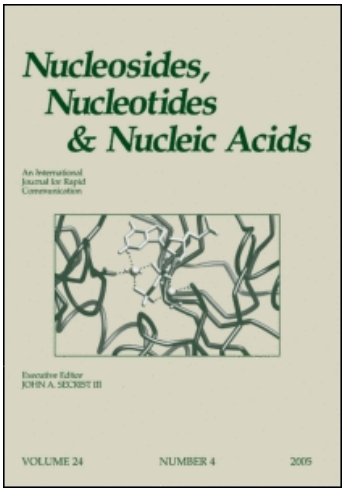

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Developing Synthetic Methods for Bioactive Phosphorus Compounds Using H-Phosphonate Chemistry: A Progress Report

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DEVELOPING SYNTHETIC METHODS FOR BIOACTIVE PHOSPHORUS COMPOUNDS USING H-PHOSPHONATE CHEMISTRY: A PROGRESS REPORT

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▫ *In this paper a short account of our recent basic studies aiming toward development of new synthetic methods for the preparation of nucleotide analogues using H-phosphonate chemistry is presented.*

Keywords H-phosphonates, Pyridylphosphonates, Stereospecific Synthesis

INTRODUCTION

Introduction of modifications at the phosphorus center of various natural products has emerged in recent decades as efficient means for modulation of their biological activity. As part of our program on exploring H-phosphonate chemistry for synthetic purposes, we have recently developed new efficient protocols for the preparation of nucleotide derivatives useful in the synthesis of oligonucleotides and their analogues.

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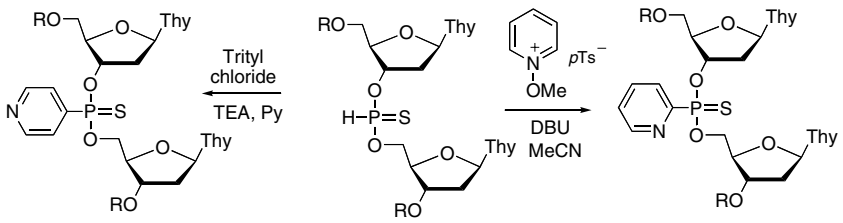


R = 4,4'-dimethoxytrityl; Thy = thymin-1-yl

SCHEME 1

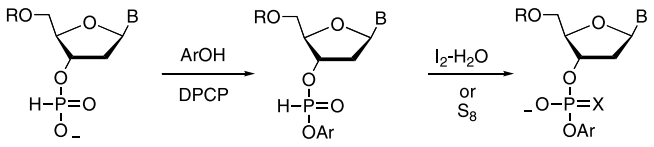
Pyridylphosphono*thio*ates—New Nucleotide Analogues

In contrast to pyridylphosphonates, their thiophosphonate counterparts are rare compounds and their biological properties are largely unexplored. Recently, synthetic and ^{31}P NMR studies led us to the development of efficient protocols for the stereospecific synthesis of this novel type of nucleotide analogues, 2-pyridyl- and 4-pyridylphosphonothioates.[1] The methods make use of easily available starting materials, dinucleoside H-phosphonothioate diesters, and afford the target pyridylphosphonothioates in high yields under mild reaction conditions. Comparing to dihydropyridylphosphonates,[2,3] the corresponding thiophosphonate derivatives are more stable and may need an additional re-aromatization step to be converted into the final pyridylphosphonates. The reaction of dinucleoside H-phosphonothioate diesters with N-methoxypyridinium salt showed some regioselectivity depending on the reaction conditions used. In the presence of DBU, the expected 2-pyridylphosphonate derivatives were formed, whereas in the presence of triethylamine, the major product of the reaction (ca. 80%) was the corresponding 4-pyridylphosphonate. The latter one is formed as the sole product when dinucleoside H-phosphonothioate diesters are treated with pyridine-trityl chloride reagent system in the presence of triethylamine (Scheme 1).

The underlying chemical reactions are stereospecific and can be extended to the preparation of other types of biologically important phosphorus compounds with this type of modifications.

Synthesis of Nucleoside Aryl Phosphates and Thiophosphates

We have developed a new, efficient method for the synthesis of aryl nucleoside phosphate and phosphorothioate diesters, based on H-phosphonate



B = a protected nucleobase; DPCP = diphenyl chlorophosphate; X = O or S

SCHEME 2

chemistry (Scheme 2). Starting from one type of substrate—easily accessible nucleoside H-phosphonates and phenols—and using a simple "one-pot" reaction, it was possible by changing the oxidation protocol to obtain, in high yields, aryl nucleoside phosphate or phosphorothioate diesters, derived from phenols of a wide range of acidity (e.g., 2 (4)-chlorophenyl, 2,4-, 3,4-, 3,5-dichlorophenyl, 2,4,6-trichlorophenyl, 4-nitrophenyl derivatives etc.).[4] These can be used as building blocks for oligonucleotide synthesis via the phosphotriester method or, in case of nucleosides with antiviral properties,[5] for the preparation of prodrugs for the corresponding nucleoside monophosphates.

