Chinese Journal of Chemistry, **2004**, *22*, 225—227 **Communication**

Novel and Convenient Approach to Synthesis of AZT/d4T H-phosphonates

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A convenient, efficient and general method has been developed for synthesis of H-phosphonate mono and diesters of AZT and d4T through one-pot reaction of phosphonic acid with AZT or d4T and different alcohols using pivaloyl chloride as condensing agent under mild conditions.

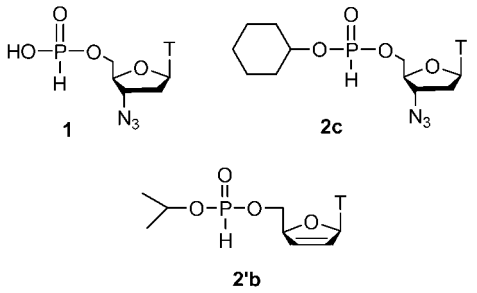
Keywords H-phosphonate, AZT, d4T, phosphonic acid, pivaloyl chloride

Intensive efforts have been made to develop effective chemotherapeutic agents against the human immunodeficiency virus (HIV).[1] Nucleoside analogues are widely used as antiviral agents in the treatments of AIDS and the AIDS related complex.[2] Currently, among the diversity of compounds, 3'-azido-3'-deoxythymidine (AZT) and 2',3'-dideoxy-2',3'-dehydrothymidine (d4T) were most extensively studied.[3] However, it has been proved that they must be phosphorylated intracellularly to their active triphosphates before acting as competitive inhibitors or alternate substrates (chain terminators) of HIV RT.[4] Furthermore, some clinical drawbacks such as the toxicity of AZT are subject to diurnal effects which may be related to monophosphorylation.[5] Consequently, in order to overcome the decreased intracellular phosphorylation, a number of methods have been developed for the delivery of monophosphorylated antiviral nucleoside.[6]

In various prodrugs, 5'-H-phosphonates of AZT and d4T as potent antiviral agents have shown promising, since in some cases they have exhibited enhanced antiviral activity and reduced cytotoxicity compared to the parent nucleosides. For example, 5'-H-phosphonate of AZT (Scheme 1, **1**), which is in Phase I clinical trials currently, is much less toxic than AZT, CC_{50} values of **1** and AZT are 2.5 mmol • L^{-1} and 210 μmol • L^{-1} respectively.[7] Selective indexes of AZT 5'-cyclohexylphosphite (Scheme 1, **2c**) and d4T 5'-isopropylphosphite (Scheme 1, **2'b**) are 18.9 and more than 6.11 times respectively compared with AZT and d4T.[8]

H-phosphonate monoesters were usually synthesized through the reaction of alcohols with phosphorus trichloride/triazole and the following hydrolysis in triethylammonium bicarbonate aqueous solution[9] or the mono-substitution of diphenyl phosphite by alcohols and the subsequent hydrolysis in triethylamine/water.[10] H-phosphonate diesters are prepared by condensation of H-phosphonate monoesters with alcohols in the presence of condensing agents[11] or hydrolysis of phosphoramidates in the presence of 1*H*-tetrazole.[12] But these methods suffered from laborious synthetic procedure, variable yields, and incompatibility with the common protecting groups utilized in natural product chemistry. Therefore, we try to develop a convenient, efficient, and general method for the preparation of H-phosphonate mono and diesters.

Scheme 1



In this paper, mono and diesters of H-phosphonates were synthesized through reaction of phosphonic acid with different alcohols in the presence of pivaloyl chloride (PV-Cl). As shown in Scheme 2, the general procedure was as follows: At room temperature, 1.1 equiv. of PV-Cl was added dropwise to the mixture of AZT or

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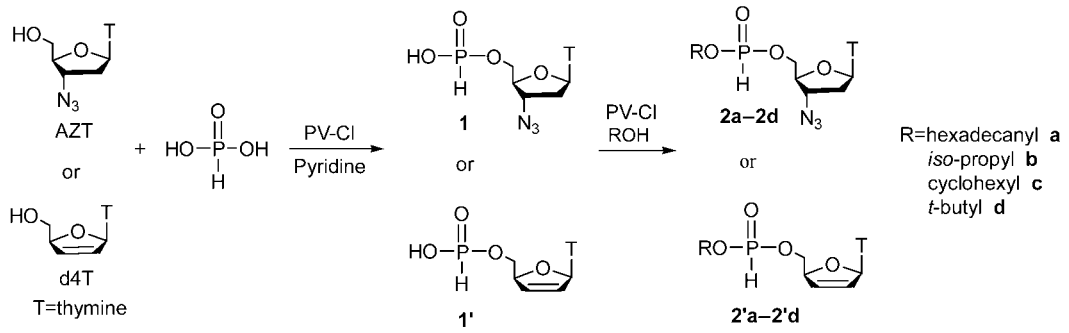
Scheme 2



d4T and 1.1 equiv. of phosphonic acid in pyridine, and the reaction completed in a few minutes. ^{31}P NMR spectrum showed more than 90% yields of **1** and **1'**. Further condensation of **1** or **1'** with 2 equiv. of alcohols in the presence of PV-Cl produced **2a**—**2d** and **2'a**—**2'd** in 53%—64% yield.

