

A NEW STRATEGY FOR DINUCLEOTIDE SYNTHESIS VIA PHOSPHITE ROUTE INVOLVING PHOSPHOROCHLORIDATES
 AS INTERMEDIATES

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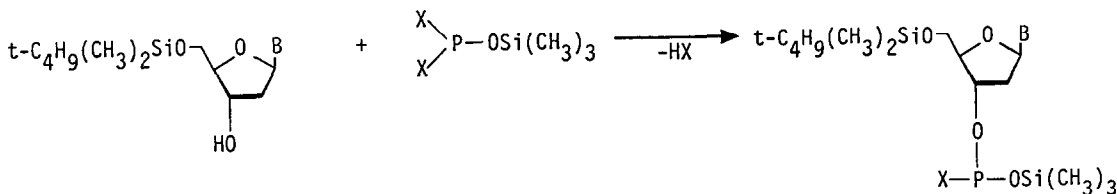
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Readily available nucleoside trimethylsilyl phosphites and analogous compounds are transformed in high yield into the corresponding phosphorochloridates by reaction with sulfuryl chloride. These compounds are employed as efficient reagents for internucleotide linkage formation.

One of the challenging areas of research in the chemistry of nucleic acids is the elaboration of methods for the chemical synthesis of oligo- and polynucleotides. The most important developments are connected with the phosphotriester approach and its recent extension via phosphite intermediates¹. Phosphite esters of nucleosides are readily available including those containing O-silyl protecting groups^{2,3}.

Based on our studies of the reactions of silyl phosphites with halogens, pseudohalogenes and halogenating agents⁴ we anticipated that trimethylsilyl phosphites derived from nucleosides could be transformed into the corresponding phosphorochloridates. The phosphorochloridates of this type have been postulated as reactive intermediates in formation of internucleotide linkages⁵.

The nucleoside phosphites 3 were synthesized by the reaction of a 5'-O-protected nucleoside 1 with bis(2,2,2-trifluoroethyl)trimethylsilylphosphite³ (2a) or bis(N,N-diisopropylamino)trimethylsilylphosphite (2b); we established the structure of this intermediate by both phosphorus and proton NMR (³¹P-NMR: +108.23 ppm, relative to the external standard 85% H₃PO₄; ¹H-NMR: 0.3 (s, 9H), 0.9 (d, 24H) and 3.0 - 3.7 (m, 4H) ppm in CDCl₃ relative to the internal standard tetramethylsilane).



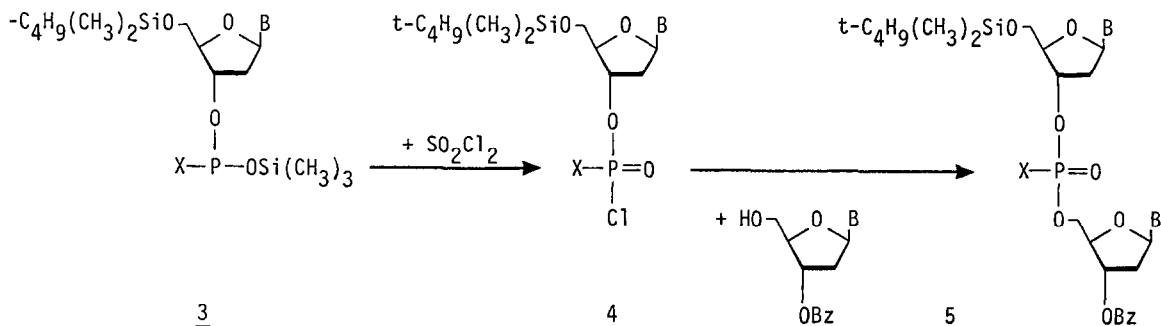
1 B = thymine or
 N(6)-benzoyladenine

2a X = CF₃CH₂O
2b X = [(CH₃)₂CH]₂N

3

The transformation of the silyl esters 3 into the chloridates 4 is smoothly accomplished by treating 3 with one equivalent of sulfuryl chloride. The reaction is quantitative and the volatile byproducts, trimethylsilyl chloride and sulfur dioxide, do not interfere with further applications of 4. The chloridate 4 is used as very efficient phosphorylating reagent towards

3'-protected nucleosides in the presence of a tertiary amine to give 5. This synthesis can also advantageously be performed without purification of the chloridate 4. This strategy promises to find application in the synthesis of oligonucleotides and other phosphates of biological interest. Moreover, the reaction is general for trimethylsilyl esters $>P-O-Si(CH_3)_3$ and provides access to the chloroanhydrides $>P(O)Cl$.



In a typical experiment 20 ml solution of 3 (0.01 mol) in dry CH_2Cl_2 was cooled to 0°C and a solution of SO_2Cl_2 (0.01 mol) in dry CH_2Cl_2 (20 ml) was added with stirring. The reaction mixture was kept 30 min at room temperature. The solvents, sulfur dioxide and trimethylchlorosilane were removed in vacuo. The residue was dissolved in CH_2Cl_2 (20 ml) and a solution of the 5'-hydroxyl component (0.01 mol) and triethylamine (0.01 mol) in CH_2Cl_2 (20 ml) was added. After 1 h the mixture was evaporated to a gum and dissolved in CH_2Cl_2 , washed with 0.1 M triethylammonium bicarbonate buffer once and then with water twice, dried (Na_2SO_4) and concentrated. The course of the reaction was monitored by thin layer chromatography on silica gel plates (Table 1), the products were purified by chromatography on silica gel columns.

Table 1. Properties of compounds 4 and 5^{a)}

X	Nucleobase B and B'	³¹ P-NMR ^{b)} of <u>4</u>		Yields of <u>5</u> (%)	R _f values of <u>5</u>
CF ₃ CH ₂ O	Thymine	+4.53,	+4.97	80	0.20, 0.25 ^{c)}
CF ₃ CH ₂ O	N(6)-Benzoyladenine	+4.83,	+5.12	83	0.23, 0.28 ^{c)}
[(CH ₃) ₂ CH] ₂ N	Thymine	+14.71,	+15.01	95	0.41, 0.48 ^{d)}
[(CH ₃) ₂ CH] ₂ N	N(6)-Benzoyladenine	+14.62,	+14.71	91	0.51, 0.54 ^{d)}

^{a)} All compounds 4 and 5 formed in the above reaction were identical with original samples prepared on different ways; ^{b)} Chemical shifts are given in ppm relative to the external standard 85% H_3PO_4 ; ^{c)} Eluents $\text{CHCl}_3/\text{MeOH}$, 5/1, v/v; ^{d)} Eluents $\text{EtOAc}/\text{CH}_2\text{Cl}_2/\text{Et}_3\text{N}$, 60/35/5, v/v.

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