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THE CHEMISTRY OF 2',3'-SECONUCLEOSIDES IV. SYNTHESIS AND REACTIONS OF 3'-AZIDO-2',3'-DIDEOXY-2',3'- SECOTHYMIDINE AND RELATED ANALOGUES**

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Abstract - The synthesis of a series of 2',3'-seconucleoside analogues related to nucleosides previously shown to posssess anti-HIV activity is described. Using strategies to distinguish between the three primary hydroxyl groups of a 2',3'-seconucleoside, 3'-azido-2',3'-dideoxy-2'3'-secothymidine and 2',3'-dideoxy-2',3'-secothymidine have been prepared. These compounds, several of the intermediates in their synthesis, as well as $O^2$2'-anhydro-3'-azido-3'-deoxy-2',3'-secothymidine and several 2',3'-secouridine analogues have been tested for their ability to prevent infection of cells by HIV. None showed any significant toxicity or antiviral activity.

In the past few years, we have published a number of papers on seconucleosides[1-4] - a term coined by McCormick and McElhinney[5] to describe some acyclic nucleosides. We have specifically been interested in the 2',3'-seconucleosides, which can be considered to be normal nucleosides which lack the C2'-C3' bond. Using a variety of strategies and blocking groups, we have shown that it is possible chemically to differentiate in reactivity between the three primary hydroxyl groups formed if a ribonucleoside is treated successively with sodium periodate and then reduced with borohydride. This is possible in the pyrimidine[1,2] or purine[3] field and we have also shown that some selectivity is retained by phosphodiesterases towards the diasterioisomeric p-nitrophenyl esters of the 5'- and 3'-phosphates of 2',3'-secouridine.[4]

** We dedicate this paper to Professor Yu Wang on the occasion of his 80th birthday.

Other relevant work in the 2',3'-seconucleosides field has been reported by Shugar and co-workers.[6-10] They have confirmed the expected flexibility of the seconucleosides[7] but have shown that they do prefer a conformation where the acyclic chain is in an extended form.[8] They have also shown that, unlike acyclovir, 2',3'-secoguanosine is not a substrate for the thymidine kinase of herpes simplex virus type-1[9] and that 2',3'-secocytidine is only partially phosphorylated by wheat shoot nucleoside phosphotransferase.[10] However, it has been known for some time that when $tRNA^{Phe}$ is treated with periodate, and the terminal dialdehyde residue reduced with borohydride to give a 2',3'-secoadenosine 3'-terminus, this can still be correctly and efficiently aminoacylated,[11] showing that it is possible for a seconucleoside to adopt a biologically recognizable conformation. Recently the synthesis of some relevant 3',4'- and 1',2'-seconucleosides has been reported.[12]

We here wish to report the synthesis of some 2',3'-secopyrimidinenucleosides which are related to the known anti-HIV agents 3'-azido-2',3'-dideoxythymidine and 2',3'-dideoxythymidine.[14] The replication cycle of HIV includes a number of steps which present targets for attack by antiviral agents. Most of the effective nucleoside analogues reported so far are thought to work by chain-terminating the synthesis of viral DNA transcribed from RNA by the viral RNA-dependent DNA polymerase or reverse transcriptase. However, to be an effective substrate for this enzyme, the nucleoside 5'-triphosphate is required and in the absence of an efficient method of transporting phosphate derivatives directly into cells, the nucleoside has to be a substrate for a 5'-kinase. Unlike the herpes viruses, HIV does not code for its own kinase and thus this initial phosphorylation depends upon the recognition of the analogue by a host cell kinase (usually a pyrimidine or even a thymidine-kinase) which normally has a very stringent specificity. Thus, this approach requires that any analogue should not differ very markedly from the normal substrate, otherwise phosphorylation will not occur, but this also inevitably means that the chances are that the triphosphate eventually produced will be a substrate not only for the reverse transcriptase but also for the cellular DNA polymerase and will thus be toxic.

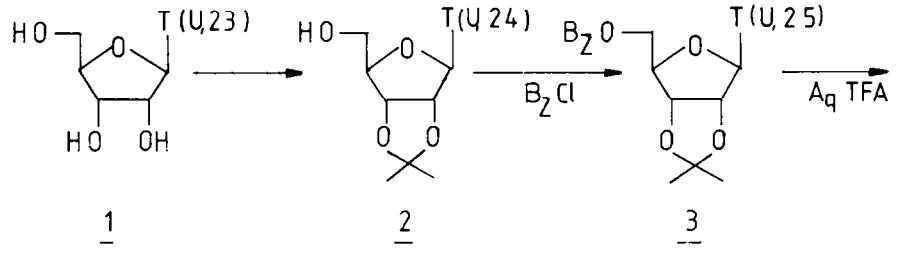
As we had aquired the ability to alter at will the functional groups of 2',3'-seconucleosides, we thought it might be of interest to prepare some 2',3'-seco-analogues of known anti-HIV agents in the hope that they might still be kinase substrates but the triphosphates of which might have some increased selectivity in their recognition by the transcriptase and the polymerase.

