An Efficient Synthesis of Cyclic IDP- and Cyclic 8-Bromo-IDP-Carbocyclic-Riboses Using a Modified Hata Condensation Method To Form an Intramolecular Pyrophosphate Linkage as a Key Step. An Entry to a General Method for the Chemical Synthesis of Cyclic ADP-Ribose Analogues[1]

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An efficient synthesis of cyclic IDP-carbocyclic-ribose (**3**) and its 8-bromo derivative **6**, as stable mimics of cyclic ADP-ribose, was achieved, and a condensation reaction with phenylthiophosphate-type substrate **15** or **16** to form an intramolecular pyrophosphate linkage was a key step. *N*-1-Carbocyclic-ribosylinosine derivative **28** and the corresponding 8-bromo congener **24** were prepared via condensation between *N*-1-(2,4-dinitrophenyl)inosine derivative **17** and a known optically active carbocyclic amine **18**. Compounds **24** and **28** were then converted to the corresponding 5″-phosphoryl-5′-phenylthiophosphate derivatives **15** and **16**, respectively, which were substrates for the condensation reaction to form an intramolecular pyrophosphate linkage. Treatment of 8-bromo substrate **15** with I_2 or $AgNO_3$ in the presence of molecular sieves 3A (MS 3A) in pyridine at room temperature gave the desired cyclic product **12** quantitatively, while the yield was quite low without MS. The similar reaction of 8-unsubstituted substrate **16** gave the corresponding cyclized product **32** in 81% yield. Acidic treatment of these cyclic pyrophosphates **12** and **32** readily gave the targets **6** and **3**, respectively. This result suggests that the construction of *N*-1-substituted hypoxanthine nucleoside structures from *N*-1-(2,4-dinitrophenyl)inosine derivatives and the intramolecular condensation by activation of the phenylthiophosphate group with I_2 or $AgNO_3$/MS 3A combine to provide a very efficient route for the synthesis of analogues of cyclic ADP-ribose such as **3** and **6**. Thus, this may be an entry to a general method for synthesizing biologically important cyclic nucleotides of this type.

Introduction

Cyclic ADP-ribose (cADPR, **1**; Figure 1)[2] is a newly discovered general mediator involved in Ca^{2+} signaling.[3] The synthesis of cADPR analogues has been extensively studied by enzymatic and chemoenzymatic methods using ADP-ribosylcyclase, due to their biological importance.[4] ADP-ribosylcyclase from *Aplysia californa* mediates the intramolecular ribosylation of NAD^+ and some modified NAD^+, which are prepared chemically or enzymatically, at the *N*-1-position of the purine moiety to yield cADPR or the corresponding analogues.[4] However, the analogues that can be obtained by this method are limited due to the substrate specificity of the enzyme. Furthermore, even though ADP-ribosylcyclase catalyzes the cyclization of NAD^+ analogues, in some cases the newly formed glycosidic bond is attached to the *N*-7 nitrogen of the purine ring: e.g., the product of the enzymatic reaction of an inosine or guanosine analogue of NAD^+ is not the desired *N*-1-cyclized product, but rather the *N*-7-cyclized product.[4g] Accordingly, the development of flexible methods for synthesizing cADPR and a variety of its analogues is needed.

In cells, cADPR is synthesized from NAD^+ by ADP-ribosylcyclase and acts as a second messenger; it is hydrolyzed promptly by cADPR hydrolase to give ADP-ribose and inactivated under physiological conditions.[3] cADPR is also known to be readily hydrolyzed nonenzymatically at the unstable *N*-1-glycosidic linkage of its

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(1) This paper constitutes part 195 of Nucleosides and Nucleotides. Part 194: Abe, H.; Shuto, S.; Matsuda, A. *Tetrahedron Lett.* **2000**, *41*, 2391−2394.

(2) Clapper, D. L.; Walseth, T. F.; Dargie, P. J.; Lee, H. C. *J. Biol. Chem.* **1987**, *262*, 9561−9568.

