Synthesis of an Aromatic Hydrocarbon Diol Epoxide–Cytosine Adduct, 5′-*O*-(9-Phenylxanthen-9-yl)-*N*⁴-[(±)-1β,2α,3α-triacetoxy-1,2,3,4-tetrahydro-4β-naphthyl]-2′-deoxycytidine,† Suitable for Incorporation into Synthetic Oligodeoxyribonucleotides

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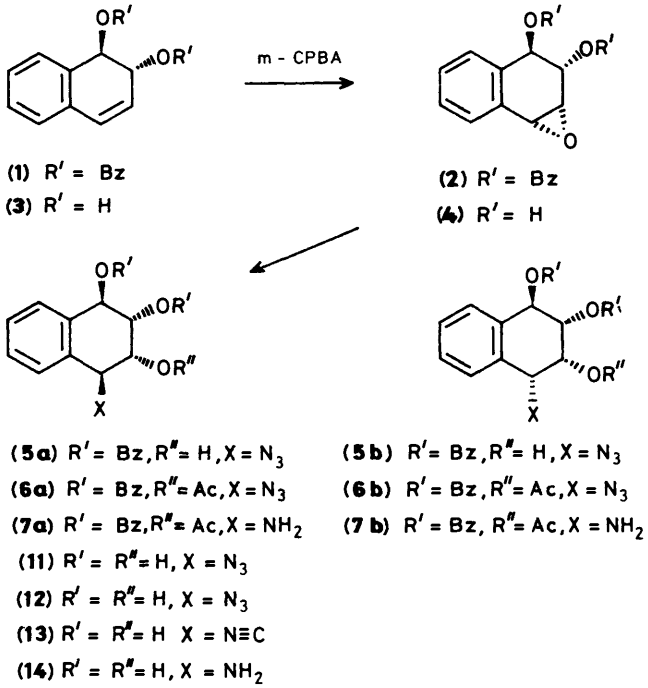
The first total synthesis of a model polyaromatic hydrocarbon diol epoxide–cytosine adduct is described, suitable for incorporation into oligodeoxyribonucleotide synthesis. An approach to control the relative stereochemistry of the diol epoxide–nucleoside adduct is discussed. This synthesis yields 5′-*O*-(9-phenylxanthen-9-yl)-*N*⁴-[(±)-1β,2α,3α-triacetoxy-1,2,3,4-tetrahydro-4β-naphthyl]-2′-deoxycytidine as a mixture of diastereoisomers.†

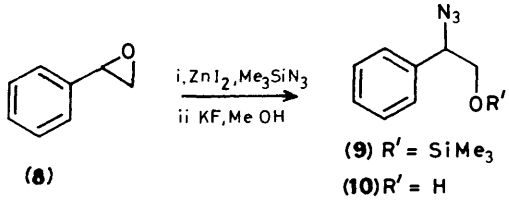
The metabolic activation of many carcinogenic hydrocarbons follows an analogous route to benzo[*a*]pyrene[1] with the formation of diol epoxides which react with DNA.[2] The major adducts formed between benzo[*a*]pyrene and DNA involves a covalent linkage between the amino groups of guanine, adenine, and cytosine and the 10-position of the diol epoxide.[3] We have begun a programme to synthesize aromatic hydrocarbon diol epoxide–deoxyribonucleotide adducts, for incorporation into synthetic oligonucleotides.

The major aims of this synthesis are to produce an aromatic hydrocarbon diol epoxide–deoxynucleoside adduct with control of both the regiochemistry and relative stereochemistry of the adduct. It is essential to see if this type of modified nucleoside can be incorporated into a synthetic oligodeoxyribonucleotide using the phosphotriester chemistry and is stable to the strongly basic deprotection conditions.[4] As the synthesis of the diol epoxides of many polyaromatic hydrocarbons, both racemic and optically active, follows the same basic chemical transformations,[5] the choice of naphthalene as a model seemed reasonable.

The synthetic strategy envisaged disconnection of the naphthalene diol epoxide–deoxycytidine adduct between the C-4 position of the pyrimidine ring and the exocyclic amino group. This type of approach should also allow the synthesis of the guanine and adenine adducts. This study has initially been carried out using *trans*-1,2-dibenzyloxy-1,2-dihydronaphthalene (**1**) as starting material.

Racemic 3α,4α-epoxy-1,2,3,4-tetrahydronaphthalene-1β,2α-diol (**4**) was synthesized from the dihydro diol (**3**).[6,7] This dihydro diol (**3**) prefers the conformation in which the hydroxy groups are pseudo-equatorial[8] and this conformation stereoselectivity directs the epoxidation to give the diol epoxide (**4**) where the 1-hydroxy group and the oxirane ring are *trans* (the *anti*-configuration).[9] Similarly, epoxidation of *trans*-1,2-dibenzoyloxy-1,2-dihydronaphthalene (**1**) gave 1β,2α-dibenzoyloxy-3α,4α-epoxy-1,2,3,4-tetrahydronaphthalene (**2**) as a single diastereoisomer. Treatment of dibenzoyloxy epoxide (**2**) with silver azide for prolonged periods at high temperature in dimethylformamide, gave as the major products the 4-azido 3-alcohols (**5a, b**) (51% (see Scheme 1). The 4-azido 3-alcohols (**5a, b**) were acetylated to give the 4-azido 3-acetates (**6a, b**), and then hydrogenated using Adam's catalyst to the 4-amino 3-acetates (**7a, b**). The lack of stereochemical control in the azide opening





Scheme.

of the epoxide (**2**) with silver azide, and the inability to separate any of the diastereoisomeric mixtures by silica gel chromatography, prompted us to look at other azide nucleophiles. We chose styrene oxide (**8**) as a model in order to ascertain the regiochemical control of various azide nucleophiles. Treatment of styrene oxide (**8**) with trimethylsilyl azide, using zinc iodide as catalyst, gave an intermediate (**9**) which was not isolated but converted directly into 2-azido-2-phenylethanol (**10**) in high yield. Treatment of the dibenzyloxy epoxide (**2**) under the same conditions failed to react. However, the reaction of the diol