The following series of compounds were made starting from ribothymidine and from uridine. We describe the chemistry in detail for the former series here as they are all novel compounds; the analogous reactions were also performed using uridine and some of the individual steps have been reported previously.[1,2]

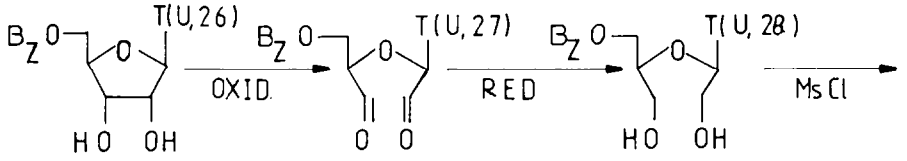
Because of the acidic conditions required using HI during a subsequent reaction, the 5'-hydroxyl group of ribothymidine had to be benzoylated rather than tritylated. This was achieved *via* the 2',3'-O-isopropylidene derivative in a predictable series of reactions. (Compounds 1-4, Scheme 1). Subsequent oxidation (5), reduction (6) and mesylation gave 5'-O-benzoyl-2',3'-di-O-mesyl-2',3'-secoribothymidine (7) using standard conditions described before.[1,2] Treatment of compound (7) with DBU gave the O^2,2'-anhydro analogue (8) which, on treatment with HI to open the anhydro ring (9), followed by catalytic reduction using 10% Pd/C and hydrogen (10), displacement of the mesyl group with LiN_3 (11) and removal of the 5'-O-benzoyl group with ammonia, gave the desired 3'-azido-2',3'-dideoxy-2',3'-secothymidine (12) in reasonable yield

The preparation of 2',3'-dideoxy-2',3'-secothymidine (17) was achieved by periodate oxidation followed by borohydride reduction of the readily available 5'-O-tritylribothymidine. Reactions to give compounds

SCHEME 1



1 2 3



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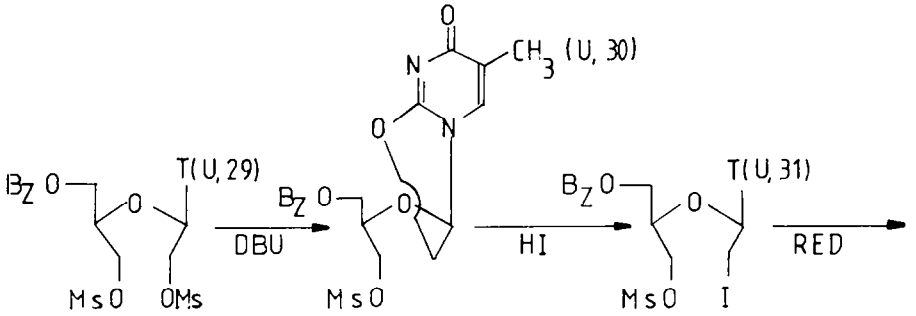
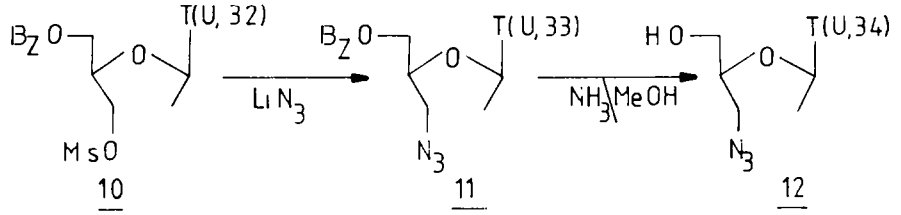


