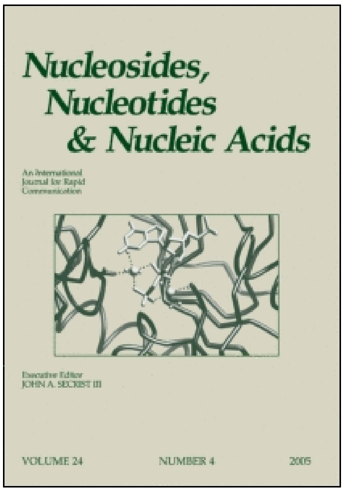

Nucleosides, Nucleotides and Nucleic Acids

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Synthesis of 5-Ethynyl-2'-Deoxyuridine-5'-Boranomono Phosphate as a Potential Thymidylate Synthase Inhibitor

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SYNTHESIS OF 5-ETHYNYL-2′-DEOXYURIDINE-5′-BORANOMONO PHOSPHATE AS A POTENTIAL THYMIDYLATE SYNTHASE INHIBITOR

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▫ *The 5-ethynyl-2′-deoxyuridine nucleoside and the 5′-boranomonophosphate nucleotide were synthesized as analogs of 5-fluoro-2′-deoxyuridine monophosphate (5-FdUMP), a widely used mechanism-based inhibitor of thymidylate synthase. Synthesis was carried out from protected 5-iodo-2′-deoxyuridine and trimethylsilylacetylene by Sonogashira palladium-catalyzed cross coupling reaction followed by selective phosphorylation and finally boronation.*

Keywords Oligonucleotides, Thymidylate Synthase, Boranomonophosphate

INTRODUCTION

Nucleoside analogs with modifications in the base, sugar, or phosphate residue can be used as probes for substrate properties, kinetic pathways, and transition states of enzymes involved in nucleic acid metabolism. 5-Fluorouracil (FU) and its derivative, 5-fluoro-2′-deoxyuridine monophosphate (5-FdUMP),[1] are strong inhibitors of thymidylate synthase (TS), an enzyme responsible for de novo synthesis of DNA thymidine and a major enzyme target for cancer therapeutics. Intrinsic and acquired drug resistance is a limitation of cancer chemotherapy using 5-fluoropyrimidines, which reduce the response rates. It is thus worthwhile to synthesize new analogs and their prodrugs combining the modifications at the phosphate center and C5 pyrimidine position. The substitution of a non-bridging oxygen in a phosphate ester group by a borane (BH_3) results in a class of phosphate modified nucleotides, e.g., boranomonophosphate.[2] Because the BH_3 group is isoelectronic with oxygen and isosteric with a methyl group, it shares a number of chemical and biochemical properties such as good aqueous solubility, increased lipophilicity, and phosphatase resistance. Boranophosphates of nucleosides and

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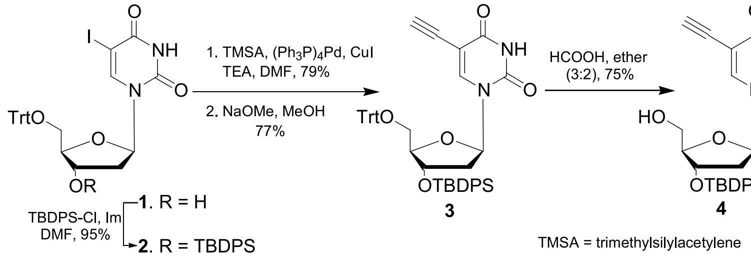
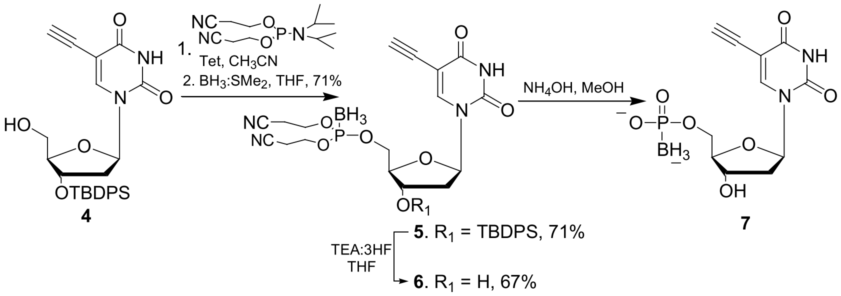