(3) (a) Galione, A. *Science* **1993**, *259*, 325−326. (b) Lee, H. C.; Galione A.; Walseth, T. F. *Vitam. Horm. (San Diego)* **1994**, *48*, 199−257. (c) Dousa, T. P.; Chini, E. N.; Beers, K. W. *Am. J. Physiol.* **1996**, *271*, C1007−C1024. (d) Lee H. C. *Physiol. Rev.* **1997**, *77*, 1133−1164. (e) Lee, H. C. *Cell Biochem. Biophys.* **1998**, *28*, 1−17. (f) Galione, A.; Cui, Y.; Empson, R.; Iino, S.; Wilson, H.; Terrar, D. *Cell Biochem. Biophys.* **1998**, *28*, 19−30. (g) Guse, A. H. *Cell Signalling* **1999**, *11*, 309−316.

(4) (a) Walseth, T. F.; Lee, H. C. *Biochim. Biophys. Acta* **1993**, *1178*, 235−242. (b) Lee, H. C.; Aarhus, R.; Walseth, T. F. *Science* **1993**, *261*, 352−355. (c) Graeff, R. M.; Walseth, T. J.; Fryxell, K.; Branton, W. D.; Lee, H. C. *J. Biol. Chem.* **1994**, *269*, 30260−30267. (d) Zhang, F.-J.; Sih, C. J. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 1701−1706. (e) Zhang, F.-J.; Gu, Q.-M.; Jing, P. C.; Sih, C. J. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 2267−2272. (f) Zhang, F.-J.; Yamada, S.; Gu, Q.-M.; Sih, C. J. *Bioorg. Med. Chem. Lett.* **1996**, *6*, 1203−1208. (g) Zhang, F.-J.; Sih, C. J. *Tetrahedron Lett.* **1995**, *63*, 9289−9292. (h) Zhang, F.-J.; Sih, C. J. *Bioorg. Med. Chem. Lett.* **1996**, *6*, 2311−2316. (i) Ashamu, G. A.; Galione, A.; Potter, B. V. L. *J. Chem. Soc., Chem. Commun.* **1995**, 1359−1356. (j) Bailey, B. C.; Fortt, S. M.; Summerhill, R. J.; Galione, A.; Potter, B. V. L. *FEBS Lett.* **1996**, *379*, 227−230. (k) Bailey, V. C.; Sethi, J. K.; Fortt, S. M.; Galione, A.; Potter, B. V. L. *Chem. Biol.* **1997**, *4*, 51−60. (l) Bailey, V. C.; Sethi, J. K.; Galione, A.; Potter, B. V. L. *J. Chem. Soc., Chem. Commun.* **1997**, 695−696. (m) Sethi, J. K.; Empson, R. M.; Bailey, V. C.; Potter, B. V. L. Galione, A. *J. Biol. Chem.* **1997**, *272*, 16358−16363. (n) Ashamu, G. A.; Sethi, J. K.; Galione, A.; Potter, B. V. L. *Biochemistry* **1997**, *36*, 9509−9517. (o) Zhang, F.-J.; Gu, Q.-M.; Sih, C. J. *Bioorg. Med. Chem.* **1999**, *7*, 653−664. (p) Wong, L.; Aarhus, R.; Lee, H. C.; Walseth, T. F. *Biochim. Biophys. Acta* **1999**, *1472*, 555−564.

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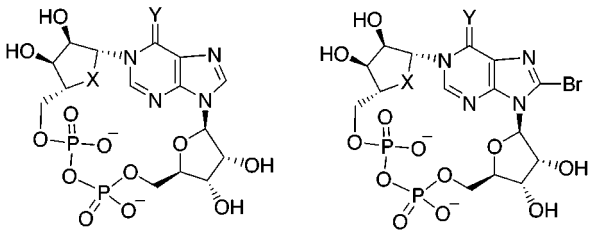


1 (cADPR): X = O, Y = NH
2: X = CH_2, Y = NH
3: X = CH_2, Y = O

4: X = O, Y = NH
5: X = CH_2, Y = NH
6: X = CH_2, Y = O

Figure 1.

adenine moiety to give ADP-ribose, even in neutral aqueous solution.[5] Although further intensive studies of cADPR are needed because of its biological importance, this biological as well as chemical instability of cADPR limits studies of its physiological role, at least to some extent. Therefore, stable analogues of cADPR that exhibit a Ca^{2+}-mobilizing activity in cells similar to that of cADPR are urgently required.

