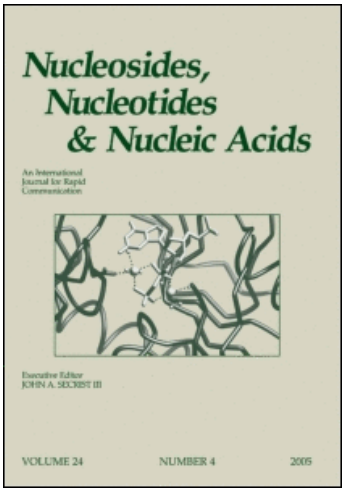

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SYNTHETIC APPROACHES TO OLIGODEOXYRIBONUCLEOSIDE PHOSPHORODITHIOATES USING TERVALENT PHOSPHORUS MONOMERS

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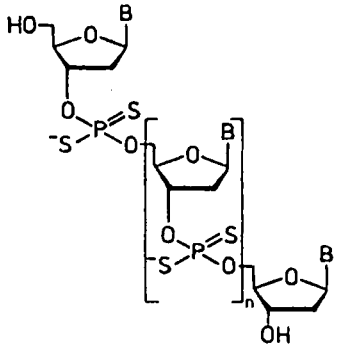
Abstract. Two routes to the title compounds using nucleoside phosphorodiamidites or thiophosphoramidites have been explored. A suitably substituted thymidine thiophosphoramidite could be used for tetrazole catalysed solid support syntheses of oligomers (2- to 10-mers).

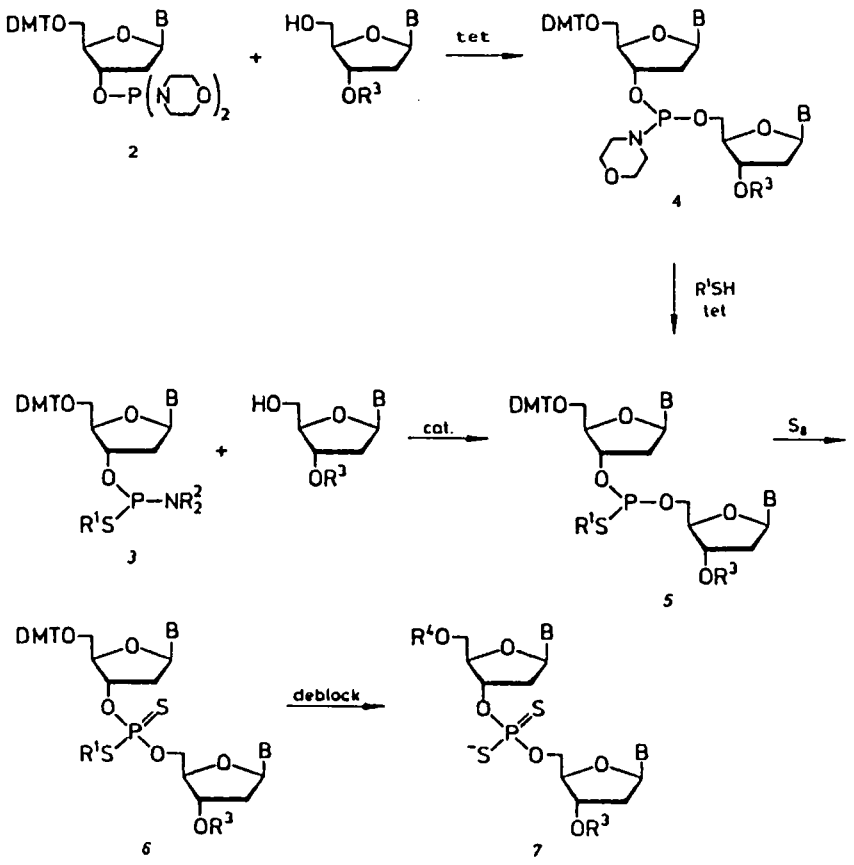
Modified oligodeoxyribonucleotides containing phosphorodithioate groups, 1, are promising alternatives to phosphorothioates as nuclease-resistant antisense probes since they



1

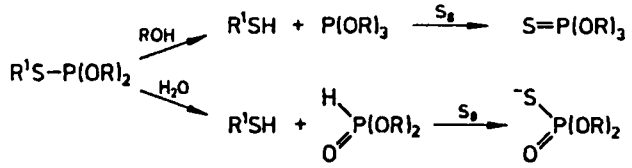
lack chiral phosphorus centers. The first member of this class, a thymidine dimer (1, n = 0, B = T), has recently been described.[1] We have investigated two potential routes to prepare 1 from the tervalent monomers 2 or 3 (Scheme 1).

