JOC Note
pubs.acs.org/joc

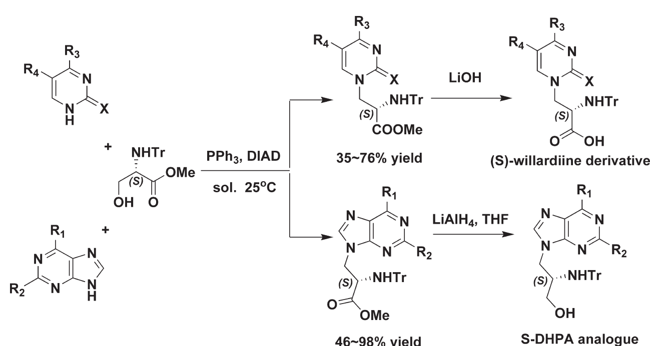
Synthesis of Acyclic Nucleosides with a Chiral Amino Side Chain by the Mitsunobu Coupling Reaction

Hai-Ming Guo,*,† Yan-Yan Wu,† Hong-Ying Niu,‡ Dong-Chao Wang,† and Gui-Rong Qu*,†

†*College of Chemistry and Environmental Science, Key Laboratory of Green Chemical Media and Reactions of Ministry of Education, Henan Normal University, Xinxiang 453007, Henan, China, and* ‡*School of Chemistry and Chemical Engineering, Henan Institute of Science and Technology, Xinxiang 453003, China*

ghm@htu.cn; quguir@sina.com.cn

Received March 3, 2010



A novel and efficient synthetic method has been developed for the preparation of chiral acyclic nucleosides with a chiral amino side chain by Mitsunobu reaction between nucleoside bases and protected L-serine. This method suggests a potentially more efficient and complementary process to acquire chiral acyclic nucleosides.

Acyclic nucleoside and nucleotide analogues are currently used as antiviral drugs possessing a broad spectrum of activities. The discovery of acyclovir[1] has stimulated extensive research in the synthesis of new acyclonucleosides in which the carbohydrate moieties are acyclic chains mimicking the sugar portion of naturally occurring nucleosides. Some acyclonucleosides containing chiral carbons in the acyclic side chain, such as S-HPMPC (Cidofovir),[2] S-HPMPA,[3] R-PMPA (Tenofovir),[4] S-DHPA,[5] et al., have been shown to possess antiviral activities (Figure 1). The biological activity spectrum of the acyclic nucleoside is markedly influenced not only by the base moiety, but also by the structure and absolute configuration of the aliphatic side chain. For example, for DHPA and HPMPA, only their *S*-enantiomers exhibit antiviral activities whereas the *R*-enantiomers are markedly less active.[5a] Reversely, the *R*-enantiomers of PMPA and PMPDAP show 10−100-fold higher activity against human immunodeficiency virus than their *S*-counterparts.[6] It should also be mentioned that the antiviral activity found for a racemic mixture is not necessarily half of the activity of the more active enantiomer. The markedly different activities of respective enantiomers make it very important to synthesize optically pure compounds for antiviral evaluation.

The reported synthesis methods of acyclic nucleoside analogues involve alkylation of purine or pyrimidine bases with various alkylating agents. The most frequently used alkylating agents are halogenated compounds.[7] Some alkylating agents are mesylate[8] or tosylate.[9] Another straightforward method for the synthesis of acyclic nucleoside is Michael addition, which usually provides racemic or achiral acyclic nucleosides.[10] All these methods suffer serious disadvantages because they are multistep and laborious, and are targeted at the synthesis of the analogues of one type.