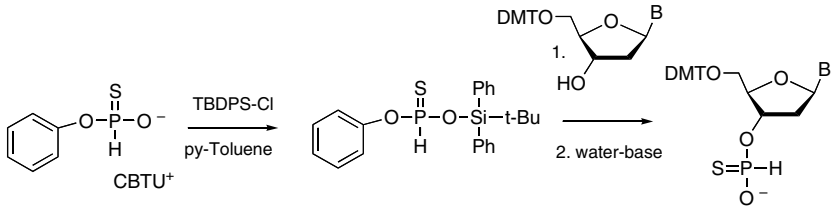
The method seems to be rather general and thus applicable to the preparation of other phosphorus-containing natural products and their analogues, or various alkyl aryl phosphates and phosphorothioates for synthetic and biochemical applications.

New Entry to Nucleoside H-Phosphonothioate Monoesters

Phenyl H-phosphonothioate monoester upon in situ silylation with *t*-butyldiphenylsilyl chloride in pyridine-toluene easily undergoes transesterification with suitably protected nucleosides to produce the corresponding 3′-H-phosphonothioates in good yields. The reagent is easy to prepare on a large scale via sulfohydrogenolysis[6] of the commercially available diphenyl H-phosphonate, is crystalline, and can be stored at room temperature for prolonged period of time (Scheme 3). This approach can be probably extended to other hydroxylic compounds and thus expands the array of synthetic methods available for the preparation of H-phosphonothioate monoesters.[7]

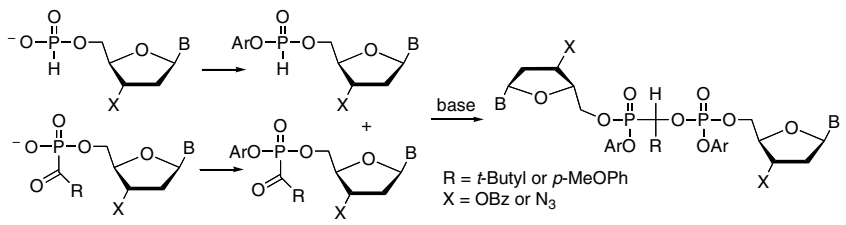
Nucleotide Analogues with Internucleosidic Phosphonate-Phosphate Bond

Searching for new nucleoside kinase bypass lipophilic pronucleotides,[8] we considered nucleotide analogues with a phosphonate-phosphate internucleoside bond as unique vehicles for delivery of biologically active nucleotides into the cell. On the synthetic part, we found that aryl nucleoside H-phosphonates and aryl nucleoside P-acylphosphonates, generated in situ from the appropriate H-phosphonate and acylphosphonate monoesters, respectively, reacted rapidly in



CBTU = p-chlorobenzylthiouronium; TBDPS-Cl = t-butyldiphenylsilyl chloride
DMT = 4,4'-dimethoxytrityl; B = aprotected nucleobase

SCHEME 3

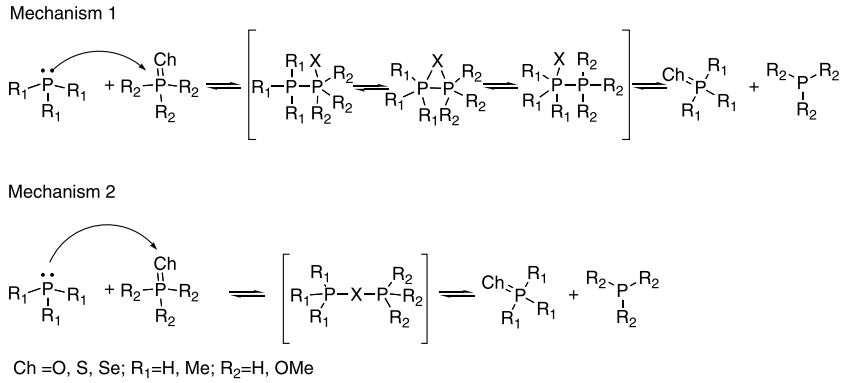


SCHEME 4

the presence of tertiary amines to produce in high yields the extended pyrophosphate analogues, diaryl dinucleoside phosphonate-phosphate derivatives.[9] These, depending on a substituent on the α-carbon of the phosphonate moiety afforded upon hydrolysis either the dinucleoside phosphonate-phosphate or the nucleoside H-phosphonates and aryl nucleoside phosphate. Since the corresponding AZT derivatives under mild conditions can generate AZT 5′-H-phosphonate and AZT 5′-phenyl phosphate, this type of compound can be considered as potential lipophilic prodrugs for delivery of one or two kinds of antiviral nucleoside monophosphates into the cell (Scheme 4).

