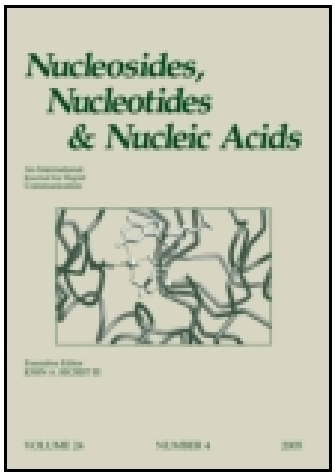


Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lncn20

MODEL SYNTHESIS OF NUCLEOSIDE BORANOPHOSPHORAMIDATE WITH AMINO ACID FOR PRODRUG PURPOSE

Ping Li [a] & Barbara Ramsay Shaw [a]
[a] Department of Chemistry , Duke University , Durham, North Carolina, USA
Published online: 15 Nov 2011.

To cite this article: Ping Li & Barbara Ramsay Shaw (2005) MODEL SYNTHESIS OF NUCLEOSIDE BORANOPHOSPHORAMIDATE WITH AMINO ACID FOR PRODRUG PURPOSE, Nucleosides, Nucleotides and Nucleic Acids, 24:5-7, 675-678, DOI: 10.1081/NCN-200060244

To link to this article: http://dx.doi.org/10.1081/NCN-200060244

Nucleosides, Nucleotides, and Nucleic Acids, 24 (5–7):675–678, (2005)

ISSN: 1525-7770 print/ 1532-2335 online
DOI: 10.1081/NCN-200060244

MODEL SYNTHESIS OF NUCLEOSIDE BORANOPHOSPHORAMIDATE WITH AMINO ACID FOR PRODRUG PURPOSE

Ping Li and Barbara Ramsay Shaw □ *Department of Chemistry, Duke University, Durham, North Carolina, USA*

□ *A model synthesis of a nucleoside boranophosphoramidate prodrug with (L)-tryptophan methyl ester was accomplished in a one-pot reaction via an H-phosphonate approach. This new type of compound is expected to possess the potent antiviral and anticancer advantages conferred by boranophosphates and normal nucleoside amino acid phosphoramidate.*

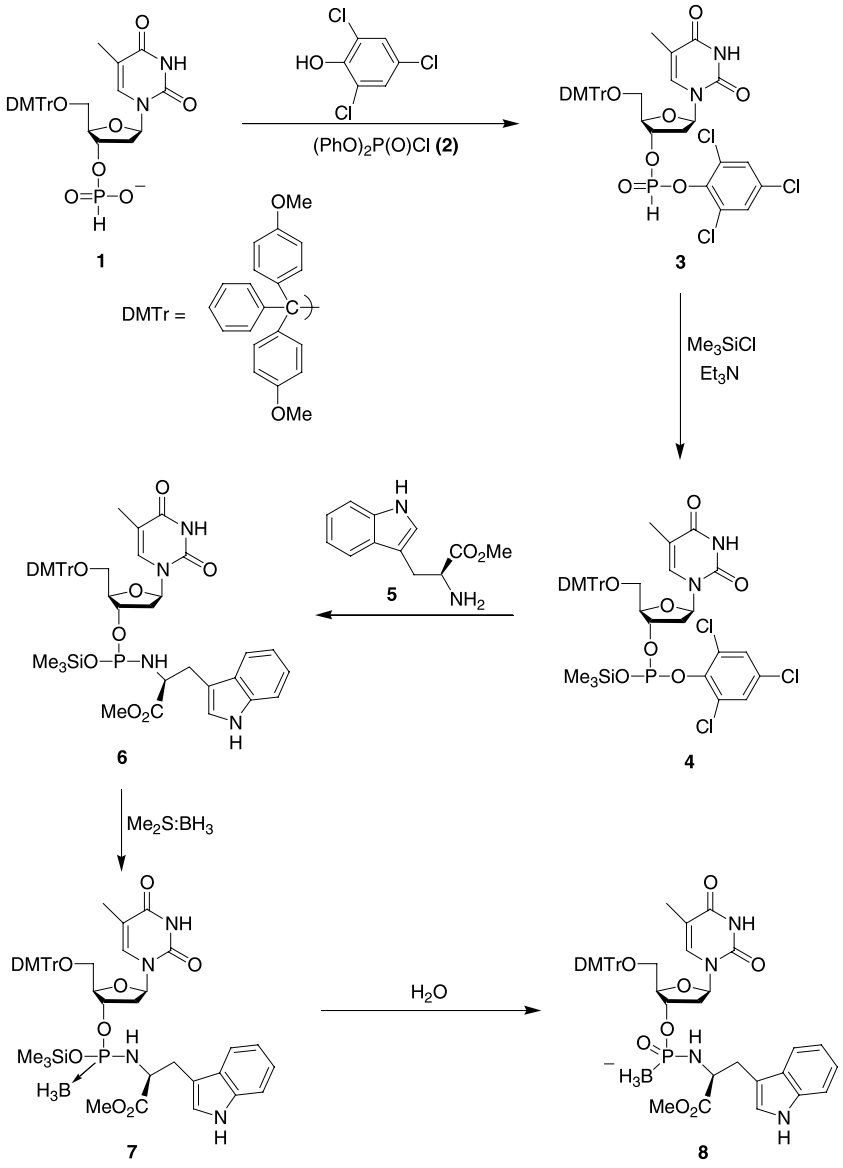
Keywords Nucleoside Boranophosphoramidate, Amino Acid, Prodrug, H-Phosphonate

