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A novel approach to the synthesis of oligodeoxyribonucleotide boranophosphates

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ABSTRACT

Deoxyribonucleoside 3'-boranophosphate derivatives including adenine, cytosine, guanine, and thymine bases were synthesized in good yields by the use of a new boranophosphorylation reaction. The reaction was found to be effective for the formation of internucleotidic boranophosphate linkages.

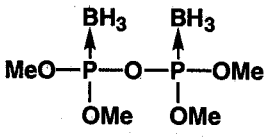
INTRODUCTION

Oligodeoxyribonucleotides bearing boranophosphate internucleotidic linkages (boranophosphate DNA) are regarded as potentially useful molecules for antisense therapeutics.[1] The conventional method for the synthesis of this DNA analog requires the construction of an oligonucleotide chain by the phosphoramidite or *H*-phosphonate approach, followed by boronation of the corresponding trivalent phosphite intermediates.[2-5] However, in the boronation step, undesirable side reactions occur at the base moieties by the borane reagents.[3,4] Therefore, the methods are applicable only to the thymine derivatives, which are less reactive to the borane reagents. In the present study, we wish to describe an alternative strategy for the synthesis of oligonucleotide boranophosphates by the use of a new boranophosphorylation reaction.

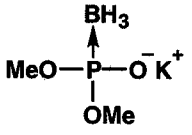
RESULTS AND DISCUSSION

Mononucleoside 3'-boranophosphate derivatives have not been used as starting materials for the synthesis of oligonucleotides having boranophosphate linkages to date. Imamoto *et al.* have employed tetramethyl boranopyrophosphate (**1**) and potassium dimethyl boranophosphate (**2**) as new reagents for boranophosphorylation of alcohols and organic halides.[6] Since the boranophosphorylating reagent **1** is less reactive to the nucleophilic attack of an alcohol, activation of the hydroxyl function should be required. In their case, *t*-BuLi was used as a strong base to generate the corresponding alkoxides.[6] In contrast to the above facts, we tried to activate the boranophosphorylating reagent **1** by using a nucleophilic catalyst. As a result, it was found that 3-nitro-1,2,4-triazole (NT)[7] was an effective activator for the boranophosphorylation of deoxyribonucleosides with **1**. When 5'-*O*-dimethoxytritylthymidine (**5t**) was allowed to react with **1** in the presence of NT and Et_3N in THF, 5'-*O*-dimethoxytritylthymidin-3'-yl dimethyl boranophosphate (**6t**) was obtained in 41% yield. In this reaction, a putative intermediate, dimethyl boranophosphoryl 3-nitro-1,2,4-triazolide **3** would be highly reactive and susceptible to hydrolysis.

Scheme 1



1



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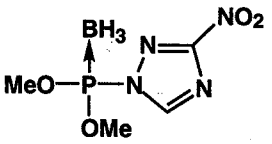

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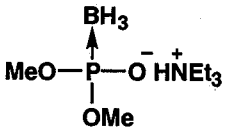