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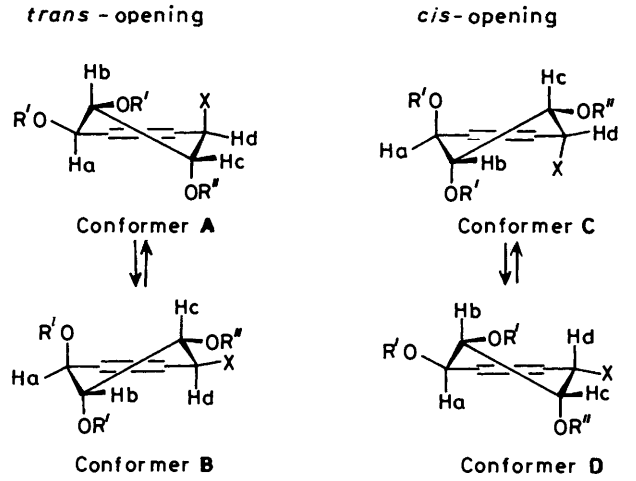
† Throughout, relative stereochemistry is described by α and β, the substituent at the 1-position being regarded as β.

epoxide (**4**) with trimethylsilyl azide and catalytic zinc iodide gave racemic 4β-azido-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**11**) in disappointingly low yield. Acetylation of this azido triol (**11**) gave the azido triacetate (**12**) in high yield. However, this overall route was dismissed in favour of a higher yielding one.

The recent use of trimethylsilyl cyanide, with zinc iodide as catalyst, has been shown to react with α-hydroxy epoxides with total regiochemical and stereochemical specificity and allows simple conversion of the intermediate isonitrile into the amine.[10] Treatment of the diol epoxide (**4**) with trimethylsilyl cyanide and zinc iodide, followed by potassium fluoride in methanol, gave 4β-isocyano-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**13**) in reasonable yield (54%) and as a single diastereoisomer. Finally, the isocyano triol (**13**) was treated with hydrogen chloride gas in methanol, neutralised with base, and the product chromatographed on Sephadex LH20 to give 4β-amino-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**14**). The synthesis of the racemic amino triol (**14**) represents one half of our convergent synthesis (see Scheme 1).

The structural assignments have been justified by the following three factors. Firstly, under Lewis-acid conditions, using catalytic zinc iodide, nucleophiles preferentially attack aryl-substituted epoxides at the benzylic carbon atom.[11] This was substantiated by the regiospecific opening of styrene oxide (**8**) with trimethylsilyl azide at the benzylic position. Secondly, the use of the epoxides (**2**) and (**4**) with known relative stereochemistry predetermines the relative stereochemistry between positions C-1 and C-2 to be *trans*, and *cis* between positions C-2 and C-3 in all cases. Finally, the epoxide (**4**) is expected to undergo *trans*-diaxial ring opening with trimethylsilyl cyanide and zinc iodide to yield the isonitrile (**13**) by comparison with reactions in the literature.[10] The use of ^{1}H n.m.r. coupling constants for determination of the relative stereochemistry at C-4 in this ring system is complicated by ring-inversion of the half-chair conformation. Thus, ^{1}H n.m.r. coupling constants alone cannot be used to ascertain the relative stereochemistry of the substituent at C-4. However, combined with the supporting chemical evidence, the ^{1}H n.m.r. coupling constants can be rationalised. Compounds with the β-configuration at position C-4 (*trans*-opening) can exist as either conformers **A** or **B**, and compounds with the α-configuration at C-4 (*cis*-opening) can exist as either conformers **C** or **D** (see Table). Glusker *et al.* have shown that the observed ^{1}H n.m.r. coupling constants can be presumed to be a weighted average of the ratio of conformers in solution.[12] The ^{1}H n.m.r. coupling constants of the *cis*- and *trans*-opened products of the 1,2,3,4-tetrahydronaphthalene system can be interpreted by comparison with those in a cyclohex-5-ene-1,2,3,4-tetrol[13] and a 7,8,9,10-tetrahydrobenzo[*a*]pyrene.[6,14] In the cyclohex-5-ene system the *trans*-opened product exists in an approximately 1:1 ratio of conformers **A** and **B**. However, in the 7,8,9,10-tetrahydrobenzo[*a*]pyrene system the *trans*-opened product exists almost exclusively as conformer **A**, since the substituent at C-10 resides in a quasi-axial environment in order to avoid adverse steric interaction with the bay region hydrogen at C-11.[6,15] The *cis*-opened cyclohex-5-ene exists almost exclusively as conformer **D** in solution, whereas in the benzo[*a*]pyrene system the substituent at C-10 prefers a pseudo-axial environment (conformer **C**). Analysis of the coupling constants in the 1,2,3,4-tetrahydronaphthalene system indicates that the products of *trans*-opening generally show no conformational preference and therefore resemble the cyclohex-5-ene system. The products of *cis*-opening also exhibit no conformational preference unlike either the cyclohex-5-ene system (conformer **D**) or the 7,8,9,10-tetrahydrobenzo[*a*]pyrene system (conformer **C**). This lack of any conformational preference for either the *cis*- or *trans*-opened products in the 1,2,3,4-tetrahydronaphthalene system invalidates the use of the ^{1}H n.m.r. coupling constants for assigning the relative stereochemistry at C-4.