INTRODUCTION

One of the focuses in current antiviral and anticancer research is the development of nucleotide prodrug or pronucleotide approaches, in which the prodrugs or pronucleotides could be delivered into cells, could bypass the initial kinase dependency step, and could be intracellularly activated to form the corresponding active nucleoside triphosphates.[1] Nucleoside amino acid phosphoramidates have shown promise as a potential pronucleotide strategy.[2,3] Studies by Wagner et al. have demonstrated that nucleoside phosphoramidate monoesters are potent antiviral and/or anticancer agents with enhanced activity yet reduced cytotoxicity.[4] These nucleoside phosphoramidate monoesters are thought to exert their biological functions through a P–N bond cleavage by phosphoramidases.[5] Nucleoside boranophosphates, in which one of the non-bridging oxygen atoms in the phosphate group is replaced by a borane (BH_3) group, have shown promising therapeutic applications in antiviral drug design.[6] For example, steady-state and pre-steady-state kinetic analyses indicate that whereas the efficiency of the single nucleotide incorporation for ddCTP by MMLV RT is nearly two orders lower than

Received October 2004, accepted February 2005.
This work was supported by an NIH grant (R01 AI52061) to B.R.S. and a Paul M. Gross Fellowship from Duke University to P.L.
Address correspondence to Barbara Ramsay Shaw, Department of Chemistry, Duke University, Box 90346, Durham, NC 27708-0346, USA; E-mail: brshaw@chem.duke.edu

for natural dCTP, the introduction of an α-boranophosphate group increases the efficiency of incorporation for ddCTPαB by 30-fold.[7] It is expected that combining nucleoside amino acid phosphoramidates and nucleoside boranophosphates would create a promising type of antiviral and anticancer prodrugs with the advantages conferred by both phosphoramidates and boranophosphates. In this communication, we present the model synthesis of a nucleoside boranophosphoramidate with an amino acid via an H-phosphonate approach.



SCHEME 1 Model synthesis of boranophosphoramidate with an amino acid.

RESULTS AND DISCUSSION

As shown in Scheme 1, in order to explore the viability of the H-phosphonate approach and to save time and cost, we started with the commercially available 5′-O-dimethoxytrityl (DMT) 2′-deoxythymidine 3′-H-phosphonate **1**. After reacting with 2,4,6-trichlorophenol in the presence of condensing reagent diphenyl chlorophosphate (DPCP, 2 equiv) **2**, a highly reactive intermediate, 2′-deoxy thymidine aryl H-phosphonate diester **3**, was formed. The reaction was completed in 1 hr as monitored by TLC. Silylation of H-phosphonate **3** with trimethylchlorosilane and excess triethylamine produced 2′-deoxythymidine phosphite triester **4**. The activated intermediate **4** was then subjected to aminolysis with tryptophanyl methyl ester **5** to afford the silylated nucleoside amino acid phosphoramidite **6**, whose ^{31}P NMR showed two peaks at δ 139.5 and 139.7 ppm corresponding to the diastereomers. Direct boronation of **6** with borane-dimethyl sulfide for 45 min resulted in the formation of the borane-complex **7**. The desired compound, nucleoside amino acid boranophosphoramidate **8**, was obtained after the reaction mixture was treated with H_2O for 30 min. The reaction was followed by ^{31}P NMR, where the broad peak centered at δ 118.0 ppm for intermediate **7** became a quartet peak at δ 93.5 ppm for boranophosphoramidate **8**. The condensing reagent DPCP was removed by repeated water extraction from dichloromethane solution containing final product **8**, which was further purified by silica gel chromatography in 32% yield.

In conclusion, we have accomplished the model synthesis of the nucleoside boranophosphoramidate with an amino acid, specifically 5′-O-dimethoxytrityl-2′-deoxythymidin-3′-yl boranophosphoramidate **8** conjugated with (*L*)-tryptophan methyl ester, in a one-pot reaction via an H-phosphonate approach with satisfactory yield. Due to the mildness of the reaction conditions, the H-phosphonate approach can be considered a general synthetic protocol for preparing nucleoside amino acid prodrugs with boranophosphoramidate linkages. Further investigation of this approach on other nucleoside analogues and amino acids is still in progress as well as studies on the chemical and biological properties of the corresponding boranophosphoramidates.

REFERENCES

1. De Clercq, E. Strategies in the design of antiviral drugs. Nat. Rev., Drug Discov. **2002**, *1*, 13–25.
2. Cahard, D.; McGuigan, C.; Balzarini, J. Aryloxy phosphoramidate triesters as pro-tides. Mini-Rev. Med. Chem. **2004**, *4*, 371–381.
3. Drontle, D.P.; Wagner, C.R. Designing a pronucleotide stratagem: lessons from amino acid phosphoramidates of anticancer and antiviral pyrimidines. Mini-Rev. Med. Chem. **2004**, *4*, 409–419.
4. Chang, S.L.; Griesgraber, W.; Southern, P.J.; Wagner, C.R. Amino acid phosphoramidate monoesters of 3′-azido-3′-deoxythymidine: relationship between antiviral potency and intracellular metabolism. J. Med. Chem. **2001**, *44*, 223–231.
5. Wagner, C.R.; Kim, J.; Chou, T.F. Direct measurement of antiviral nucleoside monophosphate delivery from phosphoramidate pronucleotides by stable isotope labeling and LC-ESI-MS/MS. Antivir. Res. **2004**, *62*, A27.
6. Shaw, B.R.; Dobrikov, M.; Wang, X.; Wan, J.; He, K.Z.; Lin, J.L.; Li, P.; Rait, V.; Sergueeva, Z.; Sergueev, D.

Reading, writing, and modulating genetic information with boranophosphate mimics of nucleotides, DNA, and RNA. Ann. N. Y. Acad. Sci. **2003**, *1002*, 12–29.

7. Dobrikov, M.I.; Sergueeva, Z.A.; Shaw, B.R. Incorporation of (alpha-*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.