Encouraged by the properties of these chiral acyclic nucleosides and based on the previous work of our studies on nucleoside analogues,[11] herein, we report the synthesis of novel chiral acyclic nucleoside analogues through the Mitsunobu reaction between purine or pyrimidine bases and

(1) (a) Wagstaff, A. J.; Faulds, D.; Goa, K. L. *Drugs* **1994**, *47*, 153. (b) Darby, G. *J. Med. Virol.* **1993**, *1* (Suppl.), 134. (c) Welsby, P. D. *J. Infect.* **1994**, *28*, 121. (d) Ruiter, A. D.; Thin, R. N. *Drugs* **1994**, *47*, 297. (e) Sally, F.; John, M. G. *Mol. Biotechnol.* **1996**, *5*, 125.

(2) (a) De Clercq, E. *Collect. Czech. Chem. Commun.* **1998**, *63*, 480. (b) Safrin, S. J.; Jaffe, H. S. *Adv. Exp. Med. Biol.* **1999**, *458*, 111.

(3) (a) De Clercq, E.; Holý, A.; Rosenberg, I.; Sakuma, T.; Balzarini, J.; Maudgal, P. C. *Science* (*Washington, D.C.*) **1986**, *323*, 464. (b) Webb, R. R.; Martin, J. C. *Tetrahedron Lett.* **1987**, *28*, 4953.

(4) Doláková, P.; Dračínský, M.; Masojídková, M.; Šolínová, V.; Kašička, V.; Holý, A. *Eur. J. Med. Chem.* **2009**, *44*, 2408.

(5) (a) Ullas, G. V.; Chu, C. K.; Ahn, M. K.; Kosugi, Y. *J. Org .Chem.* **1988**, *53*, 2413. (b) De Clercq, E.; Descamps, J.; De Somer, P.; Holý, A. *Science* (*Washington, D.C.*) **1978**, *200*, 563.

(6) (a) Balzarini, J.; Aquaro, S.; Perno, C. F.; Witvrouw, M.; Holý, A.; Clercq, E. D. *Biochem. Biophys. Res. Commun.* **1996**, *219*, 337. (b) Šolínová, V.; Kašička, V.; Sázelová, P.; Holý, A. *Electrophoresis* **2009**, *30*, 2245.

(7) (a) Votruba, A. I. *J. Med. Chem.* **2002**, *45*, 1918. (b) Ullas, G. V.; Chu, K. C.; Ahn, M. K.; Kosugi, Y. *J. Org. Chem.* **1988**, *53*, 2413. (c) Diederichsen, U.; Weicherding, D.; Diezemanna, N. *Org. Biomol. Chem.* **2005**, *3*, 1058. (d) Harnden, M. R.; Jarvest, R. L.; Bacon, T. H.; Boyd, M. R. *J. Med. Chem.* **1987**, *30*, 1636. (e) Sheikha, G. A.; Colla, P. L.; Loi, A. G. *Nucleosides, Nucleotides Nucleic Acids* **2002**, *21*, 619. (f) Schaeffer, H. J.; Beauchamp, L.; De Miranda, P.; Elion, G. B. *Nature* **1978**, *272*, 583. (g) Rana, V. S.; Kumarp, V. A.; Ganesh, K. N. *Tetrahedron* **2001**, *57*, 1311. (h) Rana, V. S.; Kumar, V. A.; Ganesh, K. N. *Bioorg. Med. Chem. Lett.* **1997**, *7*, 2837. (i) Cheng, C.; Shimo, T.; Somekawa, K.; Baba, M. *Tetrahedron* **1998**, *54*, 2031.

(8) (a) Holý, A.; Dvořáková, H.; Masojídková, M.; Jindřich, J. *J. Med. Chem.* **1996**, *39*, 4073. (b) Zakirova, N. F.; Shipitsyn, A. V.; Belanovb, E. F.; Jasko, M. V. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 3357.

(9) (a) Vandendriessche, F.; Snoeck, R.; Janssen, G. *J. Med. Chem.* **1992**, *35*, 1458. (b) Franchetti, P.; Sheikha, G. A.; Cappellacci, L. *J. Med. Chem.* **1995**, *38*, 4007. (c) Vrbková, S.; Dranský, M.; Holý, A. *Tetrahedron: Asymmetry* **2007**, *18*, 2233.