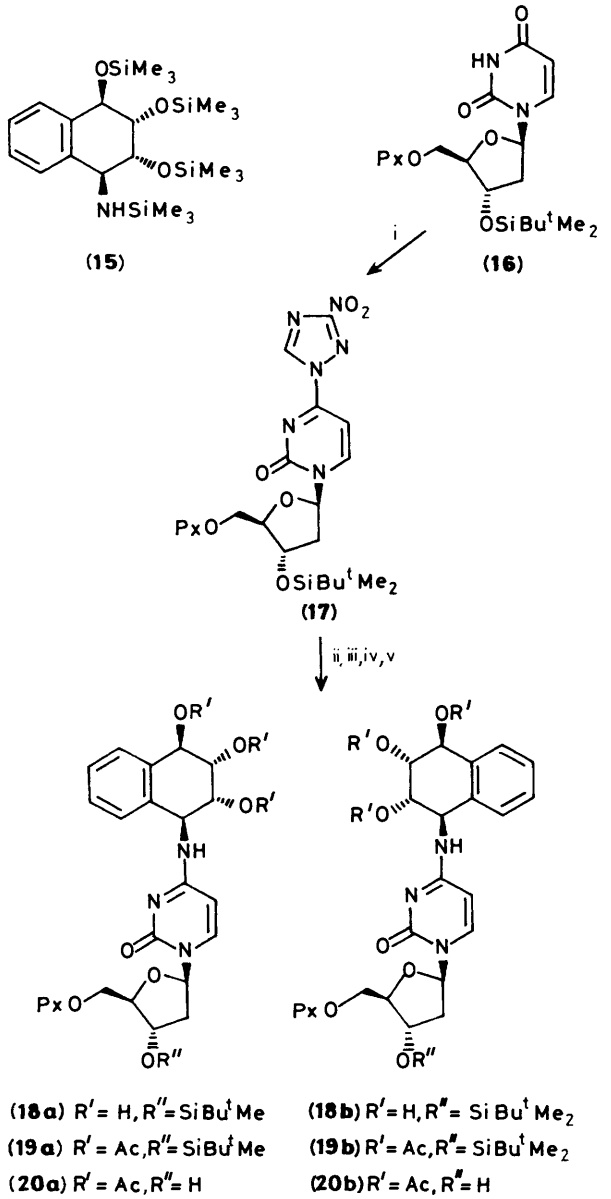
Table. Comparison of ^{1}H n.m.r. coupling constants



Compd.	Ring system	R′	R″	X	*trans*-Opening $J_{a,b}$	$J_{b,c}$	$J_{c,d}$	*cis*-Opening $J_{a,b}$	$J_{b,c}$	$J_{c,d}$
(**5a, b**)	N	Bz	H	N_3	5.5	2.0	7.5	5.5	2.5	7.0
(**6a, b**)	N	Bz	Ac	N_3	6.0	2.0	7.5	5.5	2.0	6.0
(**7a, b**)	N	Bz	Ac	NH_2	5.5	2.0	7.0	6.0	2.0	5.5
(**11**)	N	H	H	N_3	4.5	2.0	8.0			
(**12**)	N	Ac	Ac	N_3	5.1	2.3	7.7			
(**13**)	N	H	H	N≡C	4.0	2.0	7.8			
(**14**)	N	H	H	NH_2	5.2	2.0	7.0			
(**19a, b**)	N	Ac	Ac	dC	6.0	2.2	7.1			
(**20a, b**)	N	Ac	Ac	dC	6.0	2.2	7.1			
Ref. 13	C	H	H	OH	5.4	2.3	5.4	8.1	2.1	3.0
Ref. 14	B	Ac	Ac	OAc	8.9	2.3	3.8	3.5	2.3	4.9

N = 1,2,3,4-tetrahydronaphthalene ring system, B = 7,8,9,10-tetrahydrobenzo[*a*]pyrene ring system, C = cyclohex-5-ene ring system, dC = deoxycytidine nucleoside.

The comparable ease of nucleophilic displacement of the 3-nitro-1,2,4-triazolo group from the 4-position of pyrimidine nucleosides,[16] prompted us to look at this type of activated pyrimidine nucleoside for the synthesis of our modified cytidine base. Conversion of the protected deoxyuridine base (**16**) with diphenyl phosphorochloridate and 3-nitro-1,2,4-triazole in acetonitrile and triethylamine gave exclusively the activated pyrimidine nucleoside (**17**). The conditions used for coupling the aminotriol (**14**) with the activated nucleoside (**17**) have not yet been fully optimised. The method involved transient protection of the hydroxy groups of the amino triol (**14**) with trimethylsilyl chloride in pyridine to yield the amide (**15**), and this was then treated with the activated nucleoside (**17**) (see Scheme 2). The use of the racemic amine (**14**) for this coupling reaction with the enantiomerically pure activated nucleoside (**17**) yields, after work-up, (**18a, b**) as a mixture of diastereoisomers. The products (**18a, b**) were not isolated since they were found to be unstable to purification on silica gel chromatography, but converted directly into the desired triacetates (**19a, b**). The latter were obtained pure only after silica gel chromatography followed by preparative reverse-phase h.p.l.c.; this allowed partial resolution of the two diastereoisomers. The diastereoisomerically pure nucleoside (**19a**) or (**19b**) was used for assigning the ^{1}H n.m.r. spectrum of the major fraction, isolated from the h.p.l.c. purification as a



Scheme 2. *Reagents:* i, diphenylphosphorochloridate, 3-nitro-1,2,4-triazole, triethylamine, acetonitrile; ii, 1,8-diazabicyclo[5.4.0]undec-7-ene, pyridine; iii, sodium hydrogen carbonate; iv, acetic anhydride, pyridine: v, tetrabutylammonium fluoride, tetrahydrofuran; Px = 9-phenylxanthen-9-yl

mixture of diastereoisomers. Finally, removal of the 3′-dimethyl-t-butylsilyl protecting group of (**19a**, **b**), using tetrabutylammonium fluoride in tetrahydrofuran, gave the desired deoxycytidine adducts (**20a**, **b**) in good yield. The products (**20a**, **b**) were isolated as a mixture of diastereoisomers by short-column silica gel chromatography. The modified nucleosides (**20a**, **b**) are now suitably protected for incorporation into a synthetic oligonucleotide and this work will be reported shortly.

The use of both racemic and optically active polyaromatic hydrocarbon dihydro diols can now be considered which possess a wide spectrum of biological activity.