We designed cyclic ADP-carbocyclic-ribose (**2**) and its inosine congener **3** (cIDP-carbocyclic-ribose)[6] as stable mimics of cADPR, in which an oxygen atom in the ribose ring of cADPR is replaced by a methylene group. The mimics **2** and **3** should be resistant to both enzymatic and chemical hydrolysis, since they lack the unstable *N*-1-glycosidic linkage of cADPR. These analogues preserve all of the functional groups of cADPR, except for this ring oxygen, and should have a conformation similar to that of cADPR. Therefore, we expect that these analogues would effectively mobilize intracellular Ca^{2+}, like cADPR, so that they could be used as pharmacological tools for studying the mechanism of cADPR-modulated Ca^{2+} signaling pathways. The 8-bromo derivatives of cADP- and cIDP-carbocyclic-riboses (**5** and **6**, respectively) were also our synthetic targets, since Lee and coworkers found that cyclic 8-bromo-ADPR (**4**) is an antagonist of cADPR.[4a] Therefore, **5** and/or **6** may be a biologically and chemically stable antagonist of cADPR, which would also be very useful in biological studies.

We previously achieved the synthesis of the inosine congener **3**,[6] which is the first total synthesis of a cADPR analogue.[7] However, the overall yield was very low, and its biological activity has not been evaluated. In this synthesis, the intramolecular condensation to form the pyrophosphate linkage was a key step, but was very difficult. The difficulty of forming such an intramolecular pyrophosphate linkage, which prevented the completion of the synthesis of target cADPR analogues, has also been experienced by other groups.[4o,7,8] Therefore, the development of an efficient method for forming the intramolecular pyrophosphate linkage should be very beneficial.

In this paper, we describe an efficient method for preparing **3** and **6** using a modified Hata condensation reaction[9] with phenylthiophosphate-type substrates to form the intramolecular pyrophosphate linkage as a key step, and this may be an entry to a general synthetic method for cADPR-related compounds.[10]

Results and Discussion

Problems with Our Previous Synthesis of cIDP-Carbocyclic-Ribose. Our previous synthetic route is summarized in Scheme 1. An S_N2 reaction between the protected 8-bromoinosine derivative **7** and carbocyclic unit **8** provided the 8-bromo-*N*-1-(carbocyclic-ribosyl)inosine derivative **9**, which was then converted to bisphosphate **11**. Intramolecular condensation between the two phosphate groups of bisphosphate **11** was achieved by treating it with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC) in *N*-methylpyrrolidone (NMP), and subsequent reductive debromination and deprotection gave **3**. In this synthesis, intramolecular condensation to form the pyrophosphate linkage was the key step, but was very difficult. The two phosphate moieties in the molecule are perhaps separated due to electrostatic repulsion, which would prevent the desired condensation reaction.[4o,7,8] We found that the key intramolecular condensation reaction between the two phosphate groups of **11** proceeded only when a bromo substituent was introduced at the 8-position of the hypoxanthine ring of the substrate.[6] It is likely that the molecule is conformationally restricted in a *syn*-form around its glycosidic linkage due to the 8-bromo substituent,[11] in which case the two phosphate moieties would be rather close to each other to facilitate the condensation, at least to some extent.[6] However, the yield of the condensation reaction was insufficient (23%), even when the 8-bromo substrate was used.

Other problems in the previous study were rather long reaction steps, including an enzymatic optical resolution to construct the optically active carbocyclic unit **8** from cyclopentadiene, and inadequate yields in the coupling between inosine unit **7** and carbocyclic unit **8** (44%).