(10) (a) Scheiner, P.; Geer, A.; Bucknor, A. M.; Imbach, J. L.; Schinazii, R. F. *J. Med. Chem.* **1989**, *32*, 73. (b) Lira, E. P.; Huffman, C. W. *J. Org. Chem.* **1966**, *31*, 2188. (c) Guillarme, S.; Legoupy, S.; Aubertin, A. M.; Olicard, C. *Tetrahedron* **2003**, *59*, 2177. (d) Qu, G. R.; Zhang, Z. G.; Geng, M. W.; Xia, R.; Zhao, L.; Guo, H. M. *Synlett* **2007**, *5*, 721. (e) He, L.; Liu, Y. M.; Zhang, W.; Li, M.; Chen, Q. H. *Tetrahedron* **2005**, *65*, 8505.

(11) (a) Qu, G. R.; Mao, Z. J.; Niu, H. Y.; Wang, D. C.; Xia, C; Guo, H. M. *Org. Lett.* **2009**, *11*, 1745. (b) Qu, G. R.; Xia, R.; Yang, X. N.; Li, J. G.; Wang, D. C.; Guo, H. M. *J. Org. Chem.* **2008**, *73*, 2416. (c) Qu, G. R.; Ren, B.; Niu, H. Y.; Mao, Z. J.; Guo, H. M. *J. Org. Chem.* **2008**, *73*, 2450. (d) Qu, G. R.; Zhao, L.; Wang, D. C.; Wu, J.; Guo, H. M. *Green Chem.* **2008**, *10*, 287. (e) Qu, G. R.; Wu, J.; Wu, Y. Y.; Zhang, F.; Guo, H. M. *Green Chem* **2009**, *11*, 760.

DOI: 10.1021/jo100397a Published on Web 04/30/2010
© 2010 American Chemical Society

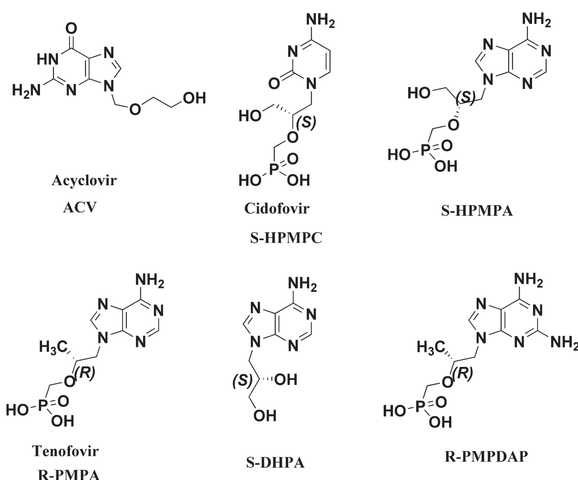
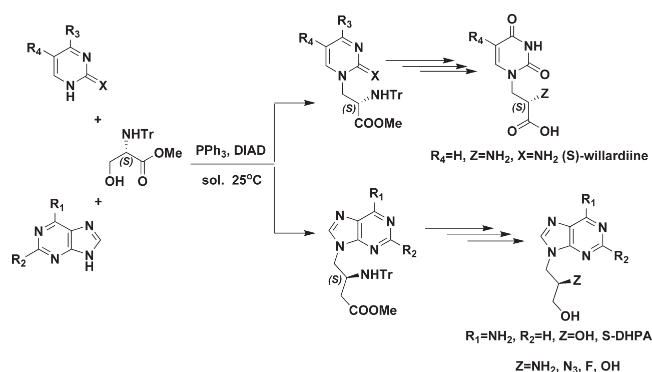


FIGURE 1. Structures of some acyclonucleosides possessing antiviral activities.

SCHEME 1. Preparation of Both Chiral Acyclic Nucleosides and Nucleotides



N-protected L-serine methyl ester. These novel chiral acyclic nucleoside ananogues can be used as key intermediates for the synthesis not only of naturally occurring *S*-willardiine and its analogues, but also of S-DHPA-like nucleoside analogues. The proposed uniform scheme allows the preparation of both chiral acyclic nucleosides and nucleotides (Scheme 1).