Experimental

N.m.r. spectra were run on a Bruker WM240 (250 MHz), Varian EM360A (60 MHz), or Jeol FT 200 MHz spectrometer. Chemical shifts are reported as p.p.m. downfield from tetramethylsilane. Mass spectra were run with a V.G. 70SEQ instrument at the Medical Research Council, Carshalton, Surrey. High performance liquid chromatography (h.p.l.c.) was carried out on a Gilson 320 with 620 datamaster, Shimadzu SPD6A U.V. spectrophotometric detector, Waters Z-module, and Waters 8MBC18 10μ reverse-phase column. Compounds were precipitated from dichloromethane in light petroleum (b.p. 30—40 °C), and centrifuged on a M.S.E. Mistral 3000 instrument. M.p.s were recorded on an Electrothermal melting point apparatus and are uncorrected. Flash-column silica gel chromatography was carried out using Merck Kieselgel 60 (Art. 9385) and t.l.c. on Merck silica gel $60F_{254}$ aluminium-backed plates. All compounds were obtained from Aldrich Chemical Co., Gillingham, Dorset. The compounds described were visualised as one spot on t.l.c.

Epoxidation of the Olefin (**1**).—Solid *m*-chloroperbenzoic acid (1.27 g, 7.36 mmol, 100) was added to a stirred solution of *trans*-1,2-dibenzyl-1,2-dihydronaphthalene (**1**) (2.593 g, 7.01 mmol) in dichloromethane (250 ml) at room temperature under nitrogen. The reaction mixture was heated under reflux for 12 h, cooled to 20 °C, and *m*-chloroperbenzoic acid (1.27 g, 7.36 mmol) added. The reaction was heated under reflux for a further 2 h and then poured into saturated aqueous sodium hydrogen carbonate (100 ml). The aqueous fraction was extracted with dichloromethane (3 × 100 ml), and the total organic fraction washed with saturated brine (100 ml), dried ($MgSO_4$), filtered, and evaporated under reduced pressure to give a white solid (3.638 g). The product, R_F[dichloromethane–hexane, 1:1 (v:v)] 0.2, was purified by flash column chromatography on silica gel (230 g) using dichloromethane–hexane (1:1) to yield racemic 1β,2α-*dibenzoyl*-3α,4α-*epoxy*-1,2,3,4-*tetrahydronaphthalene* (**2**) (1.652 g, 61%), m.p. 132—133 °C (from diethyl ether–hexane) (Found: C, 74.45; H, 4.8. $C_{24}H_{18}O_5$ requires C, 74.6; H, 4.7%); $\nu_{max.}$(Nujol) 3 075 (epoxide) and 1 722 cm^{-1} (OBz); δ_H(200 MHz, $CDCl_3$) 8.13—8.00 (4 H, m, 7-, 8-, 9-, and 10-H), 7.59—7.17 (10 H, m, 2 OBz), 6.74 (1 H, d, $J_{1,2}$ 9.7 Hz, 1-H), 5.57 (1 H, d, $J_{1,2}$ 9.7 Hz, 2-H), 4.10 (1 H, d, $J_{3,4}$ 4.5 Hz, 4-H), and 4.02 (1 H, d, $J_{3,4}$ 4.5 Hz, 3-H).

Silver Azide Opening of the Epoxide (**2**).—Freshly prepared moist silver azide (12.84 mmol, 5.0 equiv.) was added with care to a stirred solution of 1β,2α-dibenzoyloxy-3α,4α-epoxy-1,2,3,4-tetrahydronaphthalene (**2**) (0.991 g, 2.567 mmol) in dry dimethylformamide (100 ml) at room temperature under nitrogen. The mixture was then heated at 110—115 °C for 1 week. The dimethylformamide was removed under reduced pressure and the residue diluted with water (200 ml). The aqueous fraction was extracted with ethyl acetate (4 × 100 ml), and the total organic fraction washed with brine (100 ml), dried ($MgSO_4$), and evaporated under reduced pressure to give a yellow foam. The product, R_F[dichloromethane–hexane, 9:1 (v:v)] 0.15, was purified by flash column chromatography on silica gel (50 g) and elution in a gradient of dichloromethane–hexane (4:1—95:5) gave 4β- *and* 4α-*azido*-1β,2α-*dibenzoyloxy*-1,2,3,4-*tetrahydronaphthalen*-3α-*ol* (**5a**, **b**) (0.558 g, 51%), m.p. 61—63 °C (Found: C, 66.95; H, 4.55; N, 9.7. $C_{24}H_{19}N_3O_5$ requires C, 67.15; H, 4.45; N, 9.8%); $\nu_{max.}$($CHCl_3$) 3 460 (OH), 2 100 (N_3), and 1 718 cm^{-1} (OBz); δ_H(60 MHz, $CDCl_3$) 8.10—7.78 (4 H, m, 7-, 8-, 9-, and 10-H), 7.54—7.20 (10 H, m, 2 OBz), 6.57 (1 H, d, $J_{1,2}$ 5.5 Hz, 1-H), 6.32 (1 H, d, $J_{1*,2*}$ 5.5 Hz, 1*-H), 5.74 (1 H, dd, $J_{1,2}$ 5.5 Hz, $J_{2,3}$ 2.0 Hz, 2-H), 5.64 (1 H, dd, $J_{1*,2*}$ 5.5 Hz, $J_{2*,3*}$ 2.5 Hz, 2*-H), 5.02 (1 H, d, $J_{3,4}$ 7.5 Hz, 4-H), 4.79 (1 H, d, $J_{3*,4*}$ 7.0 Hz, 4*-H), 4.60 (1 H, br s, 3- and 3*-H), and 3.23 (1 H, s, OH).