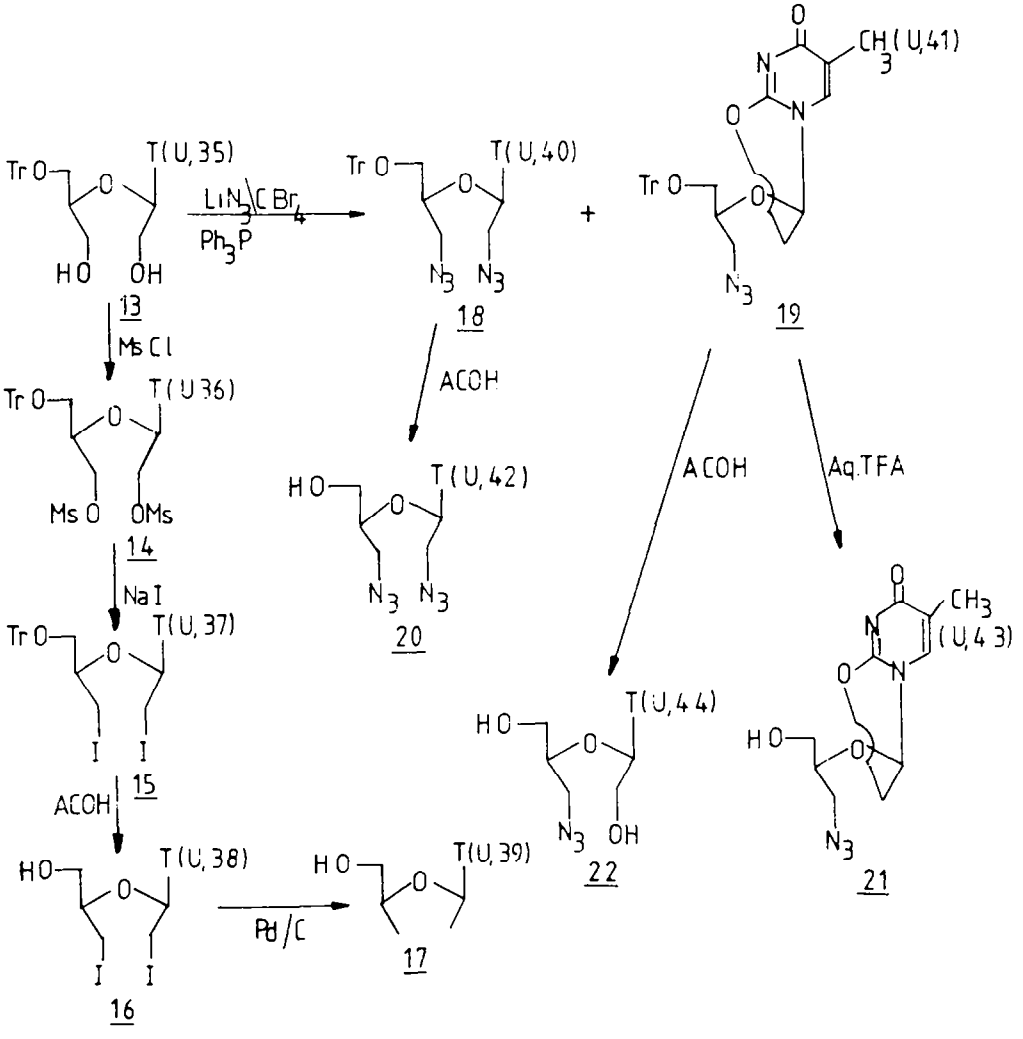
7 8 9



10 11 12

(14–17) followed an entirely predictable course to give the desired compound (17) in good yield. The diol (13) was also used as the starting material for the preparation of a more rigid analogue, O^2,2'-anhydro-3'-azido-2',3'-dideoxy-2',3'-secothymidine (21). Treatment of compound (13) with lithium azide under the conditions described by Hata[13], gave two compounds (18, 19) in the ratio 3:5 which could be separated and characterized. Compound (18) could be detritylated to give (20) and compound (19) could be detritylated under conditions which opened the anhydro ring to give (22) or which left it intact to give compound (21).

SCHEME 2



Similar reactions were carried out starting from uridine and where novel, the experimental details are reported.

All the compounds described here were tested for their ability to inhibit the infection of cells by HIV but no significant efficacy or toxicity was seen. This is probably because the compounds are not substrates for the host-cell thymidine kinase and attempts are currently being made to synthesise derivatives which circumvent this block. Compounds 12, 16, 17, 20, 21, 22, 34, 38, 39, 42, 43 and 44 were tested against HIV and several other virus strains but no activity or toxicity of any kind was detected.

EXPERIMENTAL

N.m.r. spectra were recorded on one of the following instruments: Varian XL100, Jeol FX90Q or Jeol GX270 with $(CD_3)_2$ SO as the solvent unless otherwise stated. U.v. spectra were measured in ethanol on a Perkin Elmer 552 spectrophotometer. Mass spectra were recorded on a Kratos MS 80 RF instrument using the fast atom bombardment technique, sodium chloride was added as required. Column chromatography was carried out on silica gel either Kieselgel 60 type 7734, 0.063–0.200 mm, 70–230 mesh ASTM or Kieselgel 60 type 9385, 0.04–0.063 mm, 230–400 mesh ASTM (E. Merck A.G. Darmstadt, W. Germany). All experiments were carried out under scrupulously dry conditions unless otherwise stated and all evaporations of solvents were carried out under reduced pressure.

Synthesis of compounds 35–38, 40, 42, 44 and 13–14 has been previously reported in ref. 1 and 2 respectively.

2',3'-O-Isopropylideneribothymidine (2). Ribothymidine (10 g, 33.5 mmol) was suspended in dry acetone (250 ml) and then anhydrous copper (II) sulphate (20 g) and concentrated sulphuric acid (0.25 ml) was added The mixture was then stirred in a sealed bottle for 48 hours and filtered. The filtrate was then neutralised with dry calcium hydroxide powder (10 g) for one hour and filtered. The product which crystallises while excess solvent is removed can be further recrystallised from methanol to give 10.9 g (94%) of fine needles. U.v. λ_{max}, 266 nm, ϵ = 9,820 (pH 6 in water) n.m.r. δ 11.4 (1H,s,NH), 7.6 (1H,s,H-6), 5.9 (1H,d,H-1'), 4.9 (2H,m,H-2',3'), 4.1 (1H,q,H-4'), 3.6 (2H,m,H-5'), 1.8 (3H,s,CH_3), 1.55 (3H,s,$(CH_3)_2C$), 1.36 (3H,s,$(CH_3)_2C$) Mass spec. m/z 299 $(M+H)^+$, 321 (M+Na).

5'-O-Benzoylribothymidine (4). To a solution of 2',3'-O-isopropylideneribothymidine (16.8 g, 41 mmol) in pyridine (200 ml) was added dropwise benzyoyl chloride (9.24 g, 65 7 mmol) in pyridine (80 ml). After 4 hours at room temperature the solvent was removed and the residue coevaporated with toluene. The resulting syrup was dissolved in dichloromethane and the solution was extracted three times with water. After drying the organic phase was evaporated to dryness under reduced pressure. The residue was dissolved in trifluoroacetic acid (33.6 ml and water (7 ml) and the solution was left for 20 mins at room temperature. Removal of the solvent gave a residue that was purified on a chromatographic column eluting with ethanol: chloroform (1.3) to give (17.2 g, 83%) a white solid. U v. λ_{max}, 260 nm, ϵ = 10,350 (ethanol) n.m.r. δ 11.4 (1H,s,NH), 8 1 (1H,s,H-6), 8–7.4 (5H,m,benzoyl protons), 5.9 (1H,d,H-1'), 4.5 (3H,m,CH_2-5',H-4'), 4.1 (4H,m,OH-3',OH-2',H-3',H-2'). Mass spec. m/z 364 $(M+H)^+$.