Initially, we investigated the Mitsunobu reaction between 6-chloropurine and *N*-*tert*-butoxycarbonyl (Boc) L-serine methyl ester. To our disappointment, the primary product was not the expected coupling product but the elimination product of *N*-*tert*-butoxycarbonyl (Boc) L-serine methyl ester. A survey of the literature[12] demonstrated that when Cbz or Boc was used as the N-protecting group for serine, the elimination product was in 73% or 91% yield, respectively, along with a small amount of the nucleophilic substitution product. Consistent with the literature, we decided on trityl (Tr) as the N-protecting group. Gratifyingly the elimination product was avoided, but the yield was still quite low.

To obtain satisfactory yield, various factors were investigated to optimize the reaction condition. First, the addition order of the reactants and solvents was studied. The typical procedure of the Mitsunobu reaction was as follows: diisopropyl azodicarboxylate (DIAD) was added dropwise to the mixture of triarylphosphine, alcohol, and the nucleophile. Disappointingly, the reaction was poor according to the above typical adding order. A survey of the adding order showed that the yield can be improved significantly according to the following order: alcohol was first dissolved in the solvent, followed by the addition of nucleophile and triarylphosphine sequentially, then DIAD was added dropwise last.

Next, the effects of solvent, temperature, and concentration were examined (Table 1). As shown in Table 1, xylene was shown to be the best choice for the Mitsunobu reaction (entries 1−9). In xylene, the reaction proceeded smoothly and gave the desired product in excellent yield (entry 6). Temperature also made a difference on the reaction. At 0 °C, the yield was moderate (entry 10); when the temperature was increased to 25 °C, the yield was excellent and the reaction time was shortened to 1 h (entry 6). However, when the temperature was higher than 25 °C (entries 11 and 12), the yields decreased obviously. Therefore, 25 °C is the optimized temperature. The results of the survey of reaction concentration (entries 6 and 13−16) indicated that the reaction proceeded well in dilute environment and 0.1 mol/L turned out to be the best choice.

To evaluate the generality of the reaction, a number of purines with various substituents were subjected to the optimized reaction conditions. However, the result came as a surprise. Some of the substrates gave low yields; for most of them, no reaction was observed. To extend the application of the reaction, we decided to try different solvents. To our delight, each substrate could conduct well in at least one of the investigated solvents (for details see the Supporting Information). For most substrates, DCM proved to be the best solvent. The best solvent for each substrate was listed in Table 2.

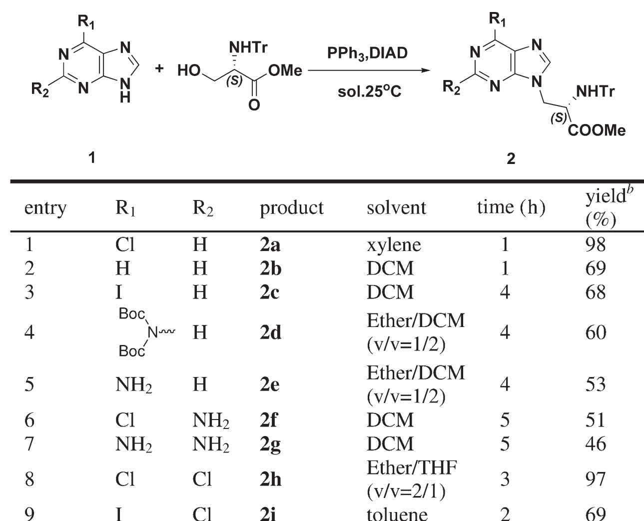
TABLE 1. Optimization of Reaction Conditions[a]

entry	solvent	temp (°C)	concn (mol/L)	yield[b] (%)
1	DCM	25	0.1	60
2	DCE	25	0.1	53
3	ether	25	0.1	64
4	THF	25	0.1	NR
5	toluene	25	0.1	64
6	xylene	25	0.1	98
7	dioxane	25	0.1	35
8	ether/DCM (v/v = 1/2)	25	0.1	24
9	ether/THF (v/v = 2/1)	25	0.1	NR
10	xylene	0	0.1	63
11	xylene	60	0.1	82
12	xylene	80	0.1	74
13	xylene	25	0.025	90
14	xylene	25	0.05	96
15	xylene	25	0.2	25
16	xylene	25	0.4	32

