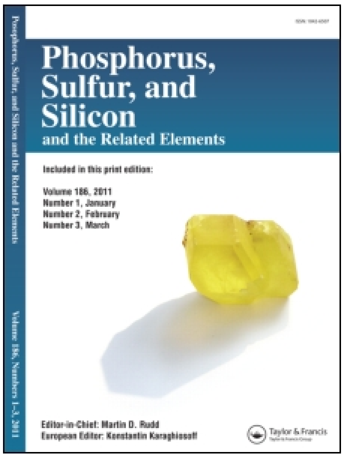

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Synthesis and Characterization of Novel Bile Acids Derived *H*-Phosphonates Conjugates

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Synthesis and Characterization of Novel Bile Acids Derived *H*-Phosphonates Conjugates

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The H-phosphnates of bile acid, conjugated with several alcohols and nucleosides, were synthesized in one pot by a tandem transesterification with diphenyl phosphate. The structures of these novel bile acid conjugates were confirmed by IR, ^{31}P NMR, ^{1}H NMR and HRMS.

Keywords Bile acid; *H*-phosphonates; transesterification

INTRODUCTION

Bile acids are natural origin compounds and play a very important role in biological systems.[1] The medicinal applications of bile acids and their conjugates were reported in numerous literature,[2] such as antiviral and the antifungal properties,[3] absorption enhancersm[4] and carriers of liver-specific drugs.[5] Their biological importance, the amphiphilic properties, the rigid concave of the backbone, and the unique disposition of hydroxyl groups make these compounds attracting building blocks for the design of synthetic receptors.[5]

Cholic acid **1** is one of the most commonly occuring bile acids (Scheme 1).[2] It possesses one carboxylic acid and three hydroxyl groups, which renders various chemical modifications possible. However, one of the major problems associated with the synthesis of cholic acid derivatives is that the "natural" hydroxyl groups is relatively inert and difficult to derivatise, especially the 7α- and 12α-OH.[6] Several studies have reported the conversion of the hydroxyl groups to other functional components, which result in novel amphiphilic structures with new properties.[6–8]

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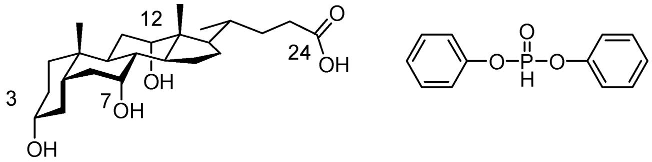
In this respect, we report the preparation of bile acid–derived poly-*H*-phosphonates. *H*-Phosphonate has proven to be a versatile building block in organic syntheses, which facilitate the conversion of hydroxyl groups into multifunctional components. It can be further transferred to phosphates, phosphorothionates, and phosphoroselenoates by oxidation[9] and phosphoramidate conjugates by Atherton-Todd reaction in high yield.[10] Previously, we have reported the synthesis of many bioconjugates with diphenyl phosphite **2** (DPP) as a phosphorylation reagent.[9–11] DPP can undergo fast transesterification with various alcohols in pyridine. In order to obtain mono-phenyl H-phosphonates, a large access of DPP (7 eqiv.) was required to prevent the formation of double-exchange *H*-phosphonates.[11] However, if one of the hydroxyl components is sterically hindered, the desired mono-phenyl *H*-phosphonate could be obtained in high yield with equal amounts of DPP.[12] The hydroxyls in the bile acids are oriented along one face and are typically steric hindered groups. Taking account of these facts, bile acids are ideal starting templates that result in a novel multifunctional scaffold containing phosphorus. It is worth noting that a few *H*-phosphonates and phosphoramides derivatives, such as carbohydrates[13] and nucleic acids,[12] have been fully studied. To the best of our knowledge, however, there is little information about the synthesis and properties of steroid *H*-phosphonates conjugates.