5'-O-Benzoylribothymidine dialdehyde (5). To a solution of 5'-O-benzoylribothymidine (9 0 g, 24.8 mmol) in ethanol (300 ml) was added sodium periodate (5.3 g) in water (130 ml) and the solution was left for 24 hours at room temperature in the dark. The solvent was then removed to yield a white solid that was extracted with acetone. Removal of the acetone under reduced pressure gave the product (8.2 g, 92%). U.v. λ_{max}, 260 nm, ϵ = 10,000 pH 6 0 in water n.m.r. ^{1}H, δ (d6-DMSO) no assignable resonances, spectrum not resolved. This is typical of a nucleoside dialdehyde.[15]

5'-O-Benzoyl-2',3'-secoribothymidine (6). To a stirred solution of 5'-O-benzoylribothymidine dialdehyde (9.0 g, 24.9 mmol) in ethanol-water (1 1) (600 ml) was added sodium borohydride in small portions (3.7 g) until complete reaction had occurred. The pH was then adjusted to pH 7 and the solvent removed under reduced pressure. The resulting white solid was extracted with acetone, yielding a white solid on evaporation. Final purification was carried out on column chromatography (toluene: acetone (3:7) (7.4 g, 82%). U.v. λ_{max}, 260 nm, ϵ = 10,200; n.m.r. δ 11.4 (1H,bs,NH), 8.0–7.5 (6H,m,H-6,benzoyl protons), 5.9 (1H,t,H-1'), 4.4 (2H,m,CH_2-5'), 3.7 (5H,m,CH_2-2',CH_2-3',H-4'), 1.8 (3H,s,CH_3). Mass spec. m/z 365

$(M+H)^+$.

5'-O-Benzoyl-2',3'-di-O-mesyl-2',3'-secoribothymidine (7). To a stirred solution of 5'-O-benzoyl-2',3'-secoribothymidine (5.0 g, 13.7 mmol) in pyridine (65 ml) at 0°C was added dropwise mesyl chloride (6.2 g, 54.26 mmol) in pyridine (6.5 ml). After 2 hours at 0°C the solvent was removed under reduced pressure and the residue coevaporated with toluene. The residue was then triturated with water and then dried by coevaporation with acetone. The resulting solid was further purified by column chromatography eluting with toluene: acetone (7:3) to give a white solid (5.9 g, 82%). U.V. λ_{max}, 260 nm, ϵ = 9,980; n.m.r. δ 11.4 (1H,s,NH), 8-7.4 (6H,m,H-6,benzoyl protons), 5.9 (1H,t,H-1'), 4.6-4.1 (3H,m,H-4',H-3', H-2'), 3.2 (6H,d,2 x $CH_3SO_2^-$), 2.3 (2H,m,CH_2-5'), 1.6 (3H,s,CH_3).

0^2,2'-Anhydro-5'-O-benzoyl-3'-O-mesyl-2',3'-secothymidine (8). To a solution of 5'-O-benzoyl-2',3'-di-O-mesyl-2',3'-secoribothymidine (3.5 g, 6.7 mmol) in dry dichloromethane (33 ml) was added DBU (1.0 g, 6.57 mmol). After 2 hours at room temperature the solvent was removed and the residue coevaporated with acetone to yield a yellow solid. The solid was purified by column chromatography, eluting with acetone, yielding a white solid (2.5 g, 89%). (Found: C, 49.5; H, 4.5; N, 6.1; $C_{18}H_{21}N_2O_8S$ requires: C, 49.9; H, 4.65; N, 6.4). U.v. λ_{max} 250 nm ϵ, 7,800. n.m.r. δ 7.9-7.4 (6H,m,H-6',benzoyl protons), 6.1 (1H,q,H-1') 4.8-4.3 (6H,m,CH_2-5', H-4', H-3', CH_2-2'), 3.2 (3H,s,$CH_3SO_2^-$), 1.6 (3H,s,CH_3). Mass spec. M/z 425 $(M+H)^+$.

5'-Benzoyl-2'-deoxy-2'-iodo-3'-O-mesyl-2',3'-secothymidine (9). A solution of 0^2,2'-anhydro-5'-O-benzoyl-3'-O-mesyl-2',3'-secothymidine (3.5 g, 8.2 mmol) in DMF (150 ml) and hydroiodic acid (32 ml) was left for 16 hours at room temperature. The solution was then neutralised by addition of sodium bicarbonate. After removal of the solvent, the residue was partitioned between dichloromethane and water. The organic phase was separated, washed twice with water and then evaporated to dryness. The resulting solid was purified on a chromatographic column eluting with toluene: acetone (1:1) to give a white foam (2.5 g, 55%). U.v. λ_{max} 260 nm, ϵ, 10,100, n.m.r. δ 11.4 (1H,bs,NH), 8-7.4 (6H,m,H-6,benzoyl protons) 6.04 (1H,t,H-1'), 4.5 (2H,H-3'), 4.4 (3H,m,CH_2-2',H-4'), 3.62 (2H,d,CH_2-5'), 3.3 (3H,s,$CH_3SO_2^-$), 1.7 (3H,s,CH_3). Mass spec. m/z 553 $(M+H)^+$.

5'-O-Benzoyl-2'-deoxy-3'-O-mesyl-2',3'-secothymidine (10). 5'-O-Benzoyl-2'-deoxy-2'-iodo-3'-O-mesyl-2',3'-secothymidine (2.49, 4.5 mmol), sodium acetate trihydrate (3.4 g) and palladium charcoal (10%) (0.78 g) were combined in 80% dioxan (166 ml). The suspension was stirred under an atmosphere of hydrogen overnight. The charcoal was then filtered off and the filtrate evaporated to dryness. The resulting solid was partitioned between dichloromethane and water. The dichloromethane layer was separated washed with sodium bisulphite solution and then twice with water. The organic phase was then dried and evaporated to dryness to yield a yellow glass (1.7 g, 90%). (Found: C, 50.9; H, 5.5; N, 6.7; $C_{18}H_{22}N_2O_8S$ requires: C, 50.7; H, 5.2; N, 6.6). U.v. λ_{max} 260 nm, (ϵ, 10,500). n.m.r. δ 11.4 (1H,s,NH), 7.9-7.5 (6H,m,H-6, benzoyl protons) 6.0 (1H,q,H-1'), 4.6-4.0 (5H,m,CH_2-3', H-4',CH_2-5'), 3.25 (3H,s,$CH_3SO_2^-$), 1.8 (3H,s,CH_3), 1.45 (3H,d,CH_3-2'). Mass spec. m/z 427 $(M+H)^+$.