[a]Reaction conditions: reaction time, 1.5 h; alcohol (0.3 mmol) was dissolved in the solvent, then 6-chloropurine (0.45 mmol) and PPh_3 (0.48 mmol) were added sequentially, finally DIAD (0.48 mmol) was added dropwise. [b]Isolated yields based on alcohol.

(12) Cherney, R. J.; Wang., L. *J. Org. Chem.* **1996**, *61*, 2544.

TABLE 2. Reaction of Purines with Various Substituents[a]



entry	R_1	R_2	product	solvent	time (h)	yield[b] (%)
1	Cl	H	**2a**	xylene	1	98
2	H	H	**2b**	DCM	1	69
3	I	H	**2c**	DCM	4	68
4	$(Boc)_2N$–	H	**2d**	Ether/DCM (v/v=1/2)	4	60
5	NH_2	H	**2e**	Ether/DCM (v/v=1/2)	4	53
6	Cl	NH_2	**2f**	DCM	5	51
7	NH_2	NH_2	**2g**	DCM	5	46
8	Cl	Cl	**2h**	Ether/THF (v/v=2/1)	3	97
9	I	Cl	**2i**	toluene	2	69

[a]Reaction conditions: alcohol (0.3 mmol) was dissolved in solvent, then purine (0.45 mmol) and PPh_3 (0.48 mmol) were added sequentially, finally DIAD (0.48 mmol) was added dropwise. [b]Isolated yields based on alcohol.

The effect of substituents on purine was investigated, too. When there was no substituent at the C2 position, all the purine derivatives gave the desired products **2a**−**e** in moderate to good yields (entries 1−5). Replacement of H by NH_2 (entry 6) led to a lower yield after purification; by contrast, replacement of H by Cl (entry 8) led to higher yields. This indicated that the electron-donating effects on C2 could lead to a decrease of the yields whereas the electron-withdrawing effects on C2 could lead to an increase of the yields. The influence of different substituent groups at C6 was also studied. When there was no substituent at the C6 position (entry 2), the yield was moderate. Replacement of H by Cl (entry 1) led to a higher yield after purification whereas replacement of H by NH_2 (entry 8) led to a lower yield. The results indicated that the yield was in inverse proportion to the electron density at C6, which was in agreement with the effect on C2.

When this procedure was applied to pyrimidine derivatives, to our delight, the target molecules were obtained in moderate to good isolated yields (Table 3)

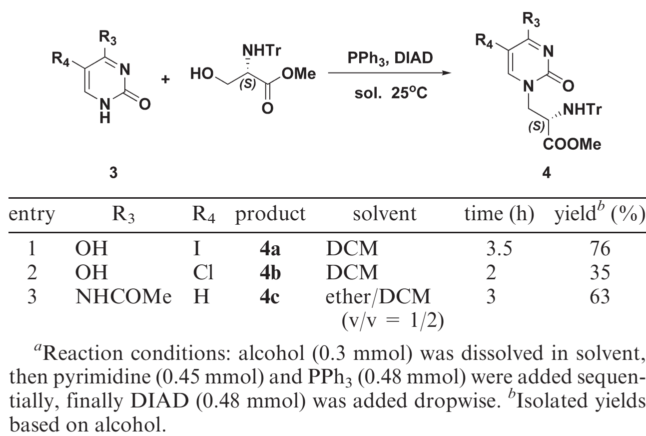
As indicated in Table 3, the reaction could proceed smoothly with pyrimidine derivatives, and the yield was moderate, except for 5-chlorouracil, which might be affected by the chlorine atom.