In the first route, the known dinucleoside phosphoramidite 4 (R^3 = Ac, B = T)[2] was treated with a thiol (R^1 = 2-cyanoethyl or 2,4-dichlorobenzyl) and tetrazole in acetonitrile. A rapid reaction occurred to give 5 (δ_P 191.6-191.8, 2 dia-



Scheme 1

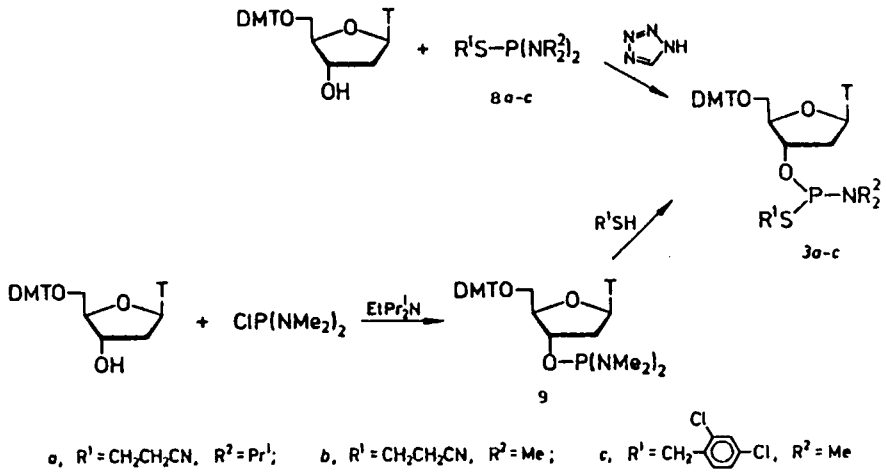
stereomers), and addition of S_8 to give 6 (δ_P 94.3-94.7, 2 diastereomers) and deblocking of the S-alkyl group with aq. NH_3 (R^1 = 2-cyanoethyl) or thiophenolate (R^1 = 2,4-dichlorobenzyl) to give 7 (R^4 = DMT, R^3 = ac, δ_P = 113-114.5, one isomer) proceeded without isolation of intermediates. According to ^{31}P NMR the crude yield of 7 was 50-80%, depending of the purity of 4 and the time lapse before S_8 addition. The main impurities were a dinucleoside H-phosphonate, a dinucleoside phosphorothioate, and a trinucleoside phosphorothioate, probably all formed by reaction of 5 with water or 3´-acetylthymidine:



The presence of these impurities indicates that R^1S is a leaving group comparable to $R^2{}_2N$ in the presence of tetrazole.

Attempts to use the monomer 2 for solid support syntheses of thymidine dimers gave low yields of 7 (R^3= R^4= H, B = T, 10-20% (^{31}P NMR) after deblocking). The main impurities were dithymidine phosphorothioate and thymidine H-phosphonate, indicating severe hydrolysis of 4 or 5 during coupling.

For the second route the thiophosphoramidites 3a-c were prepared (Scheme 2). DMTdT and 8a gave 3a (70% after column chromatography), which however was very unreactive in coupling experiments with tetrazole as catalyst. The more reactive 3b and 3c were then made from 9 (since 8b and 8c were difficult to purify) and isolated as fairly pure powders (85-90% according to ^{31}P NMR) after aq. $NaHCO_3$ extraction and precipitation from hexane ($\delta_P(CDCl_3)$ 172.6 and 170.9 for



Scheme 2

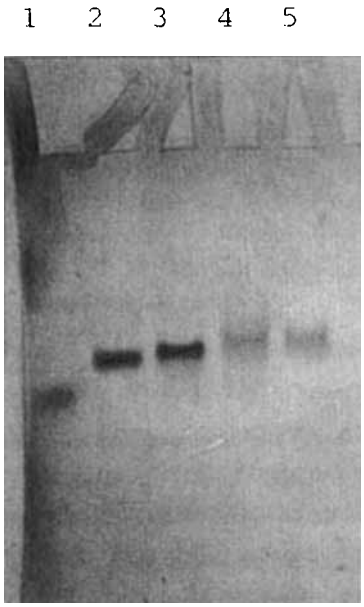


Fig. 1. PAGE (UV-shadowing), lane 1: Bromophenol blue, lane 2: $d(T)_{10}$, lane 3: $d(TpS)_9T$, lane 4 and 5: $d(TpS_2)_9T$.

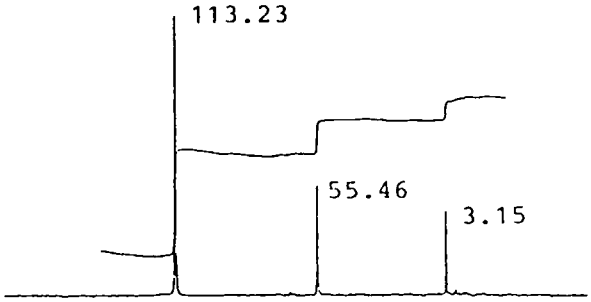


Fig. 2. ^{31}P NMR spectrum (D_2O) of crude $d(TpS_2)_9T$.

3b, 171.8 and 170.1 for 3c). Solution experiments showed that thymidine dimers 5 were formed rapidly with tetrazole catalysis (1-2 min), although subsequent side reactions consumed 5 slowly.

Solid support syntheses with 3b or 3c, using a standard amidite coupling cycle without capping (coupling 3 min, S_8 5 min), gave promising results, e.g. 1 (n=8, B=T) was synthesised with a mean DMT-efficiency of 99-101%. PAGE showed a main product running close to $d(T)_{10}$ and the phos-

phorothioate analogue (Fig. 1). However ^{31}P NMR showed that ca. 30% of phosphorus in the crude material was phosphorothioates or H-phosphonates (Fig. 2).

Work is in progress to examine the nuclease resistance and hybridisation properties of purified 1, and to extend the method to mixed sequences.

REFERENCES

1. Nielsen, J., Brill, W. K.-D., and Caruthers, M. H., Tetrahedron Lett., **1988**, 29, 2911.
2. Uznanski, B., Wilk, A., and Stec, W. J., Tetrahedron Lett., **1987**, 28, 3401.