3'-Azido-5'-O-benzoyl-2',3'-dideoxy-2',3'-secothymidine (11). To a solution of 5'-O-benzoyl-2'-deoxy-3'-O-mesyl-2',3'-secothymidine (1.0 g, 2.3 mmol) in dry DMF at 100°C was added lithium azide (0.3 g, 6.1 mmol) with vigorous stirring. After 2 hours the solvent was removed and the resulting syrup triturated with water to give an off white solid. Final purification was achieved using a silica column and eluting with toluene: acetone (7:3) to give a colourless oil (0.62 g, 72%). (Found: C, 54.7; H, 4.9; N, 18.6; $C_{17}H_{19}N_5O_5$ requires C, 54.70; H, 5.10; N, 18.75). U.v. λ_{max} 260 nm. ϵ, 10,200), n.m.r. δ 11.4 (1H,s,NH), 7.9-7.5 (6H,m,H-6,benzoyl protons, 6.1 (1H,q,H-1'), 3.7-3.1 (5H,m,CH_2-3',CH_2-5',H-4'), 1 8 (3H,s,CH_3), 1.4 (3H,d,CH_3-2'). Mass spec. m/z 374 $(M+H)^+$.

3'-Azido-2',3'-dideoxy-2',3'-secothymidine (12). 3'-Azido-5'-O-benzoyl-2',3'-dideoxy-2',3'-secothymidine (0.6 g, 1.6 mmol) was dissolved in methanolic ammonia (saturated at 0°C) (50 ml) and left at room temperature for 2 days. The solvent was then removed and the residue twice coevaporated with methanol. The residue was then purified on a chromatographic column eluting with toluene: acetone (3:7) yielding a white solid (0.34 g, 79%). (Found: C, 42.1; H, 5.1; N, 27.25; $C_{10}H_{15}N_5O_4$ requires C, 42.35; H, 5.13; N, 27.44;) U.v. λ_{max} 260 nm. (ϵ, 10,300) n.m.r. δ 11.4 (1H,s,NH), 7.7 (1H,s,H-6), 6.1 (1H,q,H-1'), 4.8 (1H,bs,OH-5'), 3.7-3.1 (5H,m,CH_2-5', CH_2-3', H-4'), 1.8 (3H,s,CH_3), 1.4 (3H,d,CH_3-2'). Mass spec.

m/z 270 $(M+H)^+$.

2',3'-Dideoxy-2',3'-di-iodo-5'-O-trityl-2',3'-secothymidine (15). To a stirred solution of 2',3'-di-O-mesyl-5'-O-trityl-2',3'-secoribothymidine (1 g, 1.52 mmol) in dry DMF (20 ml) at 100°C was added sodium iodide (1.1 g, 7.6 mmol). The solution was stirred at 100°C for 2 hours and the solvent was removed by evaporation to give a yellow oil which was triturated with water. The resulting solid was filtered off, washed with water and dried. Purification by column chromatography using toluene: acetone (1:1) as the eluant gave a white solid (0.88 g, 80%). U.v. λ_{max} 263 (ϵ, 8,500); n.m.r. δ 11.4 (1H,s,NH), 7.6 (1H,s,H-6), 7.3 (15H,s,Ph_3C), 6.0 (1H,t,H-1'), 4.0 (2H,d,H-2'), 3.8 (3H,m,H-3' and H-4'), 3.1 (2H,d,CH_2-5'), 1.7 (3H,s,CH_3).

2',3'-Dideoxy-2',3'-diiodo-2',3'-secothymidine (16). A suspension of 2',3'-dideoxy-2',3'-di-iodo-5'-O-trityl-2',3'-secothymidine (2.1 g, 2.9 mmol) in 80% aqueous acetic acid (30 ml) was heated at 100°C for 30 min. The solvent was removed by evaporation to give a syrup which was coevaporated with toluene to give a white solid. This was purified by column chromatography first using toluene: acetone (3:2) as eluant and then toluene: acetone (3:7) to give a colourless glass (0.95 g, 70%). U.v. λ_{max} 263 nm. (ϵ, 8,400) n.m.r. δ 11.4 (1H,s,NH), 7.6 (1H,s,H-6), 6.0 (1H,t,H-1'), 4.0 (1H,m,OH-5'), 3.9 (2H,d,CH_2-2'), 3.75 (3H,m,H-3',H-4'), 3.5 (2H,m,CH_2-5'), 1.8 (3H,s,CH_3).

2',3'-Dideoxy-2',3'-secothymidine (17). Palladium on charcoal (10%, 1.64 g) was added to a solution of 2',3'-dideoxy-2',3'-diiodo-2',3'-secothymidine (2.3 g, 4.9 mmol) and sodium acetate trihydrate (7.23 g) in 80% ethanol (200 ml). The suspension was stirred overnight at room temperature under an atmosphere of hydrogen. One equivalent of silver nitrate was then added to precipitate the iodide and the suspension stirred for a further 5 mins. After filtration the solution was evaporated to dryness and the resulting solid purified on a chromatographic column eluting with toluene: acetone (1:1) to give a white solid (0.79 g, 75%). (Found: C, 52.7; H, 7.2; N, 12.5; $C_{10}H_{16}N_2O_4$ requires C, 52.6; H, 7.1; N, 12.3). U.v. λ_{max} 260 nm.
(ϵ, 10,000); n.m.r. δ 11.3 (1H,bs,NH), 7.8 (1H,s,H-6), 5.9 (1H,q,H-1'), 4.6 (1H,t,OH-5'), 3.3 (2H,m,CH_2-5'), 1.7 (3H,s,CH_3), 1.38 (3H,d,CH_3-2'), 1.1 (3H,d,CH_3-3'). Mass spec. 229 $(M+H)^+$.