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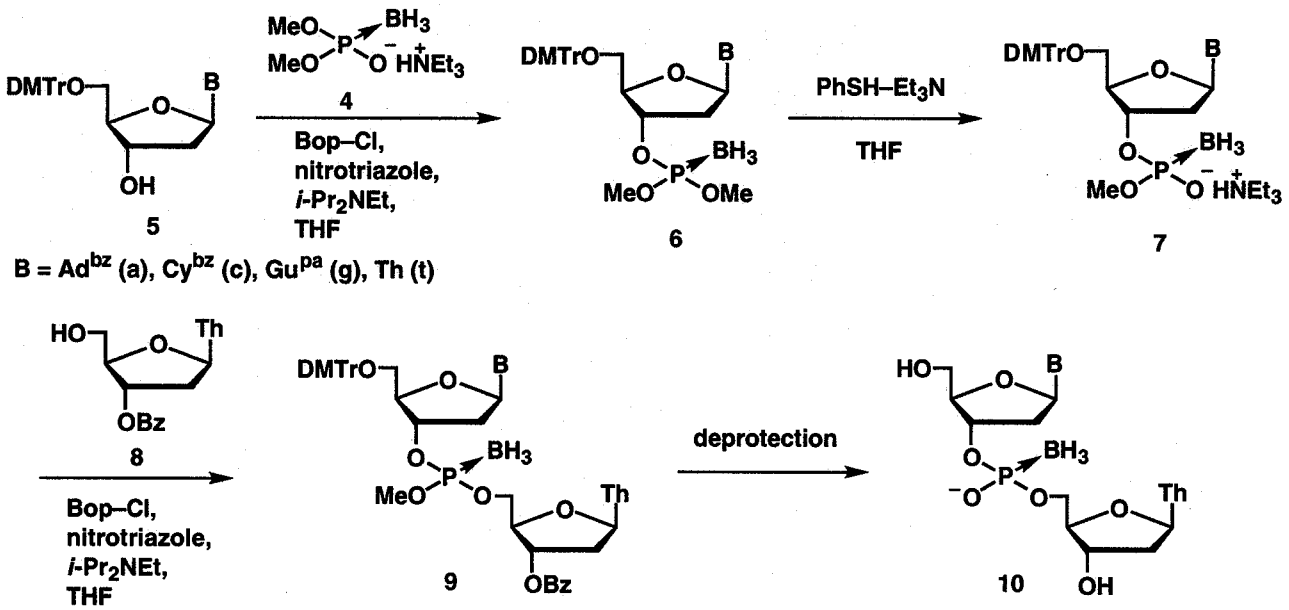
Consequently, the reactive intermediate **3** was tried to generate *in situ* with dimethyl boranophosphate and a condensing reagent to eliminate a trace of water in the reaction mixture. Triethylammonium dimethyl boranophosphate (**4**) was prepared in quantitative yield by boronation of dimethyl trimethylsilyl phosphite[8] followed by treatment with MeOH-Et_3N. The resulting **4** was condensed with **5t** in the presence of 1-mesitylenesulfonyl-3-nitro-1,2,4-triazole (MSNT)[9] and Et_3N. The reaction proceeded quickly, but a certain amount of the 5'-*O*-sulfonylated product was formed. In contrast, when *N,N'*-bis(2-oxo-3-oxazolidinyl)phosphonic chloride (Bop-Cl)[10] was employed as a condensing reagent with NT and Et_3N, the desired product **6t** was obtained in 84% yield without any side-products. In a similar manner, 5'-*O*-dimethoxytrityl-6-*N*-benzoyldeoxyadenosin-3'-yl dimethyl boranophosphate (**6a**) and 5'-*O*-dimethoxytrityl-4-*N*-benzoyldeoxycytidin-3'-yl dimethyl boranophosphate (**6c**) were synthesized from **5a** and **5c**, respectively. In the case of

Scheme 2



boranophosphorylation of deoxyguanosine derivative **5g**, the 6-*O*-boranophosphorylated product was observed to some extent by a TLC analysis. However, the dimethyl boranophosphoryl group at the 6-*O*-position was readily hydrolyze during the aqueous work-up to give the 5'-*O*-dimethoxytrityl-2-*N*-phenylacetyldeoxyguanosin-3'-yl dimethyl boranophosphate (**6g**) in good yield. Next, treatment of nucleoside 3'-dimethyl boranophosphate derivatives (**6**) with PhSH-Et_3N-THF (1:1:2, v/v/v)[11] afforded the demethyated products **7**. Condensation of the resulting diesters **7** with 3'-*O*-benzoylthymidine **8** in the presence of Bop-Cl, NT, and *i*-Pr_2NEt in THF gave the fully protected dimers **9** in good yields. Finally, all of the protecting groups in **9** were removed by the conventional procedure to yield the dinucleoside boranophosphates **10**. The present strategy will be useful for the synthesis of oligodeoxyribonucleotide boranophosphates including A, C, G, and T. Solid phase synthesis of oligomers is now in progress.

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