_6$: C, 55.88; H, 6.86; N, 13.72. Found: C, 55.90; H, 6.85; N, 13.96.

*7-[6-O-(5-Carboxypentyl)-3,4-dideoxy-β-*D-glycero-*hex-3-enopyranosyl-2-ulose]theophylline* (**11**). — A solution of pyridinium dichromate (1.375 g, 3.66 mmol) and **10** (225 mg, 0.55 mmol) in dry *N,N*-dimethylformamide (3 mL) was stirred for 5 h at room temperature, then poured slowly into stirred ethyl acetate (~70 mL), and filtered through Silica Gel G (Merck). After removal of the solvents *in vacuo*, a solution of the residue in ethyl acetate was filtered through Silica Gel G, and concentrated. P.l.c. (ethyl acetate–acetone, 8:2) of the residue gave amorphous **11** (165 mg, 71%), $[\alpha]_D^{22}$ −93° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 278 nm (ε 7900).

Anal. Calc. for $C_{19}H_{24}N_4O_7$: C, 54.28; H, 5.71; N, 13.33. Found: C, 54.80; H, 6.41; N, 13.42.

Covalent binding of **11** *to immunoglobulins.* — To a solution of **11** (25 mg, 0.06 mmol) in dry 1,4-dioxane (0.2 mL) were added *N*-hydroxysuccinimide (8 mg, 0.06 mmol) and dicyclohexylcarbodi-imide (13 mg, 0.06 mmol). The mixture was stirred for 2 h at room temperature and overnight at 4°, then filtered, and concentrated. P.l.c. (ethyl acetate–acetone 7:3) of the residue gave amorphous **12** (20 mg, 65%), $[\alpha]_D^{22}$ −27° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 277 nm (ε 8600). The active ester **12** was immediately coupled as follows. To a stirred solution of human IgG (8.5 mg, 5.3×10^{-2} nmol) in phosphate-buffered saline (PBS, pH 7.0) was added a solution of **12** (0.27–6.57 mg, 5.3–12.7 × 10^{-1} nmol) in *N,N*-dimethylformamide (1.1 mL). Stirring was continued for 2 h at room temperature and then for 2 h at 4°. The slightly cloudy solution was centrifuged (15,000*g*) for 30 min at 4° and the conjugate was isolated by gel filtration on Sephadex G-50 (elution with PBS). Protein content was determined by the dye-binding method[15] with Coomassie Blue G-250. Protein-bound ketonucleoside was determined as follows. The pH of an aliquot of the solution of purified protein was adjusted to 10, and the solution was kept for 2 h at room temperature and then freeze-dried. The residue was extracted with ethanol, and the theophylline content of the extract was determined at 272 nm.

*7-[3,4-Dideoxy-6-*O*-(6-triphenylmethoxyhexyl)-β-*D-erythro-*hex-3-enopyranosyl]theophylline* (**14**). — To a stirred solution of **10** (600 mg, 1.47 mmol) in dry dichloromethane (3.75 mL) were added chlorotriphenylmethane (500 mg, 1.79 mmol), triethylamine (0.516 mL, 3.7 mmol), and dimethylaminopyridine (15 mg, 0.12 mmol)[16]. The solution was kept overnight at room temperature and then concentrated. Flash chromatography (solvent *B*) of the residue gave amorphous **14** (860 mg, 90%), $[\alpha]_D^{22}$ −18° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 279 nm (ε 8994).

Anal. Calc. for $C_{38}H_{42}N_4O_6$: C, 70.15; H, 6.46; N, 8.61. Found: C, 69.80; H, 6.40; N, 8.25.

*7-[3,4-Dideoxy-6-*O*-(6-*O*-triphenylmethoxyhexyl)-β-*D-glycero-*hex-3-enopyranosyl-2-ulose]theophylline* (**15**). — To a solution of **14** (670 mg, 1.03 mmol) in dry dichloromethane (7 mL) were added pyridinium dichromate (603 mg, 1.6 mmol) and molecular sieve Type 3A (1.15 g). The suspension was stirred for 2 h at room