O,^{2}2'-Anhydro-3'-azido-3'-deoxy-5'-O-trityl-2',3'-secothymidine (19). To a stirred solution of 5'-O-trityl-2',3'-secoribothymidine (5.5 g, 10.9 mmol), carbon tetrabromide (10.2 g) and lithium azide (5.5 g) in dry DMF (30 ml) was added triphenylphosphine (7.82 g) in dry DMF during 1½ hour at 20°C. The reaction was left overnight at room temperature. Ethanol was then added and the solution was left for 10 mins. The solvent was removed and the resulting syrup was poured onto water (400 ml), giving a solid which was separated from the supernatant and dried by coevaporation with acetone. The two compounds were separated on a chromatographic column by eluting with toluene: acetone (2:8). The fraction containing the fastest running compound gave a white solid, 2',3'-diazido-2',3'-dideoxy-5'-O-trityl-2',3'-secothymidine (18) (1.9 g, 32%). (Found: C, 63.2; H, 5.2; N, 20.4. $C_{29}H_{28}N_8O_4$ requires C, 63.0; H, 5.1; N, 20.3); U.v. λ_{max} 262 nm. (ϵ, 11,000); n.m.r. δ 11.4 (1H,s,NH), 7.8 (1H,s,H-6), 7.3 (15H,m,Ph_3C), 6.0 (1H,t,H-1'), 3.8-3.5 (5H,m,CH_2-2',CH_2-3',H-4'), 3.1 (2H,m,CH_2-5'), 1.8 (3H,s,CH_3); the slowest running compound was identified as O,^{2}2'-Anhydro-3'-azido-3'-deoxy-5'-O-trityl-2',3'-secothymidine (19) (2.9 g, 53%). (Found: C, 68.3; H, 5.4; N, 13.4. $C_{29}H_{27}N_5O_4$ requires C, 68.36; H, 5.34; N, 13.74); U.v. λ_{max} 250 nm. (ϵ, 8,500) in ethanol; n.m.r. δ 7.82 (1H,s,H-6), 7.4-7.2 (15H,m,Ph_3C), 6.1 (1H,d,H-1'), 4.8 (2H,d-d,CH_2-2'), 4.5 (1H,t,H-4'), 3.67-3.45 (2H,m,CH_2-3'), 3.0 (2H,d,CH_2-5'), 1.66 (3H,s,CH_3). Mass spec. m/z 510 $(M+H)^+$.

3'-Azido-3'-deoxy-2',3'-secoribothymidine (22). O^2,2'-Anhydro-3'-azido-3'-deoxy-5'-O-trityl-2',3'-secothymidine (1.5 g, 2.9 mmol) in 80% acetic acid (50 ml) was heated at 100°C for 2 hours, when tlc indicated that complete detritylation had taken place. The solvent was removed and the residue coevaporated with toluene. The crude product was purified using column chromatography, eluting with ethylacetate: ethanol (8:2) to give a white solid (0.38 g, 45%). (Found: C, 42.3; H, 5.5; N, 24.7; $C_{10}H_{15}N_5O_5$ requires C, 42.1; H, 5.30; N, 24.6); U.v. λ_{max} 260 nm. (ϵ, 10,000); n.m.r. δ 11.3 (1H,s,NH), 7.8 (1H,s,H-6), 5.9 (1H,t,H-1'), 3.8-3.3 (7H,m,CH_2-5',CH_2-3',CH_2-2',H-4'), 1.7 (3H,s,CH_3). Mass spec. m/z 286 $(M+H)^+$.

2',3'-Diazido-2',3'-dideoxy-2',3'-secothymidine (20). 2',3'-Diazido-2',3'-dideoxy-5'-O-trityl-2',3'-secothymidine (2.5 g, 4.5 mmol) was deprotected using the conditions described for the preparation of compound (22) to give a white solid (0.7 g, 51%). (Found: C, 38.9; H, 4.6; N, 36.3; $C_{10}H_{14}N_8O_4$ requires C, 38.70; H, 4.6; N, 36.10); U.v. λ_{max} 260 nm. (ϵ, 9,800); n.m.r. δ 11.4 (1H,bs,NH), 7.8 (1H,s,H-6), 5.9 (1H,t,H-1'), 4.8 (1H,bs,OH-5'), 3.8-3.5 (7H,m,CH_2-5',CH_2-3',CH_2-3',H-4'), 1.7 (3H,s,CH_3). Mass spec. m/z 311 $(M+H)^+$.