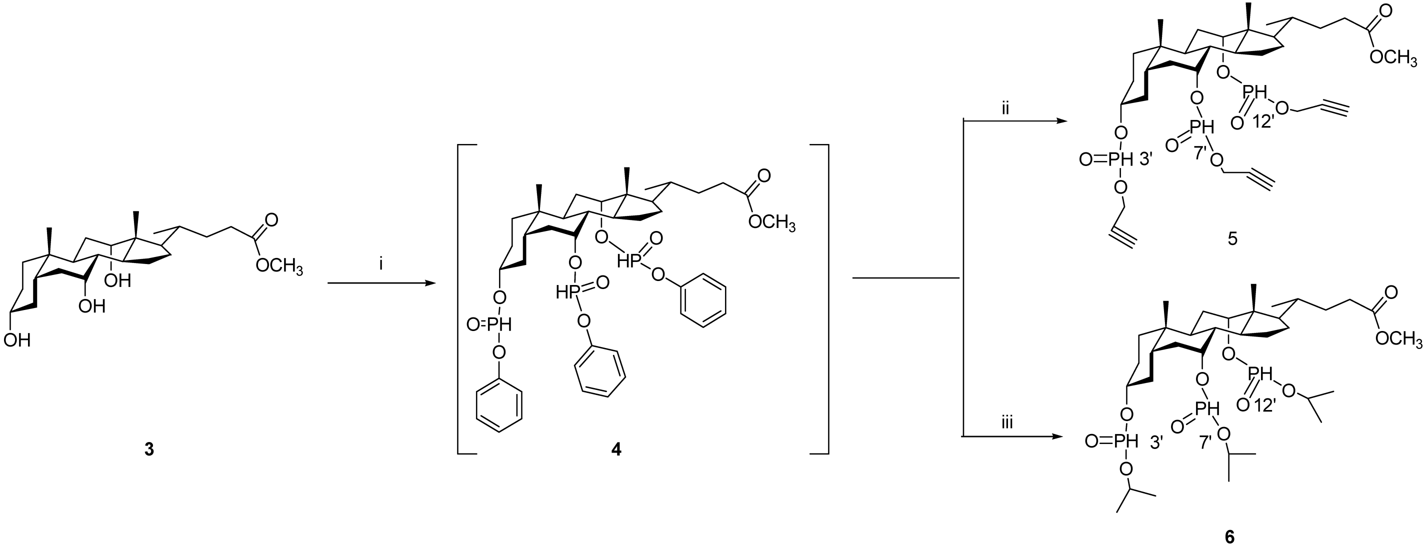


cholic acid **1** DPP **2**

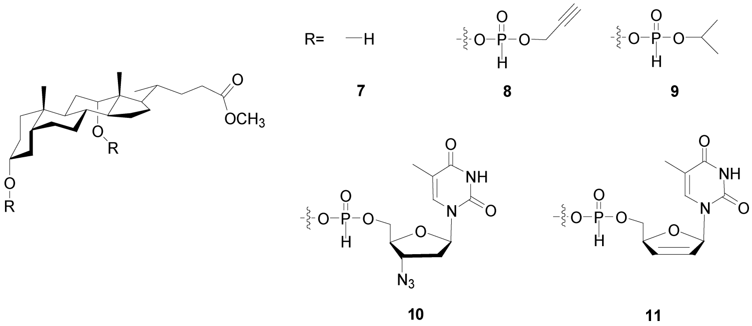
SCHEME 1 The chemical structure of cholic acid and diphenyl.

RESULTS AND DISCUSSION

The general synthetic strategy for bile acid *H*-phosphonates conjugates was shown in Schemes 2 and 3. Our approach starts from methyl cholate because the methyl esters can be prepared easily in large quantities. However, many other bile acid esters and amides are also appropriate starting materials. The reaction was monitored by ESI-MS and ^{31}P NMR. As is shown in Scheme 2, the first step of transesterification is fast and the all the hydroxyl groups were almost quantitatively converted to mono-phenyl *H*-phosphonates **4** after 5 h under anhydrous condition. The second step of transesterification is relatively slow and sensitive



SCHEME 2 The synthesis and the chemical structure of cholic acid derived *H*-phosphnates. (i) DPP (3 eqiv.)/py, 0°C, 5 h; (ii) propargyl alcohol (3 eqiv.)/py, 7 h; and (iii) *i*-PrOH (3 eqiv.)/py, 7 h.