SCHEME 1 Synthesis of 5-ethynyl-3′-O-TBDPS-2′-deoxyuridine.

oligonucleotides have recently attracted attention due to their possible therapeutic applications as anticancer and antiviral[3,4] prodrugs and antisense agents.[5] The aim is to design and synthesize nucleotide prodrugs combining the best nucleoside inhibitors with the advantages of a boranophosphate modification. We hypothesize that nucleoside boranomonophosphates should resist further phosphorylation by nucleoside monophosphate kinase and dephosphorylation by alkaline phosphatase, and thus will increase the effective concentration of the active (monophosphate) drug in the cell.

5-Ethynyl-3′-O-TBDPS-2′-deoxyuridine nucleoside **4** was synthesized following Scheme 1 in 75% yield. Commercially available 5-iodo-2′-deoxyuridine was selectively protected at the 5′- and 3′-hydroxyls by trityl **1** and t-butyldiphenylsilyl **2**, respectively. The palladium mediated cross-coupling reaction was carried out using the Sonogashira method to obtain fully protected nucleoside, which after purification, was treated with sodium methoxide to deprotect the trimethylsilyl group to obtain **3** in 77% yield.

The nucleoside **3** was treated with formic acid and ether to remove the 5′-O-trityl to obtain the 3′-TBDPS protected nucleoside **4** in 75% yield. Finally, nucleoside **4** was phosphorylated by biscyanoethyldiisopropylphosphoramidite using tetrazole in CH_3CN as an activator followed by boronation by borane-dimethylsulfide complex, as shown in Scheme 2 to obtain compound **5** in 71% yield. The 3′-silyl protecting group was removed by TEA:3HF to obtain nucleotide



SCHEME 2 Synthesis of 5-ethynyl-2′-deoxyuridine-5′-boranomonophosphate.

6 in 67% yield. Compound **6** on treatment with NH_4OH/MeOH solution gave 5-ethynyl-2′-deoxyuridine-5′-borano monophosphate **7** in good yield.

In conclusion, of several approaches (not shown) tried to obtain **7**, the approaches described in Schemes 1 and 2 were found to be optimal for synthesis of the 5-ethynyl-2′-deoxyuridine-5′-boranomonophosphate. Biscyanoethyldiisopropylphosphoramidite was the best phosphorylating reagent to obtain the final boranomonophosphate nucleoside **7**.

REFERENCES

1. Kalman, T.I.; Nie, Z. Rational design of selective antimetabolites of DNA-thymine biosynthesis: 5-propynylpyrimidine nucleoside derivatives. Nucleosides Nucleotides Nucleic Acids **2000**, *19*, 357–369.
2. Shaw, B.R.; Dobrikov, M.; Wang, X.; Wan, J.; He, K.; Lin, J.; Li, P.; Rait, V.; Sergueeva, Z.A.; Sergueev, D. Reading, writing, and modulating genetic information with boranophosphate mimics of nucleotides, DNA, and RNA. Ann. N. Y. Acad. Sci. **2003**, *1002*, 12–28.
3. Dobrikov, M.; Sergueeva, Z.A.; Shaw, B.R. Incorporation of (α-*P*-borano)-2′,3′-dideoxycytidine-5-triphosphate into DNA by drug-resistant MMLV reverse transcriptase and *Taq* DNA polymerase. Nucleosides Nucleotides Nucleic Acids **2003**, *22*, 1651–1655.
4. Selmi, B.; Boretto, B.; Sarfati, S.R.; Guerreiro, C.; Canard, B. Mechanism-based suppression of dideoxynucleotide resistance by K65R human immunodeficiency virus reverse transcriptase using an α-boranophosphate nucleoside analogue. J. Biol. Chem. **2001**, *276*, 48466–48472.
5. Wang, X.; Dobrikov, M.; Sergueeva, D.; Shaw, B.R. RNase H activation by stereoregular boranophosphate oligonucleotide. Nucleosides Nucleotides Nucleic Acids **2003**, *22*, 1151–1153.