O^2,2'-Anhydro-3'-azido-2',3'-dideoxy-2',3'-secothymidine (21). O^2,2'-Anhydro-3'-azido-3'-deoxy-5'-O-trityl-2',3'-secothymidine (0.25 g, 0.49 mmol) in n-butanol: trifluoroacetic acid (9:1) (3 ml) was stirred for 2 minutes at room temperature, when tlc indicated that complete detritylation had taken place. Excess n-butanol was then added and the solvent removed under vacuo at 30°C to give a yellow residue. The residue was then purified on a silica column eluting with chloroform: ethanol (7:3) to give a white solid (0.088 g, 67%). (Found: C, 42.1; H, 5.0; N, 24.5; $C_{10}H_{13}N_5O_4$ $1H_2O$ requires C, 42.1; H, 5.3; N, 24.7); U.v. λ_{max} 250 nm. (ϵ, 8,000) in water pH 6.0; n.m.r. δ 7.83 (1H,s,H-6), 6.1 (1H,d-d,H-1'), 5.1 (1H,t,OH-5'), 4.8 (2H,d-d,CH_2-2'), 4.5 (1H,d,H-4'), 3.56-3.3 (4H,m,CH_2-5'CH_2-3'), 1.8 (3H,s,CH_3); Mass spec. 268 $(M+H)^+$.

5'-O-Benzoyl-2',3'-O-isopropylidene uridine (25). (2.18 g, 79%). (Found: C, 59.0; H, 5.1; N, 6.9. $C_{18}H_{20}N_2O_7$ requires C, 58.8; H, 5.2; N, 7.2). U.v. λ_{max} 261 nm. (ϵ, 10,300); n.m.r. δ 11.4 (1H,bs,NH), 8.0-7.4 (6H,m,H-6,benzoyl protons), 5.84 (1H,d,H-1'), 5.54 (1H,d,H-5), 5.12 (1H,d-d,H-5'), 4.92 - 4.4 (3H,m,H-2',H-3',H-4'), 1.54 (3H,s,$(CH_3)_2C$-), 1.36 (3H,s,$(CH_3)_2C$-).

5'-O-Benzoyluridine (26). (1.67 g, 87%). U.v. λ_{max} 261 nm. ϵ, 10,200 (ethanol): n.m.r. δ 11.4 (1H,bs,NH), 8.1-7.5 (6H,m,H-6, benzoyl protons), 5.9 (1H,d,H-1'), 5.6 (1H,d,H-5), 4.5 (2H,m,CH_2-5'), 4.15 (3H,m,H-2',H-3',H-4').

5'-O-Benzoyluridine dialdehyde (27). (4.86 g, 88%). (Found: C, 54.1; H, 4.4; N, 7.8. $C_{16}H_{16}N_2O_8$ + 0.5 H_2O requires C, 54.1; H, 4.2; N, 7.9). U.v. λ_{max} 261 nm. (ϵ, 10,400); n.m.r. δ (d_6-DMSO): no assignable resonances, spectrum not resolved.

5'-O-Benzoyl-2',3'-secouridine (28). (12.86 g, 79%). (Found: C, 54.4; H, 5.0; N, 7.8; $C_{16}H_{18}N_2O_7$ requires C, 54.9; H, 5.1; N, 8.0). U.v. λ_{max} 261 nm. (ϵ, 10,000); n.m.r. δ 11.4 (1H,bs,NH), 8.0-7.4 (6H,m,H-6, benzoyl protons), 5.85 (1H,t,H-1'), 5.35 (1H,d,H-5), 4.3 (2H,m,CH_2-5'), 3.7 (5H,m,CH_2-2',CH_2-3',H-4').

5'-O-Benzoyl-2',3'-di-O-mesyl-2',3'-secouridine (29). (26.5 g, 80%). U.v. λ_{max} 261 nm. (ϵ, 10,400); n.m.r. δ 11.2 (1H,bs,NH), 8.1 to 7.4 (6H,m,H-6, benzoyl protons), 6.1 (1H,t,H-1'), 5.45 (1H,d,H-5), 4.6-4.3 (7H,m,CH_2-2',CH_2-3',H-4',CH_2-5'), 3.25 (3H,s,2'-$CH_3SO_2^-$), 3.2 (3H,s,3'-$CH_3SO_2^-$).

O^2,2'-Anhydro-5'-O-benzoyl-3'-O-mesyl-2',3'-secouridine (30). (17.94 g, 84%). (Found: C, 49.5; H, 4.6; N, 6.7; $C_{17}H_{18}N_2O_7S$ + H_2O requires C, 49.5; H, 4.9; N, 6.8). U.v. λ_{max} 250 nm. (ϵ, 7,500). n.m.r. δ 8.1-7.5 (6H,m,H-6, benzoyl protons), 6.1 (1H,d-d,H-1'), 5.8 (1H,d,H-5), 5.0-4.2 (7H,m,CH_2-2',CH_2-3',H-4',CH_2-5'), 3.3 (3H,s,$CH_3SO_2^-$). Mass spec. m/z 411 $(M+H)^+$.

5'-O-Benzoyl-2'-deoxy-2'-iodo-,3'-O-mesyl-2',3'-secouridine (31). (5.53 g, 61%). U.v. λ_{max} 261 nm. (ϵ, 10,300); n.m.r. δ 11.42 (1H,bs,NH), 8.0-7.4 (6H,m,H-6, benzoyl protons), 6.04 (1H,t,H-1'), 5.48 (1H,d-d,H5), 4.52 (2H,m,CH_2-3'), 4.3 (3H,m,CH_2-2',H-4'), 3.62 (2H,d,CH_2-5'), 3.3 (3H,s,$CH_3SO_2^-$).

5'-O-Benzoyl-2'-deoxy-,3'-O-mesyl-2',3'-secouridine (32). (3.9, 94%). U.v. λ_{max} 260 nm. (ϵ, 10,400). (Found: C, 49.4; H, 4.7; N, 6.7; $C_{17}H_{20}N_2O_8S$ requires C, 49.5; H, 4.9; N, 6.8. n.m.r. δ 11.1 (1H,s,NH), 8.0-7.4 (6H,m,H-6, benzoyl protons), 6.0 (1H,q,H-1'), 5.4 (1H,d,H-5), 4.6-4.0 (5H,m,CH_2-3',H-4',CH_2-5'), 3.25 (3H,s,$CH_3SO_2^-$), 1.45 (3H,d,CH_3-2').