A typical procedure is described as follows for the preparation of compounds **1** and **2b**. Phosphonic acid (90.2 mg, 1.1 mmol) and AZT (267 mg, 1 mmol) were dissolved in 5 mL of anhydrous pyridine and co-evaporated twice, and then dissolved in 5 mL of fresh anhydrous pyridine. Pivaloyl chloride (132.6 mg, 1.1 mmol) in 2 mL of pyridine was added dropwise to the above solution under N_2 atmosphere at room temperature. About 5 min later, ^{31}P NMR spectrum revealed that there were a strong peak at δ 5.3 and a minor peak at δ 10.03 corresponding to **1** and symmetrical di-AZT H-phosphonate respectively. Without isolation, isopropyl alcohol (72 mg, 2 equiv.) was added to the above solution, and then pivaloyl chloride (241 mg, 2 equiv.) in pyridine was dropped at room temperature. 10 min. later, ^{31}P NMR spectrum showed that the peak at δ 5.3 disappeared, and a pair of peaks at δ 7.72 and 7.16 appeared corresponding to the pair of diastereoisomers of H-phosphonate **2b** of AZT due to the chirality of phosphorus. After evaporation of pyridine and the crude product was purified by column chromatography, and **2b** was obtained in 62% yield.[13] Other *O*-alkyl-5'-*H*-phosphonates of AZT and d4T were synthesized in moderate yields as shown in Table 1 using the similar method.

In summary, H-phosphoante mono and diesters of AZT and d4T were synthesized in an easy one-pot reaction using phosphonic acid as starting material in the presence of pivaloyl chloride. Compared with other methodologies, this method is fast, convenient, efficient under mild conditions. Both phosphonic acid and pivaloyl chloride are commercially available and inexpensive reagents, therefore, this method could be applied to the large-scale production of H-phosphonate mono and diesters, such as symmetric and asymmetric dialkyl H-phosphonate, dinucleoside H-phosphonate, and carbohydrate H-phosphonate. Further study of this method is in progress.

Table 1 Compounds of 5'-H-phosphonates of AZT and d4T

Comp.	R	^{31}P NMR[a]		ESI-MS		Yield/%
		δ	$^1J_{P\text{-}H}$	$(M+H)^+$	$(M+Na)^+$	
1	H	5.30	670	332	354	91
1'	H	5.34	676	289	311	93
2a	Hexadecyl	9.21, 8.84	709	556	578	64
2b	*iso*-Propyl	7.72, 7.16	707	374	396	62
2c	Cyclohexyl	7.71, 7.04	707	414	436	59
2d	*t*-Butyl	3.75, 3.13	708	388	410	53
2'a	Hexadecyl	9.17, 8.77	701	513	535	65
2'b	*iso*-Propyl	7.59, 7.08	704	331	353	60
2'c	Cyclohexyl	7.53, 7.12	700	371	393	60
2'd	*t*-Butyl	3.92, 3.08	698	345	367	54

[a] The values were determined in $CDCl_3$ using a Bruker AMP 200 at 81 MHz (85% H_3PO_4 as external standard).

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13 Characteristics are given for a representative compound: **2b**: ^{1}H NMR ($CDCl_3$, 500 MHz) δ: 9.53 (br, 1H, H-3), 6.90, 6.89* (d, $^1J_{P\text{-}H}$=704 Hz, 1H, P-H), 7.36, 7.33* (s, 1H, H-6), 6.17, 6.16* (s, 1H, H-1'), 4.78 (t, J = 6.5 Hz, 1H, C**H**$(CH_3)_2$), 4.34—4.0 (m, 4H, CH_2-5', H-4', H-3'), 2.44—2.32 (m, 2H, H-2'), 1.90 (s, 3H, CH_3-5), 1.36—1.34 (m, 6H, CH(C**H**$_3)_2$); ^{13}C NMR ($CDCl_3$, 125 MHz) δ: 163.85, 150.29, 135.35, 111.54, 84.98, 82.13, 72.35, 64.06, 60.00, 37.34, 23.76, 12.47. HRMS (ESI) calcd for $C_{13}H_{21}N_5O_6P$ $(M+H)^+$ 374.1224, found 374.1228. **2'a**: ^{1}H NMR ($CDCl_3$, 500 MHz) δ: 9.34 (br, 1H, H-3), 6.83, 6.81* (d, $^1J_{P\text{-}H}$=703 Hz, 1H, P-H), 7.28, 7.24* (s, 1H, H-6), 7.01 (s, 1H, H-1'), 6.30 (d, J=4.5 Hz, 1H, H-2'), 5.89 (s, 1H, H-3'), 4.98 (s, 1H, H-4'), 4.31—4.20 (m, 2H, OC**H**$_2(CH_2)_{14}CH_3$), 4.11—4.02 (m, 2H, H-5'), 1.87 (s, 3H, CH_3-5), 1.67—1.62 (m, 2H, OCH_2C**H**$_2(CH_2)_{13}CH_3$), 1.29—1.21 (br, 26H, OCH_2CH_2-(C**H**$_2)_{13}CH_3$), 0.84 (t, J = 6.5 Hz, 3H, OCH_2CH_2-$(CH_2)_{13}$C**H**$_3$); ^{13}C NMR ($CDCl_3$, 125 MHz) δ: 163.94, 150.88, 135.82, 132.87, 127.78, 111.24, 89.58, 84.53, 66.48, 65.47, 31.86, 30.33, 29.62—29.30, 25.38, 22.63, 14.07, 12.34. HRMS (ESI) calcd for $C_{26}H_{46}N_2O_6P$ $(M+H)^+$ 513.3088, found 513.3083. * The starred peak represents the other of diastereoisomers.

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