Mechanism for the Chalcogens Exchange Reaction

Recently, we have proposed new selenium transferring reagents, namely triphenylphosphine selenide[10] and triphenyl phosphoroselenoate[11] for the introduction of selenium into nucleic acids. Regarding mechanistic aspects of the underlying reactions, two pathways for chalcogens transfer from P(V) to P(III) compounds were explored using density functional theory calculations (B3LYP, 6-31G*). For both mechanisms, the corresponding transition states were identified and characterized. The calculations showed that transfer of sulfur and selenium proceeds most likely via an X-philic attack of the phosphorus nucleophile on the



SCHEME 5

chalcogen, while for the oxygen transfer reaction, a mechanism involving a three-membered cyclic transition state is equally likely (Scheme 5).

The activation energies for the X-philic attack (Mechanism 1) show the expected trend that is in agreement with the observed order of reactivity of phosphine chalcogenides in the chalcogen exchange reaction between P(V) and P(III) compounds. Thus, the reactions involving transfer of oxygen had very high activation energy (ca. 56–69 kcal/mol), while those for sulfur and selenium transfer, were significantly less energy demanding (7–20 kcal/mol for sulfur and 4–10 kcal/mol for selenium). For the edge attack of phosphorus nucleophile on the P=Ch bond (Mechanism 2), very high activation energies (above 50 kcal/mol) were observed for all the chalcogens. For the oxygen transfer reaction the results are thus inconclusive, since attack of a phosphorus nucleophile on oxygen (Mechanism 1) and on the P=O group (Mechanism 2) both have similar activation energies. These energetic barriers are very high and effectively prevent reduction of P(V) phosphoryl compounds or oxidation of P(III) derivatives by phosphoryl compounds.

REFERENCES

1. Johansson, T.; Stawinski, J. Nucleoside H-phosphonates. Part 19: efficient entry to novel nucleotide analogues with 2-pyridyl- and 4-pyridylphosphonothioate internucleotide linkages. Tetrahedron **2004**, *60*, 389–395.
2. Kers, A.; Stawinski, J. A new type of nucleotide analogue with 4-pyridylphosphonate internucleotide linkage. Tetrahedron Lett. **1999**, *40*, 4263–4266.
3. Johansson, T.; Kers, A.; Stawinski, J. 2-Pyridylphosphonates: a new type of modification for nucleotide analogues. Tetrahedron Lett. **2001**, *42*, 2217–2220.
4. Cieslak, J.; Jankowska, J.; Sobkowski, M.; Wenska, M.; Stawinski, J.; Kraszewski, A. Aryl H-phosphonates. Part 13. A new, general entry to aryl nucleoside phosphate and aryl nucleoside phosphorothioate diesters. J. Chem. Soc., Perkin Trans. 1 **2002**, 31–37.
5. Sobkowski, M.; Cieslak, J.; Jankowska, J.; Stawinski, J.; Kraszewski, A. Dinucleoside aryl phosphorothioates as building blocks for large scale synthesis of chimeric oligonucleotide analogues. Collect. Symp. Ser. **2002**, *5*, 283–289.
6. Kers, I.; Kers, A.; Stawinski, J.; Kraszewski, A. Aryl H-phosphonates. 8. Simple and efficient method for the preparation of nucleoside H-phosphonothioate monoesters. Tetrahedron Lett. **1999**, *40*, 3945–3948.
7. Stawinski, J.; Kraszewski, A. How to get the most out of two phosphorus chemistries. Studies on H-phosphonates. Acc. Chem. Res. **2002**, *35*, 952–960.
8. Parang, K.; Wiebe, L.I.; Knaus, E.E. Novel approaches for designing 5′-O-ester prodrugs of 3′-azido-2′,3′-dideoxythymidine (AZT). Curr. Med. Chem. **2000**, *7*, 995–1039.
9. Szymczak, M.; Szymanska, A.; Stawinski, J.; Boryski, J.; Kraszewski, A. Aryl H-phosphonates. Part 14. Synthesis of new nucleotide analogues with phosphonate-phosphate internucleosidic linkage. Org. Lett. **2003**, *5*, 3571–3573.
10. Bollmark, M.; Stawinski, J. A new selenium-transferring reagent—triphenylphosphine selenide. Chem. Commun. **2001**, 771–772.
11. Kullberg, M.; Bollmark, M.; Stawinski, J. Triphenyl phosphoroselenoate—a new selenizing agent for P(III) compounds. Collect. Symp. Ser. **2002**, *5*, 290–294.