3'-Azido-5'-O-benzoyl-2',3'-dideoxy-2',3'-secouridine (33). (6.9 g, 78%). U.v. λ_{max} 261 nm (ϵ, 9,800); (Found: C, 53.7; H, 4.90; N, 19.6; $C_{16}H_{17}N_5O_5$ requires C, 53.48; H, 4.77; N, 19.49). n.m.r. δ 11.3 (1H,s,NH), 7.8–7.48 (6H,m,H-6,benzoyl protons), 6.1 (1H,q,H-1'), 5.4 (1H,d,H-5), 4.3–3.4 (5H,m,CH_2-5',CH_2-3',H-4'), 1.47 (3H,d,CH_3-2'); Mass spec. m/z 360 $(M+H)^+$.

3'-Azido-2',3'-dideoxy-2',3'-secouridine (34). (3.6 g, 82%). U.v. λ_{max} 261 nm. (ϵ, 10,200) pH 6.0 in water; (Found: C, 42.1; H, 5.6; N, 27.25; $C_9H_{13}N_5O_4$ requires C, 42.35; H, 5.13; N, 27.44); n.m.r. δ 11.3 (1H,s,NH), 7.7 (1H,d,H-6), 5.98 (1H,q,H-1'), 5.65 (1H,d,H-5), 4.8 (1H,t,HO-5'), 3.6–3.29 (5H,m,CH_2-5',CH_2-3',H-4'), 1.47 (3H,d,CH_3-2'); Mass spec. m/z 256 $(M+H)^+$.

2',3'-Dideoxy-secouridine (39). (0.6 g, 81%). (Found: C, 50.5; H, 6.7; N, 13.4; $C_9H_{14}N_2O_4$ requires C, 50.5; H, 6.5; N, 13.1). U.v. λ_{max} 260 nm. (ϵ, 9,400). n.m.r. δ 11.1 (1H,s,NH), 7.6 (1H,d,H-6), 5.8 (1H,q,H-1'), 5.6 (1H,d,H-5), 4.5 (1H,t,OH-5'), 3.3 (2H,m,CH_2-5'), 1.35 (3H,d,CH_3-2'), 1.05 (3H,d,CH_3-3'); Mass spec. m/z 229 $(M+H)^+$.

0,^{2}2'-Anhydro-3'-azido-3'-deoxy-2',3'-secouridine (43). (2.1 g, 75%). (Found: C, 42.9; H, 4.5; N, 27.8; $C_9H_{11}N_5O_4$ requires C, 42.7; H, 4.4; N, 27.7). U.v. λ_{max} 250 nm. (ϵ, 8,200) in water pH 6.0; n.m.r. δ 7.8 (1H,d,H-6), 6.1 (1H,d-d,H-1'), 5.8 (1H,d,H-5), 4.86 (2H,m,CH_2-2'), 4.34 (1H,m,H-4'), 3.67–3.47 (2H,m,CH_2-3'), 3.05 (2H,d,CH_2-5'); Mass spec. m/z 254 $(M+H)^+$.

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REFERENCES

1. Jones, A S.; Walker, R.T.; Wyatt, P.G.; Balzarini, J. and De Clercq, E. *J. Chem. Res.*1985, 336–337.

2. Jones, A.S.; McLean, M.J.; Tanaka, H.; Walker, R.T.; Balzarini, J. and De Clercq, E. *Tetrahedron*, 1985, 41, 5965–5972,

3. Beaton, G.; Jones, A.S. and Walker, R.T. *Tetrahedron*, 1988, 44, 6419–6428.

4. Jones, A.S.; Niwas, S.; Tanaka, H. and Walker, R.T. *Nucleic Acids Research*, 1986, 14 (3), 5409–5416

5. McCormick, J.E. and McElhinney, R.S. *J. Chem. Res.* 1981, 12, 256–260.

6. Birnbaum, G.I.; Stolarski, R.; Kazimierczuk, Z. and Shugar, D. *Can. J. Chem.* 1985, 63, 1215–1221.

7. Oivanen, M.; Lönnberg, H.; Kazimierczuk, Z. and Shugar, D. *Nucleosides and Nucleotides*, 1989 8 (1), 133–144.

8. Stolarski, R.; Kazimierczuk, Z.; Lassota, P. and Shugar, D. *Z. Naturforsch*, 1986, 41C, 758–770.

9. Stolarski, R.; Lassota, P.; Kazimierczuk, Z. and Shugar, D. *Z. Naturforsch*, 1988, 43C, 231–242.

10. Lassota, P.; Kazimierczuk, Z.; Zan-Kowalezewska, M. and Shugar, D. *Biochemical and Biophysical Research Communications*, 1986, 137(1), 453–460.

11. Von Der Haar, F.; Schlimme, E.; Gomez-Guillen, M. and Cramer, F. *Eur. J. Biochem.* 1971, 24, 296–301.

12. Scheiner, P.; Geer, A.; Imbach, J-L. and Schinazi, R.F. *J. Med. Chem.* 1989, 32, 73–76.

13. Abushanab, E. and Sarma, M.S.P. *J. Med. Chem.* 1989, 32, 76–79.

14. De Clercq, E. *J. of Antimicrobial Chemotherapy*, 1989, 23, 35–46.

15. Howarth, O.; Jones, A.S.; Walker, R.T. and Wyatt, P.G. *J. Chem. Soc. Perk. Trans. II*, 1984, 261–265.