Acetylation of the Azido Alcohols (**5a**, **b**).—Acetic anhydride (6.00 ml, 63.47 mmol) was added *via* a syringe to a stirred

solution of 4β- and 4α-azido-1β,2α-dibenzoyloxy-1,2,3,4-tetrahydronaphthalen-3α-ol (**5a**, **b**) (0.459 g, 1.07 mmol) in dry pyridine (1.70 ml) at 0 °C under nitrogen. The reaction was allowed to warm slowly to room temperature, and after 24 h was quenched by addition to saturated aqueous sodium hydrogen carbonate (250 ml). The aqueous fraction was extracted with chloroform (3 × 300 ml) and the combined extracts dried ($MgSO_4$), filtered, and reduced under reduced pressure to give 4β- and *4α-azido-3α-acetoxy-1β,2α-dibenzoyloxy-1,2,3,4-tetrahydronaphthalene* (**6a**, **b**) (0.500 g, 99%), m.p. 60—62 °C; R_F[dichloromethane–hexane, 1:1 (v:v)] 0.20 (Found: C, 66.3: H, 4.3; N, 9.1. $C_{26}H_{21}N_3O_6$ requires C, 66.25; H, 4.5; N, 8.9); $\nu_{max.}(CHCl_3)$ 2 102 (N_3), 1 750 (OAc), and 1 720 cm^{-1} (OBz); δ_H(60 MHz, $CDCl_3$) 8.16—7.78 (4 H, m, 7-, 8-, 9-, and 10-H), 7.58—7.22 (10 H, m, 2 OBz), 6.58 (1 H, d, $J_{1,2}$ 6.0 Hz, 1-H), 6.55 (1 H, d, $J_{1*,2*}$ 5.5 Hz, 1*-H), 6.03—5.66 (2 H, $J_{2,3}$ 2.0 Hz, $J_{2*,3*}$ 2.0 Hz, 2-H, 2*-H, 3-H, and 3*-H), 5.03 (1 H, d, $J_{3*,4*}$ 6.0 Hz, 4*-H), 4.98 (1 H, d, $J_{3,4}$ 7.5 Hz, 4-H), 2.13 (3 H, s, OAc), and 2.02 (3 H, s, OAc).

Hydrogenation of the Azido Esters (**6a**, **b**).—Adam's catalyst (50 mg) was added to a stirred solution of 4β- and 4α-azido-3α-acetoxy-1β,2α-dibenzoyloxy-1,2,3,4-tetrahydronaphthalene (**6a**, **b**) (0.405 g, 0.86 mmol) in dioxane (31 ml) and the mixture hydrogenated in a Brown's hydrogenator. The reaction was monitored by t.l.c. and the mixture worked up after 2.5 days by filtration through Kieselguhr white, washing of the latter with chloroform (100 ml), and evaporation of the filtrate to give an oil (0.363 g). The product, R_F(chloroform) 0.11, was purified by flash column chromatography on silica gel (12.5 g) and eluted with chloroform to give 4β- and *4α-amino-3α-acetoxy-1β,2α-dibenzoyloxy-1,2,3,4-tetrahydronaphthalene* (**7a**, **b**) (0.259 g, 68%), m.p. 61—63 °C (Found: C, 69.95; H, 5.2; N, 3.3. $C_{26}H_{23}NO_6$ requires C, 70.1; H, 5.2; N, 3.1%); $\nu_{max.}(CHCl_3)$ 3 390—3 210 (NH_2), 1 740 (OAc), and 1 722 cm^{-1} (OBz); δ_H(60 MHz, $CDCl_3$) 8.19—7.76 (4 H, m, 7-, 8-, 9-, and 10-H), 7.70—7.24 (10 H, m, 2 OBz), 6.59 (1 H, d, $J_{1,2}$ 5.5 Hz, 1-H), 6.57 (1 H, d, $J_{1*,2*}$ 6.0 Hz, 1*-H), 6.09—5.76 (2 H, m, $J_{1,2} = J_{1*,2*}$ 2.0 Hz, 2-H and 2*-H), 5.74 to 5.26 (2 H, m, 3-H, and 3*-H), 4.37 (1 H, d, $J_{3*,4*}$ 5.5 Hz, 4*-H), 4.32 (1 H, d, $J_{3,4}$ 7.0 Hz, 4-H), 2.10 (3 H, s, OAc), 2.00 (3 H, s, OAc), and 1.74 (2 H, s, NH_2).

Azide Opening of Styrene Oxide (**8**).—Trimethylsilyl azide (1.42 ml, 10.7 mmol) was added *via* a syringe to a stirred solution of styrene oxide (**8**) (0.854 g, 7.11 mmol) in dry dichloromethane (50 ml) under nitrogen at room temperature. After 5 min solid zinc iodide (10 mg) was added and the mixture heated under reflux for 4 days. The solvent was removed by distillation and replaced with ethanol-free chloroform (15 ml) and the mixture heated under reflux for a further 2 days. The solvent was then removed under reduced pressure and methanol (50 ml) and potassium fluoride (2.006 g, 21.32 mmol) were added to the residue, the reaction mixture was then stirred at room temperature for 50 min. After this, the mixture was evaporated under reduced pressure and saturated brine (50 ml) added. The aqueous phase was extracted with ethyl acetate (3 × 50 ml), and the combined extracts dried ($MgSO_4$), filtered, and evaporated under reduced pressure to give a yellow oil (1.241 g). The product, R_F(chloroform) 0.22, was purified by flash column chromatography on silica gel (32 g) using chloroform as eluant to yield *2-azido-2-phenylethanol* (**10**) (0.995 g, 86%) as a colourless oil (Found: C, 58.85; H, 5.55; N, 26.1. $C_8H_9N_3O$ requires C, 58.9; H, 5.55; N, 25.75%); $\nu_{max.}$(neat) 3 350 (OH) and 2 100 cm^{-1} (N_3); δ_H(60 MHz, $CDCl_3$) 7.30 (5 H, s, Ph), 4.60 (1 H, t, $J_{1,2}$ 6.0 Hz, 1-H), 3.70 (2 H, t, $J_{1,2}$ 6.0 Hz, $J_{2\text{-H,OH}}$ 6.0 Hz, 2-H), 2.32 (1 H, t, $J_{2\text{-H,OH}}$ 6.0 Hz, OH).