It is noteworthy that the purine target products would serve in further functionalizations to produce a molecule that is similar to S-DHPA[5] (Figure 1), a medicinal which is effective against a variety of RNA viruses. Herein we just try **2b** to do the further transformation (Scheme 2).

Besides, pyrimidine target products would also serve in further functionalizations to produce [(*S*)-1-(2-amino-2-carboxyethyl)-pyrimidine-2,4-dione] (*S*-Willardiine) derivatives (Figure 1). *S*-Willardiine1 is a naturally occurring heterocyclic excitatory amino acid present in the seeds of Acacia and Mimosa. The (*S*)- but not (*R*)-isomers of willardiine and 5-bromowillardiine were potent agonists, producing rapidly but incompletely desensitizing responses. Herein we just try **4b** to do the further transformation (Scheme 3).

In conclusion, we have developed a novel and efficient synthetic method to prepare chiral acyclic nucleosides with a

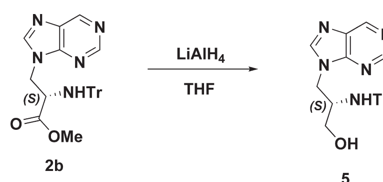
TABLE 3. Reaction of Pyrimidines with Various Substituents[a]



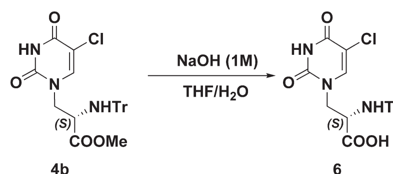
entry	R_3	R_4	product	solvent	time (h)	yield[b] (%)
1	OH	I	**4a**	DCM	3.5	76
2	OH	Cl	**4b**	DCM	2	35
3	NHCOMe	H	**4c**	ether/DCM (v/v = 1/2)	3	63

[a]Reaction conditions: alcohol (0.3 mmol) was dissolved in solvent, then pyrimidine (0.45 mmol) and PPh_3 (0.48 mmol) were added sequentially, finally DIAD (0.48 mmol) was added dropwise. [b]Isolated yields based on alcohol.

SCHEME 2. Reduction of 2b



SCHEME 3. Hydrolysis of 4b



chiral amino side chain by the Mitsunobu reaction between L-serine and a base. Compared to previously known approaches, the simplicity of this procedure and generally satisfactory yields make this method particularly attractive. Since the target molecules could be converted to many other useful compounds, this method suggests an opportunity to acquire many other useful derivatives from these chiral acyclic nucleosides.

Experimental Section

Typical Experimental Procedure for the Mitsunobu Reaction of Purines or Pyrimidines with *N*-Trityl L-Serine Methyl Ester. *N*-Trityl L-serine methyl ester (0.3 mmol) was dissolved in the solvent, then purine base **1** or pyrimidine **3** (0.45 mmol) and PPh_3 (0.48 mmol) were added sequentially, then DIAD (0.48 mmol) was added dropwise. After completion of the reaction monitored by thin layer chromatography (TLC), the solvent was evaporated, then the crude product was purified by column chromatography over silica gel with ethyl acetate and petroleum ether as the eluent, to give target product **2** or **4**.

Compound 2a: white powder, mp 90−91 °C; ^1H NMR ($CDCl_3$, 400 MHz) δ 8.70 (s, 1H), 8.31 (s, 1H), 7.34−7.32 (m, 6H), 7.23−7.15 (m, 9H), 4.55 (dd, J = 5.6, 14.0 Hz. 1H), 4.35 (dd, J = 4.8, 14.0 Hz, 1H), 3.88−3.82 (m, 1H), 3.22 (s, 3H), 2.89 (s, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 171.5, 151.2, 151.1, 150.2, 145.1, 144.0, 130.4, 127.5, 127.2, 126.0, 70.2, 55.0, 51.6, 47.0; HRMS calcd for $C_{28}H_{24}ClN_5NaO_2$ [M + Na^+] 520.1516, found 520.1519.