Thus, the development of both an efficient condensation method for forming the intramolecular pyrophosphate linkage and a more straightforward method for constructing the *N*-1-carbocyclic structure was needed.

The Synthetic Plan in This Study. The present plan for the synthesis of **3** and **6** is shown in Scheme 2. Formation of the intramolecular pyrophosphate linkage was investigated by treating 5′-phenylthiophosphates **15** and **16** as substrates with $AgNO_3$ or I_2 as a promoter. The *N*-1-carbocyclic-ribosyl structure is constructed from *N*-1-(2,4-dinitrophenyl)inosine derivative **17** and optically active carbocyclic amine **18**. Compounds **17** and **18** are readily prepared from inosine and commercially available (1*R*)-(−)-azabicyclo[2.2.1]hept-5-en-3-one.[7b]

(5) Lee, H. C.; Aarhus, R. *Biochim. Biophys. Acta* **1993**, *1164*, 68−74.

(6) Shuto, S.; Shirato, M.; Sumita, Y.; Ueno, Y.; Matsuda, A. *J. Org. Chem.* **1998**, *52*, 1986−1994.

(7) Synthetic approaches to carbocyclic analogues of cADP-ribose have been published by other groups. Although the *N*-1-carbocyclic inosine and adenosine structures have been constructed, formation of the intramolecular pyrophosphate linkage has not been achieved. (a) Fortt, S.; Potter, B. V. L. *Tetrahedron Lett.* **1997**, *38*, 5371−5374. (b) Hutchinson, E. J.; Taylor, B. F.; Blackburn, G. M. *J. Chem. Soc., Chem. Commun.* **1997**, 1859−1860.

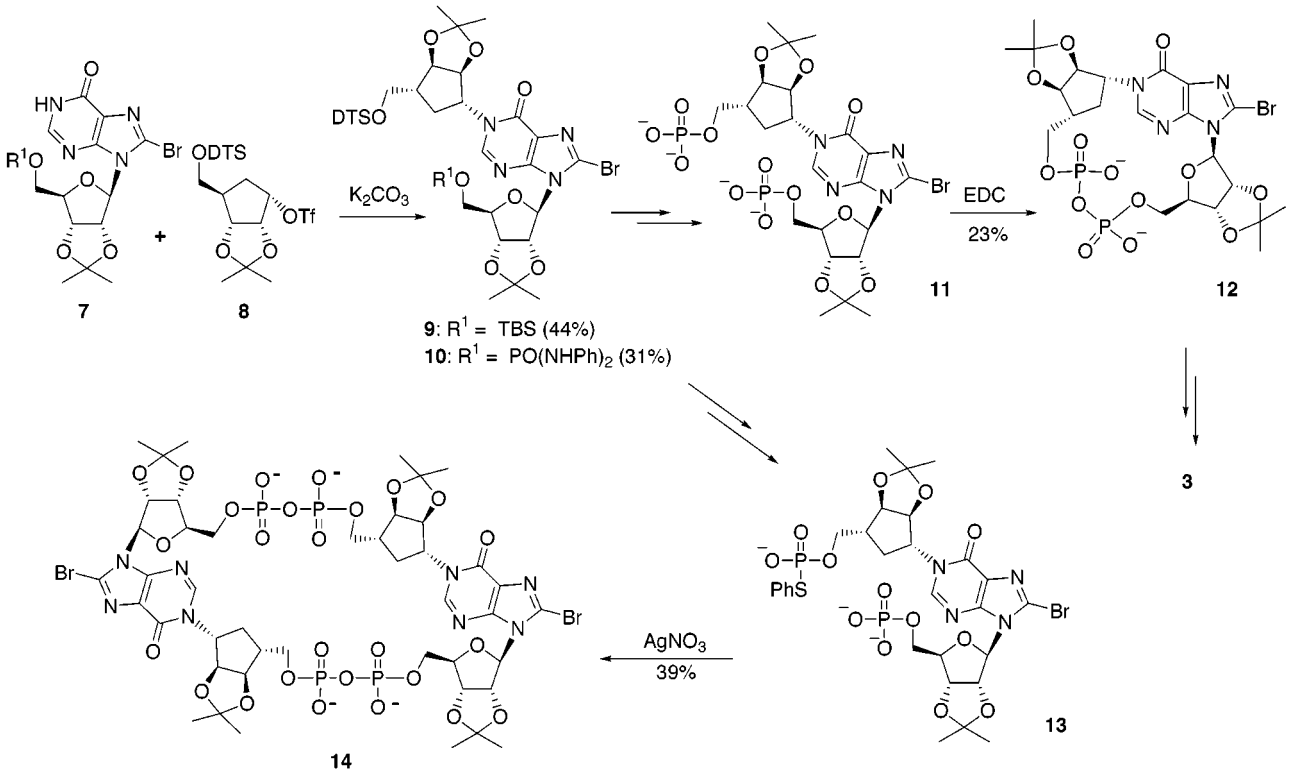
(8) Gu, Q.-M.; Sih, C. J. *J. Am. Chem. Soc.* **1994**, *116*, 7481−7486.