SCHEME 3 The synthesis and the chemical structure of deoxycholic acid derived.

to steric hindrance, thereby preventing the formation of disteroid *H*-phosphonate.

The deoxycholic acid–derived *H*-phosphonate conjugates were synthesized in the similar procedure. It is worth mentioning that bile acids are interesting drug carriers that target a specific organ and improve the lipophilicity of drug molecules.[2] The bioactive nucleosides such as 3′-Azido-3′-deoxythymidine (AZT) and 2′, 3′-didehydro-2′, 3′-dideoxythymidine (d_4T), can be attached to deoxycholic methyl ester using this method. These molecules contain both steroid and nucleosides and have the potential to act as prodrugs or drugs by themselves.[14] However, the three bulky nucleoside moieties cannot be attached to the steroid nucleus simultaneously due to the large steric hindrance of cholic acid. When the same reaction was applied to cholic acid methyl ester, it only results in a complex mixture that is unlikely to be purified on a column chromatography of silica gel to obtain the pure product.

The structures of all these new *H*-Phosphonates and phosphoramide conjugates of bile acids were confirmed by IR, ^{31}P NMR, ^{1}H NMR, and HRMS. The NMR spectra shown in Figure 1 clearly present the shifts of protons among these cholic acid derivatives. In the downfield of ^{1}H NMR spectra, the three protons that were directly connected with phosphorus are easily confirmed by the large $^{1}J_{P\text{-}H}$ coupling constant (700 Hz approximately).

In conclusion, bile acids, which possess up to three sterically hinder hydroxyl groups, have provided a unique structure for the synthesis of poly-*H*-phosphonates derivatives. The extension of this methodology may find applications in pharmacology and supramolecular chemistry.

EXPERIMENTAL

Cholic acid, deoxycholic acid, and diphenyl phosphite (DPP) were purchased from Sigma Company and used without further purification.

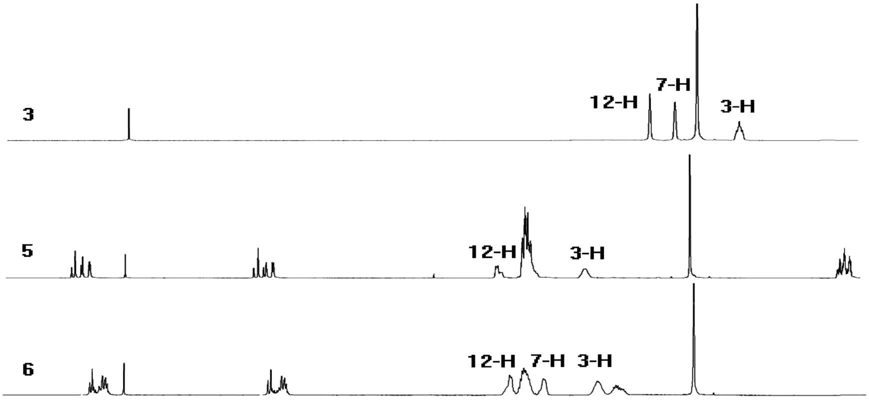


FIGURE 1 Comparison of the 600 MHz ^{1}H NMR spectra of compounds **3**, **5**, and **6** in $CDCl_3$.

Cholic acid methyl ester (CAME, **3**) and deoxycholic acid methyl ester (DCAME, **7**) were synthesized by refluxing appropriate bile acid in methanol catalyzed by hydrochloric acid and followed by a recrystallization, which was adapted to a reported procedure.[15] Pridine was purchased from Beijing Chemicals Co. and redistilled prior to use. Typical procedure for the synthesis and the spectrum data of bile acid dimers are represented by conjugate **5**.