Azide Opening of the Diol Epoxide (**4**).—Trimethylsilyl azide (1.645 ml, 11.83 mmol) and zinc iodide (15 mg) were added (the former *via* a syringe) to a stirred solution of 3α,4α-epoxy-1,2,3,4-tetrahydronaphthalene-1β,2α-diol (**4**) (0.421 g, 2.365 mmol) in dry, ethanol-free chloroform (40 ml) under nitrogen. The mixture was heated under reflux for 20 h after which the solvent was removed by careful distillation. Methanol (30 ml) and potassium fluoride (2.226 g, 23.65 mmol) were then added to the reaction mixture which was then stirred under nitrogen at room temperature for 1.5 h. The solvent was then removed under reduced pressure and saturated brine (50 ml) added. The aqueous phase was extracted with ethyl acetate (3 × 100 ml) and the combined extracts were dried ($MgSO_4$), filtered, and evaporated under reduced pressure to give a red oil (0.350 g). The product, R_F[chloroform–methanol, 9:1 (v:v)] 0.18, was purified by flash chromatography over silica gel (22 g) using chloroform–methanol (95:5) as eluant to yield 4β-*azido-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol* (**11**) (0.044 g, 8—11%), m.p. 138—140 °C (decomp.) (Found: C, 53.95; H, 5.1; N, 18.6. $C_{10}H_{11}N_3O_3$ requires C, 54.30; H, 5.0; N, 19.0%); $\nu_{max.}$(Nujol) 3 320 and 3 200 (OH) and 2 105 cm^{-1} (N_3); δ_H[250 MHz, $(CD_3)_2SO$] 7.40—7.26 (4 H, m, 7-, 8-, 9-, and 10-H), 4.63 (1 H, d, $J_{3,4}$ 8.0 Hz, 4-H), 4.45 (1 H, d, $J_{1,2}$ 4.5 Hz, 1-H), 4.00 (1 H, dd, $J_{3,4}$ 8.0 Hz, $J_{2,3}$ 2.0 Hz, 3-H), and 3.84 (1 H, dd, $J_{2,3}$ 2.0 Hz, $J_{1,2}$ 4.5 Hz, 2-H).

Acetylation of the Azido Triol (**11**).—Acetic anhydride (6.0 ml) was added *via* a syringe to a stirred solution of 4β-azido-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**11**) (0.254 g, 1.149 mmol) in pyridine (1.70 ml) at 0 °C under nitrogen and the mixture was allowed to warm slowly to room temperature. After 12 h at room temperature, the reaction mixture was quenched by addition of ethyl acetate (75 ml), and washed with aqueous sodium hydrogen carbonate (2 × 75 ml) and saturated brine (75 ml). The organic fraction was dried ($MgSO_4$), filtered, and evaporated under reduced pressure to give an off-white solid (375 mg). The product, R_F(chloroform) 0.26, was purified by silica gel chromatography (20 g) using chloroform as eluant to give 4β-*azido-1β,2α,3α-triacetoxy-1,2,3,4-tetrahydronaphthalene* (**12**) (361 mg, 90%), m.p. 134—135 °C (from ethyl acetate–hexane) (Found: C, 55.65; H, 5.2; N, 12.0. $C_{16}H_{17}N_3O_6$ requires C, 55.35; H, 4.95; N, 12.1%); $\nu_{max.}(CHCl_3)$ 2 105 (N_3) and 1 750 cm^{-1} (OAc); δ_H(250 MHz, $CDCl_3$) 7.52 (4 H, m, 7-, 8-, 9-, and 10-H), 6.12 (1 H, d, $J_{1,2}$ 5.1 Hz, 1-H), 5.55 (1 H, dd, $J_{1,2}$ 5.1, $J_{2,3}$ 2.3 Hz, 2-H), 5.50 (1 H, dd, $J_{3,4}$ 7.7 Hz, $J_{2,3}$ 2.3 Hz, 3-H), 4.81 (1 H, d, $J_{3,4}$ 7.7 Hz, 4-H), 2.13 (3 H, s, OAc), 2.12 (3 H, s, OAc), and 2.07 (3 H, s, OAc); δ_C(63.9 MHz, $CDCl_3$), 170.10, 169.99, and 169.65 ($OCOCH_3$), 132.41 and 131.88 (C-5 and C-6), 129.79, 129.49, 129.24, and 128.43 (C-7, C-8, C-9, and C-10), 71.26 (C-1), 69.40 (C-2 and C-3), 60.09 (C-4), and 21.02, 20.89, and 20.79 ($OCOCH_3$).

Isonitrile Opening of the Diol Epoxide (**4**).—Trimethylsilyl cyanide (1.43 ml, 10.73 mmol) was added to a stirred solution of 3α,4α-epoxy-1,2,3,4-tetrahydronaphthalene-1β,2α-diol (**4**) (0.562 g, 3.157 mmol) in ethanol-free chloroform (43 ml) under nitrogen. The mixture was heated under reflux for 16 h after which the solvent was removed by careful distillation. Methanol (32 ml) and potassium fluoride (2.021 g, 21.47 mmol) were added to the reaction mixture which was then stirred for 4 h at room temperature. The mixture was evaporated under reduced pressure and saturated brine (50 ml) added to the residue. The aqueous phase was extracted with ethyl acetate (4 × 100 ml), and combined extracts were dried ($MgSO_4$), filtered, and evaporated under reduced pressure to give a brown oil (0.740 g). The product, R_F[chloroform–methanol, 9:1 (v:v)] 0.21, was purified by flash column chromatography on silica gel using chloroform–methanol (95:5) as eluant to yield 4β-isocyano-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**13**) (0.350 g, 54%) as an amorphous, hygroscopic solid (1 spot on t.l.c.); δ_H(60

MHz, CD_3OD, D_2O) 7.61—7.27 (4 H, m, ArH), 5.07 (1 H, d, $J_{3,4}$ 7.8 Hz, 4-H), 4.73 (1 H, d, $J_{1,2}$ 4.0 Hz, 1-H), 4.27 (1 H, dd, $J_{3,4}$ 7.8, $J_{2,3}$ 2.0 Hz, 3-H), and 4.13 (1 H, dd, $J_{2,3}$ 2.0 Hz, $J_{1,2}$ 4.0 Hz, 2-H); δ_C[62.9 MHz, $(CD_3)_2SO$] 159.19 (N≡C), 136.57 and 130.97 (C-5 and C-6), 130.41, 128.23, 127.85, and 126.57 (C-7, C-8, C-9, and C-10), and 72.50, 70.91, 69.02, and 56.64 (C-1, C-2, C-3, and C-4).