Compound 4a: white powder, mp 76−80 °C; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.34 (s, 1H), 7.85 (s, 1H), 7.42−7.40 (m, 6H), 7.31−7.22 (m, 9H), 4.14 (d, J = 8.8 Hz, 1H), 3.70−3.64 (m, 2H), 3.29 (s, 3H), 2.89 (d, J = 9.2 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 171.7, 159.3, 149.3, 149.2, 144.1, 127.6, 127.3, 126.0, 70.3, 65.8, 54.2, 51.7, 51.0, 21.0; HRMS calcd for $C_{27}H_{24}IN_3NaO_4$ [M + Na$^+$] 604.0709, found 604.0710.

Typical Experimental Procedure for the Reduction of Methyl 3-(9*H*-Purin-9-yl)-2-(tritylamino)propanoate. $LiAlH_4$ was suspended in THF and the mixture was stirred at room temperature for 25 min. To this solution was added **2b** and the mixture was stirred for 4 h. After completion of the reaction, the solvent was evaporated, then the crude product was purified by column chromatography over silica gel with dichloromethane and methanol as the eluent, to give target product **5**.

Compound 5: white powder, mp 190−193 °C; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 9.16 (s, 1H), 8.92 (s, 1H), 7.94 (s, 1H), 7.56−7.54 (m, 6H), 7.34−7.30 (m, 6H), 7.27−7.22 (m, 3H), 4.14 (dd, J = 3.2, 14.0 Hz, 1H), 3.73 (dd, J = 6.0, 14.0 Hz, 1H), 3.16−3.14 (m, 1H), 2.98 (dd, J = 4.0, 12.0 Hz, 1H), 2.66 (dd, J = 7.2, 12.0 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 151.3, 150.9, 148.1, 145.8, 145.2, 133.0, 127.6, 127.3, 126.0, 70.3, 60.2, 53.0, 43.6; HRMS calcd for $C_{27}H_{25}N_5NaO$ [M + Na$^+$] 458.1957, found 458.1957.

Typical Experimental Procedure for the Hydrolysis of Methyl 3-(5-Chloro-2,4-dioxo-3,4-dihydropyrimidin-1(2*H*)-yl)-2-(tritylamino)-propanoate. The aqueous solution of NaOH (1 M, 1 mL) was placed in a cylindrical vessel with a cross section of 3 cm^2 and a capacity of 12 mL. Then the THF solution of ester **4b** (1 mL) was added dropwise. The vessel was placed in a thermostated bath (26 °C) and stirred for 20 h. The reaction mixture was extracted with CH_2Cl_2, the aqueous layer was acidified by 2 M HCl to pH 3 and filtered, then the residue was dried in an oven to afford the target molecular **6**.

Compound 6: white powder, mp 118−120 °C; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 10.50 (s, 1H), 7.51 (s, 1H), 7.50 (m, 6H), 7.23 (m, 10H), 4.13 (d, J = 6.4 Hz, 1H), 3.65 (s, 1H), 3.35 (d, J = 9.2 Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 146.4, 144.6, 143.2, 127.6, 127.0, 126.7, 126.4, 125.7, 125.5, 100.5, 70.0, 51.0.

Acknowledgment. We are grateful for financial support from the National Nature Science Foundation of China (grants 20772024 and 20802016), the Program for New Century Excellent Talents in University of Ministry of Education (NCET-09-0122), the Program for Innovative Research Team in University of Henan Province (2008IRTSTHN002), and the Henan National Nature Science Foundation (092300410226).

Supporting Information Available: General information, experimental procedures, optimization of reaction conditions for each purine base or pyrimidine, and characterization data for the products including spectroscopic information. This material is available free of charge via the Internet at http://pubs.acs.org.