Preparation of Methyl 3α, 7α, 12α-tri(propargy-H-phosphonate)-5β-cholan-24-ate (5)

CAME (254 mg, 0.6 mmol) was dissolved in pyridine 5 ml. Under an argon atmosphere, the solution was added slowly to DPP (0.534 g, 2.0 mmol) in 4 ml dry pyridine and cooled in an ice bath. The mixture was then stirred at room temperature for 5 h and the mono-phenyl bile acid triphosphite was obtained. Then the propargyl alcohol (0.129 ml, 2.2 mmol) was added and was stirred for additional 7 h. The obtained residue was purified on a column (silica gel; CH_2Cl_2: AcOEt = 8:3) to provide pure tri-*H*-phosphonates (244 mg, 0.34 mmol, light-yellow oil) in 56% yield. R_f: 0.5 (CH_2Cl_2:CH_3OH=20:1); IR (cm^{-1}): 3448.5, 3293.2, 3227.9, 2946.7, 2946.4, 2872.6, 2434.4, 2125.1, 1735.2, 1251.2, 977.3; ^{31}P NMR (81 MHz, $CDCl_3$): $\delta = 6.92, 7.25, 7.99, 8.08$; ^{1}H NMR (600 MHz, $CDCl_3$): $\delta = 0.67$ (s, 3H, 18–CH_3), 0.85 (s, 3H, 19–CH_3), 0.91 (d, 3H, 21–CH_3), 0.98–1.97 (m, 20H, aliphatic H), 2.14–2.32 (m, 4H, 2,4–CH_2), 2.56–2.64 (m, 3H, 3 × $\equiv$ C–H), 3.60 (s, 3H, OCH_3), 4.26 (m, 1H,

3α–CH), 4.59–4.68 (m, 7H, $3\times$–$CH_2C\equiv CH$, 7α–CH), 4.82 (m, 1H, 12α–CH), 6.86 (d, 1H, $J = 710$ Hz, $3'$–PH), 6.91, 6.93*(d, 1H, $J = 710$Hz, $7'$–PH), 6.96, 6.99* (d, 1H, $J = 710$ Hz, $12'$–PH); HRMS (ESI) found: 946.2994, $[C_{34}H_{51}O_{11}P_3+NH_4]^+$ calcd: 946.2982.

REFERENCES

[1] J. Sjövall, *Lipids,* **39,** 703 (2004).
[2] A. Enhsen, W. Krame, and G. Wess, *Drug Discovery Today*, **3,** 409 (1998).
[3] D. B. Salunke, B. G. Hazra, V. S. Pore, M. K. Bhat, P. B. Nahar, and M. V. Deshpande, *J. Med. Chem.*, **47,** 1591 (2004).
[4] B. G. Hazra, V. S. Pore, S. K. Dey, S. Datta, M. P. Darokar, D. Saikia, S. P. S. Khanuja, and A. P. Thakur, *Bioorg. Med. Chem. Lett.,* **14,** 773 (2004).
[5] E. Virtanen and E. Kolehmainen, *Eur. J. Org. Chem.*, 3385 (2004).
[6] V. Amo, L. Siracusa, T. Markidis, K. M. Bhattarai, M. Galobardes, G. Naredo, and A. P. Davis, *Org. Biomol. Chem.*, **2,** 3320 (2004).
[7] A. P. Davis and M. N. Pérez-Payán, *Synlett*, 991 (1999).
[8] C. H. Li, A. S. Peters, E. L. Meredith, G. W. Allman, and P. B. Savage, *J. Am. Chem. Soc.*, **120,** 2961 (1998).
[9] C. B. Xue, Y. W. Yin, and Y. F. Zhao, *Tetrahedron. Lett.*, **29,** 1145 (1988).
[10] Q. Xiao, J. Sun, Y. Ju, Y. F. Zhao, and Y. X. Cui, *Synthesis*, **1,** 107 (2003).
[11] Q. Xiao, J. Sun, Y. Ju, Y. F. Zhao, and Y. X. Cui, *Chem. Lett.*, **32,** 522 (2003).
[12] I. Kers, J. Cieslak, J. Jankowska, A. Kraszewski, and J. Stawinski, *Nucleosides & Nucleotides*, **18,** 1245 (1999).
[13] Y. Ju, Q. Xiao, and Y. F. Zhao, *Heteroatom. Chem.*, **14,** 208 (2003).
[14] V. M. F. Cardona, A. I. Ayi, and A. M. Aubertin, *Antivir. Res.*, **42,** 189 (1999).
[15] S. Gouin and X. X Zhu, *Steriods*, **61,** 664 (1996).