Reduction of the Isonitrile (**13**).—Hydrogen chloride gas was bubbled through a stirred solution of 4β-isocyano-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**13**) (0.441 g, 2.15 mmol) in methanol (170 ml), at 20 °C for 2 h; the reaction vessel was then stoppered and stirred for a further 2 h at room temperature. The mixture was worked up by first removing the solvent under reduced pressure and then adding aqueous sodium hydroxide (1.0M; 5.3 ml) to the residue. The aqueous phase was extracted with ethyl acetate (5 × 50 ml) and the combined extracts were dried (Na_2SO_4), filtered, and evaporated under reduced pressure to give an oily solid (0.489 g). The product was purified by Sephadex LH20 chromatography using water as eluant to yield 4β-*amino*-1,2,3,4-*tetrahydronaphthalene*-1β,2α,3α-*triol* (**14**) (0.149 g, 36%) as a lyophilysed white solid, m.p. 77—78 °C (decomp.) (Found: C, 61.35; H, 6.85; N, 6.75. $C_{10}H_{13}NO_3$ requires C, 61.55; H, 6.7; N, 7.2%); $\nu_{max.}$(Nujol) 3 600—3 100 (OH and NH_2) and 1 652 cm^{-1} (NH_2); δ_H[250 MHz; $(CD_3)_2SO$–D_2O] 7.40—7.20 (4 H, m, ArH), 4.44 (1 H, d, $J_{3,4}$ 7.0 Hz, 4-H), 4.23 (1 H, d, $J_{1,2}$ 5.2 Hz, 1-H), 4.07 (1 H, dd, $J_{2,3}$ 2.0 Hz, $J_{1,2}$ 5.2 Hz, 2-H), 3.90 (1 H, dd, $J_{3,4}$ 7.0 Hz, $J_{2,3}$ 2.0 Hz, 3-H).

Formation of the Activated Nucleoside (**17**).—Diphenyl phosphorochloridate (0.33 ml, 1.59 mmol) was added to a stirred solution of 3-nitro-1,2,4-triazole (0.181 g, 1.59 mmol) in acetonitrile (6.0 ml) and triethylamine (0.59 ml, 4.24 mmol), at 0 °C under argon. A white precipitate formed immediately and the ice-bath was removed. After the mixture had been stirred for 5 min at room temperature a solution of 5′-*O*-(9-phenylxanthen-9-yl)-3′-*O*-dimethyl-t-butylsilyl-(+)-2′-deoxyuridine (**16**) (0.634 g, 1.06 mmol) in acetonitrile (6.5 ml) was added and the reaction monitored by t.l.c. The reaction was worked up after 20 h by adding dichloromethane (30 ml) and washing the organic fraction with aqueous sodium hydrogen carbonate (2 × 30 ml). The organic fraction was dried (Na_2SO_4), filtered, and evaporated under reduced pressure. The product was purified (from baseline material on t.l.c.) by passing it rapidly through a short column of silica gel (10 g) using chloroform as eluant. Removal of the solvent under reduced pressure gave 4-(3-nitro-1,2,4-triazolo)-1-[β-D-5′-*O*-(9-phenylxanthen-9-yl)-3′-*O*-(dimethyl-t-butylsilyl)-2′-deoxyribofuranosyl]pyrimidin-2(1*H*)-one (**17**) (0.510 g, 69%), R_F(chloroform) 0.20, as an oil that was not subjected to further purification.

Coupling of the Amino Triol (**14**) *with the Activated Nucleoside* (**17**).—Trimethylsilyl chloride (2.24 ml, 177 mmol) was added to a stirred solution of 4β-amino-1,2,3,4-tetrahydronaphthalene-1β,2α,3α-triol (**14**) (0.23 g, 1.18 mmol) in pyridine (10.0 ml) and dimethylformamide (3.0 ml), at 0 °C under argon. The reaction mixture was warmed to room temperature, stirred for 0.5 h and the solvent removed under reduced pressure. More pyridine (5.0 ml) was added and the solvent again removed under reduced pressure. A solution of the activated pyrimidine nucleoside (**17**) (0.51 g, 0.733 mmol) in pyridine (6.0 ml) was added to the resulting oil at room temperature under argon with stirring. Afrer 1 h, 1,8-diazabicyclo[5.4.0]undec-7-ene (1.0 ml, 6.45 mmol) was added and the reaction mixture heated under reflux for 5 h and at 80 °C for 12 h. It was then allowed to cool to room temperature when the solvent was removed under reduced pressure. The resulting oil was added to water (200 ml) and extracted with chloroform (4 × 50 ml). The organic fraction was dried (Na_2SO_4), filtered, and evaporated under reduced pressure. Pyridine (1.28 ml) and acetic anhydride (2.40 ml) were added to the crude reaction mixture under argon which, after it had been stirred for 1 h at room temperature, was diluted with dichloromethane (50 ml) and washed with aqueous sodium hydrogen carbonate (5 × 50 ml). The organic fraction was dried (Na_2SO_4), filtered, and evaporated under reduced pressure to give an oil (0.60 g). This was chromatographed twice on silica gel (25 g and 12.5 g), first using chloroform as eluant, and then using a gradient of chloroform–dichloromethane (3:1) to chloroform (100%) to yield a light brown oil (0.200 g). This was then purified by C-18 reverse-phase h.p.l.c. using a manually controlled gradient running from pure dichloroethane to dichloroethane–acetonitrile (85:15) and precipitated to give the products 5′-O-(9-*phenylxanthen*-9-*yl*)-3′-O-(*dimethyl-t-butylsilyl*)-N^4-[(±)-1β,2α,3α-*tri*-O-*acetoxy*-4β-1,2,3,4-*tetrahydronaphthyl*]-2′-*deoxycytidine* (**19a**, **b**) (85 mg) and a sample of diastereoisomerically pure material (9 mg), overall yield (94 mg, 14.2%); R_F(chloroform) 0.31 (Found: C, 66.2: H, 6.25; N, 4.5. $C_{50}H_{55}N_3O_{11}Si$ requires C, 66.55; H, 6.15; N, 4.65%); δ_H[250 MHz; $(CD_3)_2SO$, pure diastereoisomer] 8.370 (1 H, d, $J_{NH,4}$ 8.5 Hz, NH), 7.755 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 6-H), 7.458 to 7.065 (17 H, br m, xanthenyl and naphthyl 7-, 8-, 9-, and 10-H), 6.144 (1 H, t, $J_{1,2'} = J_{1,3'}$ 6.2 Hz, sugar 1-H), 6.044 (1 H, d, $J_{1,2}$ 6.0 Hz, naphthyl 1-H), 5.728 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 5-H), 5.580 (1 H, t, $J_{3,4}$ 7.1 Hz, $J_{NH,4}$ 8.5 Hz, naphthyl 4-H), 5.472 (1 H, dd, $J_{2,3}$ 2.2 Hz, $J_{1,2}$ 6.0 Hz, naphthyl 2-H), 5.415 (1 H, dd, $J_{2,3}$ 2.2 Hz, $J_{3,4}$ 7.1 Hz, naphthyl 3-H), 4.365 (1 H, br m, sugar 3-H), 3.832 (1 H, br dd, $J_{3,4}$ 7.5 Hz, $J_{4,5}$ 3.5 Hz, sugar 4-H), 3.158 (1 H, dd, $J_{4,5}$ 3.5 Hz, $J_{5,5'}$ 11.0 Hz, sugar 5-H), 3.022 (1 H, dd, $J_{4,5}$ 3.5 Hz, $J_{5,5'}$ 11.0 Hz, sugar 5*-H), 2.234 to 2.095 (2 H, br m, sugar 2-H), 2.130 (3 H, s, OAc), 2.032 (3 H, s, OAc), 2.020 (3 H, s, OAc), 0.080 (9 H, s, Bu^tSiO), 0.008 (3 H, s, MeSiO), and 0.000 (3 H, s, MeSiO).

