

THE TETRAISOPROPYLDISILOXANE-1,3-DIYL: A VERSATILE PROTECTING GROUP FOR  
THE SYNTHESIS OF ADENYLYL-(2 $\rightarrow$ 5')-ADENYLYL-(2 $\rightarrow$ 5')-ADENOSINE (2-5A CORE).

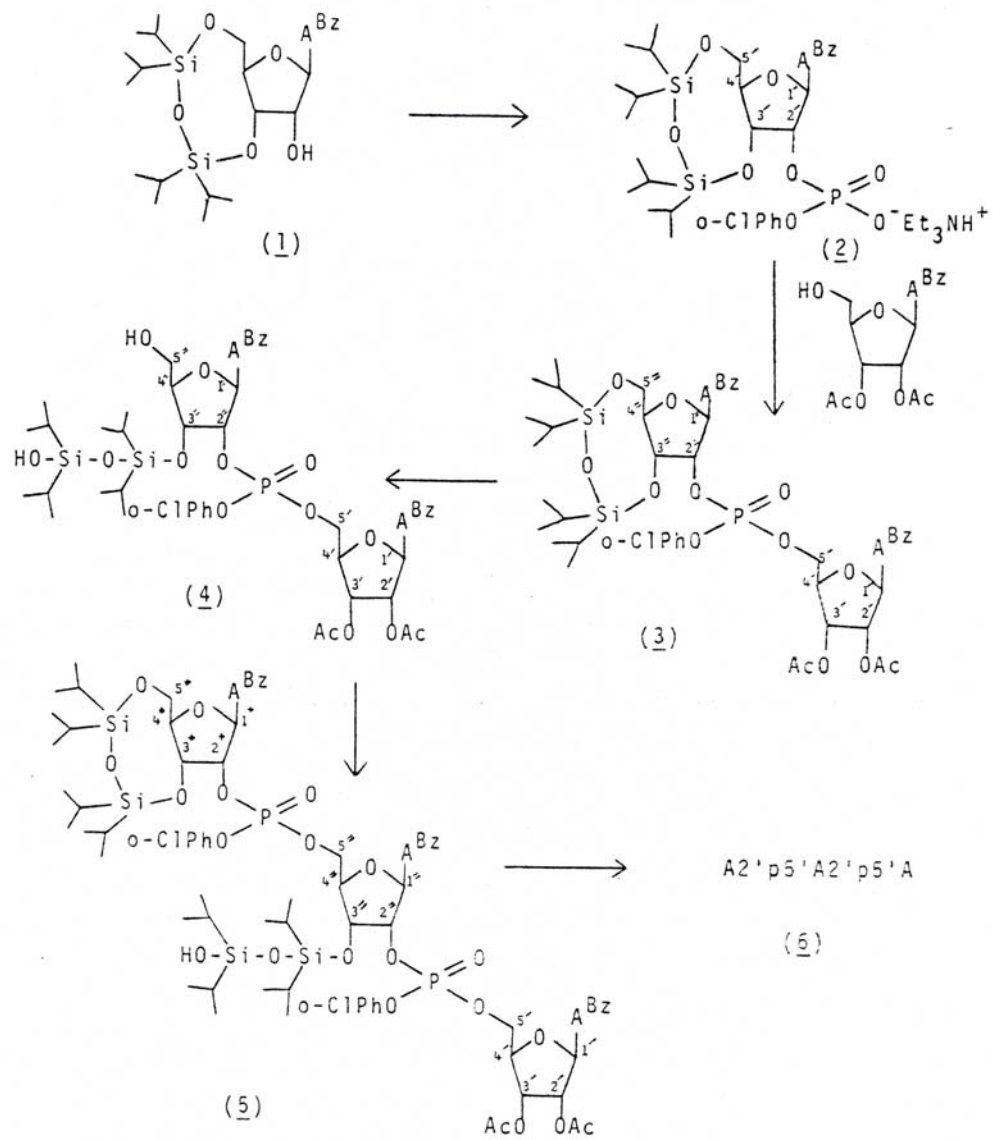
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Summary: A2'p5'A2'p5'A has been synthesised in large scale using 3', 5'-  
tetraisopropylsilyl-6-N-benzoyl adenosine as the starting material.

The antiviral and the antitumour activity of interferon has been directly correlated to the production of pppA2'p5'A2'p5'A in the interferon treated cells<sup>1</sup>. The core, A2'p5'A2'p5'A, has also been found to be remarkably active in certain biological systems<sup>2</sup>. These observations have triggered several chemical synthesis<sup>3</sup> of the core through different routes. We have recently reported<sup>4</sup> such a chemical synthesis using 9-phenylxanthen-9-yl and 2,2-dibromomethylbenzoyl groups for 5'- and 3'-hydroxy protection respectively. Most of the other syntheses<sup>3</sup> so far reported in the literature, like our own synthesis, make use of two separate protecting groups at 5'- and 3'-hydroxyls which are then removable under a different set of conditions.