temperature and then poured slowly into ether. The mixture was filtered through silica gel G (Merck), the silica gel was washed with 1:1 ethyl acetate–ether, and the combined filtrate and washings were concentrated. Flash chromatography (solvent *B*) of the residue gave amorphous **15** (480 mg, 72%), $[\alpha]_D^{22}$ −92° (*c* 0.15, chloroform); $\lambda_{max}^{CHCl_3}$ 279 nm (ε 8450). ^{1}H-N.m.r. data ($CDCl_3$): δ 7.69 (s, 1 H, H-8), 7.57–6.95 (m, 16 H, H-4′ and 3 Ph), 6.56 (d, 1 H, *J* 1.5 Hz, H-1′), 6.31 (dd, 1 H, *J* 10 Hz, and 2 Hz, H-3′), 4.88 (m, 1 H, H-5′), 3.76–3.23 [m, 4 H, H-6′,6′, and $OCH_2(CH_2)_4CH_2OTr$], 3.63 and 3.4 (2 s, 6 H, NMe-1,3), 3.05 [t, 2 H, $OCH_2(CH_2)_4CH_2OTr$], and 1.88–1.03 [m, 8 H, $OCH_2(CH_2)_4CH_2OTr$].

Anal. Calc. for $C_{38}H_{40}N_4O_6 \cdot 5\ H_2O$: C, 69.40; H, 6.24; N, 8.52. Found: C, 69.36; H, 6.36; N, 8.50.

*7-[3,4-Dideoxy-6-*O*-(6-hydroxyhexyl)-β-*D*-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline* (**16**). — To a solution of **15** (360 mg, 0.55 mmol) in ether (1.2 mL) was added aqueous 98% formic acid. The mixture was kept at room temperature for 7 min, then diluted with ethyl acetate, and washed with half-saturated aqueous NaCl, saturated aqueous $NaHCO_3$, and water. The organic layer was dried (Na_2SO_4) and concentrated. Flash chromatography (solvent *A* followed by 8:2 ethyl acetate–acetone) of the residue gave **16** (200 mg, 89%), isolated as a syrup, $[\alpha]_D^{22}$ −59° (*c* 0.18, chloroform); $\lambda_{max}^{CHCl_3}$ 278 nm (ε 11,000). ^{1}H-N.m.r. data ($CDCl_3$): δ 7.77 (s, 1 H, H-8), 7.23 (dd, 1 H, *J* 10 and 1.5 Hz, H-4′), 6.63 (d, 1 H, *J* 1.5 Hz, H-1′), 6.4 (dd, 1 H, *J* 2 Hz, H-3′), 4.95 (m, 1 H, H-5′), 3.78–3.25 (m, 6 H, H-6′,6′, and 2 OCH_2), 3.65 and 3.43 (2 s, 6 H, NMe-1,3), 2.1–1.1 (m, 8 H, 4 CH_2).

Anal. Calc. for $C_{19}H_{26}N_4O_6 \cdot 0.5\ H_2O$: C, 54.93; H, 6.50; N, 13.49. Found: C, 54.85; H, 6.55; N, 13.42.

*7-[3,4-Dideoxy-6-*O*-(6-docosahexaenoyloxyhexyl)-β-*D*-*glycero*-hex-3-enopyranosyl-2-ulose]theophylline* (**17**). — To a stirred solution of **16** (12 mg, 0.029 mmol) in dry dichloromethane (0.25 mL) were added docosahexaenoic acid (9.49 mg, 0.029 mmol), dimethylaminopyridine (0.2 mg), and dicyclohexylcarbodi-imide (8.94 mg, 0.043 mmol) under nitrogen. The mixture was kept for 2 h at room temperature, then filtered, washed with dilute acetic acid and water, dried, and concentrated. P.l.c. (1:1 ethyl acetate–light petroleum) of the residue gave amorphous **17** (8.24 mg, 40%), $[\alpha]_D^{22}$ −40° (*c* 0.1, chloroform); $\lambda_{max}^{CHCl_3}$ 278 nm (ε 6900). ^{1}H-N.m.r. data ($CDCl_3$): δ 7.80 (s, 1 H, H-8), 7.23 (dd, 1 H, *J* 10 and 1.5 Hz, H-4′), 6.67 (d, 1 H, *J* 1.5 Hz, H-1′), 6.57 (dd, 1 H, *J* 2 Hz, H-3′), 5.42 (m, 12 H, olefinic protons of docosahexaenoic acid), 3.67 and 3.43 (2 s , 6 H, NMe-1,3), 2.85 (m, 10 H, =CH–CH_2–CH=), 2.38 (m, 4 H, =CH–CH_2–CH_2).

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