Removal of the Silyl Protecting Group from the Modified Nucleoside (**19a**, **b**).—Tetrabutylammonium fluoride (1.0M in tetrahydrofuran; 0.220 ml) was added to a stirred solution of the modified nucleoside (**19a**, **b**) (66 mg, 0.0732 mmol) in tetrahydrofuran (2.20 ml) under nitrogen at room temperature. The reaction was quenched after 15 min by the addition of ethyl acetate (15 ml) and saturated brine (10 ml). The aqueous fraction was extracted with ethyl acetate (2 × 5 ml) and the combined extracts were dried (Na_2SO_4), filtered, and evaporated under reduced pressure to yield a yellow oil (95 mg). Purification of this by flash column chromatography on silica gel (3.6 g) using a gradient of chloroform to chloroform–ethanol (98.5:1.5) as eluant gave, after precipitation, 5′-O-(9-*phenylxanthen*-9-*yl*)-N^4-[(±)-1β,2α,3α-*tri*-O-*acetoxy*-1,2,3,4-*tetrahydro*-4β-*naphthyl*]-2′-*deoxycytidine* (**20a**, **b**) (42 mg, 72%), R_F(chloroform) 0.05 (Found: C, 66.8; H, 5.15; N, 5.25. $C_{44}H_{41}N_3O_{11}$ requires C, 67.1; H, 5.25; N, 5.35); δ_H[250 MHz; $(CD_3)_2SO$] for mixture of diastereoisomers, 8.355 (1 H, d, $J_{NH,4\text{-}H}$ 8.5 Hz, NH), 8.345 (1 H, d, $J_{NH,4\text{-}H}$ 8.5 Hz, NH), 7.745 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 6-H), 7.715 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 6-H), 7.448—7.087 (34 H, m, xanthenyl and naphthyl 7-, 8-, 9-, and 10-H), 6.160 (1 H, t, $J_{1,2} = J_{1,2'}$ 6.2 Hz, sugar 1-H), 6.152 (1 H, t, $J_{1,2} = J_{1,2'}$ 6.2 Hz, sugar 1-H), 6.040 (2 H, d, $J_{1,2}$ 6.0 Hz, naphthyl 1-H), 5.700 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 5-H), 5.682 (1 H, d, $J_{5,6}$ 7.5 Hz, pyrimidine 5-H), 5.587 (2 H, br t, $J_{3,4}$ 7.1 Hz, $J_{NH,4\text{-}H}$ 8.5 Hz, naphthyl 4-H), 5.470 (1 H, dd, $J_{1,2}$ 6.0 Hz, $J_{2,3}$ 2.2 Hz, naphthyl 2-H), 5.458 (1 H, d, $J_{1,2}$ 6.0 Hz, $J_{2,3}$ 2.2 Hz, naphthyl 2-H), 5.415 (2 H, dd, $J_{2,3}$ 2.2 Hz, $J_{3,4}$ 7.1 Hz, naphthyl 3-H), 5.282 (2 H, d, $J_{OH,3\text{-}H}$ 4.5 Hz, OH), 4.233 (2 H, br m, sugar 3-H), 3.880 (2 H, br dd, $J_{3,4}$ 7.5 Hz, $J_{4,5}$ 4.0 Hz, sugar 4-H), 3.162 (2 H, br d, sugar 5-H), 3.044 (1 H, dd, $J_{4,5}$ 4.0 Hz, $J_{5,5'}$ 10.5 Hz, sugar 5′-H), 3.042 (1 H, dd, $J_{4,5}$ 4.0 Hz, $J_{5,5'}$ 10.5 Hz,

sugar 5′-H), 2.310 to 2.190 (2 H, br m, sugar 2-H), 2.130 (3 H, s, OAc), 2.120—2.040 (2 H, br m, sugar 2′-H), 2.125 (3 H, s, OAc), 2.036 (3 H, s, OAc), 2.028 (3 H, s, OAc), 2.025 (3 H, s, OAc), and 1.998 (3 H, s, OAc); *m/z* (xenon, fast atom bombardment) 788 (M^+, 2.19%), 532 (M^+ − 256, 2.29), and 257 (M^+ − 531, 100).

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