We now report a chemical synthesis of the core(6) using a bifunctional protecting group, tetraisopropylsilyloxane-1,3-diyl(TIPDSi)<sup>5</sup>, for simultaneous 3'- and 5'-hydroxy protection of 6-N-benzoyl adenosine. The 3',5'-O-TIPDSi-6-N-benzoyl adenosine<sup>16</sup> can be obtained in 10g quantity in one step from 6-N-benzoyl adenosine. The 3',5'-O-TIPDSi protecting group has been reported to migrate<sup>6</sup>, in presence of an acid when DMF is used as solvent, to 2',3'-O-TIPDSi derivative. However, it is stable<sup>5</sup> in most of the reaction conditions which are normally employed for the introduction of a phosphodiester or a triester function. Thus we introduced a phosphodiester function at 2'-position of (1) by reacting (1) with o-chlorophenylphosphorobis-(1,2,4-triazolide) under a standard condition reported in the literature<sup>7</sup>. That the triethylammonium phosphodiester function was specifically introduced at 2'-position in (2), was unequivocally ascertained by the characteristic downfield shift of H-2' absorption ( $\delta$ 5.4) in the <sup>1</sup>H-NMR spectrum of (2)<sup>8</sup> when compared with <sup>1</sup>H-NMR spectrum of (1)<sup>5</sup> ( $\delta$ 4.6 dd, J<sub>1,2</sub>=0.6Hz, J<sub>2,3</sub>=3Hz; H-2'). The pure phosphodiester salt (2)<sup>8</sup>, R<sub>f</sub><sup>14</sup>=0, (1.26 mmol) was then condensed with 6-N-benzoyl-2',3'-di-O-acetyl adenosine (0.84 mmol) in dry pyridine solution (15 ml) in presence of an excess of 1-mesitylenesulphonyl-3-nitro-1,2,4-triazole (MS-NT)<sup>7,14</sup> to obtain a pure, fully protected dinucleoside monophosphate (3)<sup>9</sup>, R<sub>f</sub><sup>14</sup>=0.66, in 87% isolated yield. We could then selectively unmask<sup>5,10</sup> the 5'-hydroxyl function by treating (3) for 1 h at 20°C with 0.2M HCl in dioxane solution (10 ml).



The selective hydrolysis of 3',5'-O-TIPDSi derivative (3) to 5'-hydroxy dinucleoside monophosphate (4)<sup>11</sup> was quantitative and the product (4)<sup>17</sup> was homogenous on TLC,  $R_f^{14}=0.62$ . This could be directly used for the second condensation reaction with a slight excess of (2) under a identical condition to that of the first condensation. The reaction resulted in a single product which was isolated in 94% yield after a usual work-up and a chromatography over a short column of silica gel. The product,  $R_f^{14}=0.69$ , thus obtained, could be conveniently identified as fully protected A2'p5'A2'p5'A (5) by <sup>1</sup>H-NMR spectroscopy<sup>12</sup>. The (5) was deprotected to (6) in the following order: (i) 4-nitrobenzaloximate ion in aq. dioxane<sup>13</sup> for 18h at 20°C, (ii) aq. NH<sub>3</sub>(d0.9) for 50h, (iii) n-Bu<sub>4</sub>NF (0.3M) in THF-pyridine 8:2,v/v at 20°C for 6h. The core was then purified through a DEAE-Sephadex A 25 column using Et<sub>3</sub>NH<sup>+</sup>HCO<sub>3</sub><sup>-</sup> (pH 7.6) for gradient elution (0.001M-0.6M) and was obtained in 91% isolated yield. Finally, the identity of the product was confirmed with an authentic sample<sup>4</sup> by <sup>1</sup>H-NMR, UV and TLC in two different solvent systems<sup>5</sup>:(a)  $R_f=0.9$  in i-Pr-OH:NH<sub>3</sub>(d0.9):H<sub>2</sub>O (55:109:35,v/v) and (b)  $R_f=0.64$  in i-butyric acid:NH<sub>3</sub>(d0.9):H<sub>2</sub>O (66:1:33,v/v). The product, A2'p5'A2'p5'A, was completely digested by Crotalus adamantus snake venom phosphodiesterase and was totally resistant to T<sub>2</sub> RNase which cleaves only 3'→5' diester bond.

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Tetrahedron 3075 (1980).
8.  $^1\text{H-NMR}$  ( $\text{CDCl}_3+\text{D}_2\text{O}$ ):  $\delta$  8.75(s, 1H), H-2, 8.2(s, 1H), H-8, 8.08(m, 2H), 1-&5-H of 6-N-Bz, 7.28(m, 4H), o-ClPh, 6.32(d,  $J_{1,2}=2.5\text{Hz}$ ), H-1', 5.4(m, 2H), H-2'&3', 4.3(m, 1H), H-4', 4.15(m, 2H), 5'- $\text{CH}_2$ , 3.05(q, 6H), 2.65(m, 4H) and 1.15(m, 33H).  $^{31}\text{P-NMR}$  ( $\text{CDCl}_3$ ): -6.0.
9.  $^1\text{H-NMR}$  ( $\text{CDCl}_3+\text{D}_2\text{O}$ ): 8.71(s, 1H), 8.65(s, 1H), H-2 protons, 8.45(s, 1H) & 8.27(s, 1H), H-8 protons, 8.02(m, 4H), 1-&5-H of 6-N-Bz, 7.44(m, 6H), 2-, 3-&4-H of 6-N-Bz, 7.25(m, 4H), o-ClPh, 6.4(d,  $J_{1,2}=2\text{Hz}$ , 1H) & 6.3(d,  $J_{1,2}=2.5\text{Hz}$ , 1H) are anomeric protons, 5.75(m, 3H), H-2'', H-2'&H-3', 5.08(m, 1H), H-3'', 4.6(m, 3H), 5'- $\text{CH}_2$ , & H-4'', 4.18(m, 1H), H-4', 4.06(m, 2H), 5''- $\text{CH}_2$ , 2.7(m, 4H), 2.06(s, 3H) & 1.94(s, 3H), two acetate groups, 1.06(m, 24H).
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11.  $^1\text{H-NMR}$  ( $\text{CDCl}_3+\text{D}_2\text{O}$ ):  $\delta$  8.77(s, 1H), 8.69(s, 1H), H-2 protons, 8.4(s, 1H) & 8.23(s, 1H), H-8 protons, 8.0(m, 4H), 1-&5-H of 6-N-Bz, 7.5(m, 6H), 2-, 3-&4-H of 6-N-Bz, 7.25(m, 4H), o-ClPh, 6.32(d,  $J_{1,2}=2.5\text{Hz}$ , 1H) & 6.24(d,  $J_{1,2}=1.8\text{Hz}$ , 1H) are assigned to two anomeric protons, 5.68(m, 3H), H-2'', H-2'&H-3', 4.91(m, 1H), H-3'', 4.28(m, 4H), H-4', H-4'' & 5'- $\text{CH}_2$ , 3.84(m, 2H), 5''- $\text{CH}_2$ , 2.7(m, 4H), 2.12 & 2.03 (two s, 3H each), 1.05(m, 24H).
12.  $^1\text{H-NMR}$  ( $\text{CDCl}_3+\text{D}_2\text{O}$ ):  $\delta$  8.76(s, 1H), 8.67(s, 1H) & 8.69(s, 1H), H-2 protons, 8.36(s, 1H), 8.33(s, 1H) & 8.31(s, 1H), H-8 protons, 8.12(m, 6H), 1-&5-H of 6-N-Bz, 7.5(m, 9H), 2-, 3-&4-H of 6-N-Bz, 7.3(m, 8H), o-ClPh, 6.45(d,  $J_{1,2}=3.5\text{Hz}$ , 1H), 6.28(d,  $J_{1,2}=3.1\text{Hz}$ , 1H), 6.22(d,  $J_{1,2}=3.2\text{Hz}$ , 1H) are anomeric protons, 5.7(m, 4H), H-2'', H-2'&H-2', H-3', 5.1(m, 2H), H-3' & H-3'', 4.5(m, 7H), H-4'', H-4'', H-4', 5'- $\text{CH}_2$  & 5''- $\text{CH}_2$ , 4.1(m, 2H), 5''- $\text{CH}_2$ , 2.6(m, 4H), 2.19(s, 3H) & 2.13(s, 3H), two acetate groups, 1.0(m, 24H).
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14. Merck Silica gel 60 F<sub>254</sub> pre-coated plates have been used for TLC in 10% Methanol -  $\text{CHCl}_3$  solvent system.
15. DC-plasticfolien cellulose F<sub>254</sub> sheets have been used.
16. HPLC analysis showed that (1) was pure and free of 2',3'-isomer (reverse phase C<sub>18</sub> column: linear gradient, H<sub>2</sub>O to 8%  $\text{CH}_3\text{CN}-\text{H}_2\text{O}$ ,  $R_T=11.7$  min for 3',5'-isomer (1) and  $R_T=9.8$  min for 2',3'-isomer).
17. HPLC (reverse phase C<sub>18</sub> column: linear gradient, H<sub>2</sub>O to 11%  $\text{CH}_3\text{CN}-\text{H}_2\text{O}$ ,  $R_T=14.3$  min) showed a single compound and thus, it is concluded that the cleavage of (3) to (4) was virtually regiospecific.

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