(9) (a) Nakagawa, I.; Kony, S.; Ohtani, S.; Hata, T. *Synthesis* **1980**, 556−557. (b) Sekine, M.; Kamimura, T.; Hata, T. *J. Chem. Soc., Perkin Trans. 1* **1985**, 997−1000. (c) Sekine, M.; Nishiyama, S.; Kamimura, T.; Osaki, Y.; Hata, T. *Bull. Chem. Soc. Jpn.* **1985**, *58*, 850−860. (d) Fukuoka, K.; Suda, F.; Suzuki, R.; Ishikawa, M.; Takaku, H.; Hata, T. *Nucleosides Nucleotides* **1994**, *13*, 1557−1567.

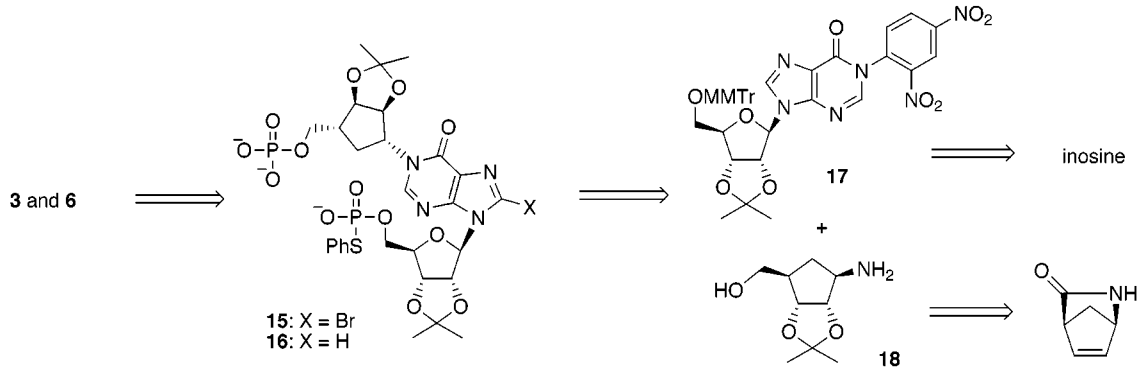
(10) A preliminary account of this study has been published previously: Fukuoka, M.; Shuto, S.; Minakawa, N.; Ueno, Y.; Matsuda, A. *Tetrahedron Lett.* **1999**, *40*, 5361−5364.

(11) Although a predominance of *anti*- over *syn*-conformers is well-known for natural nucleosides and their analogues, introducing a bulky substituent, such as a bromo group, into the 8-position of purine nucleosides restricts the conformation in a *syn*-form, through steric repulsion for the ribose moiety: Saenger, W. *Principles of Nucleic Acid Structure*; Springer-Verlag: New York, 1983.

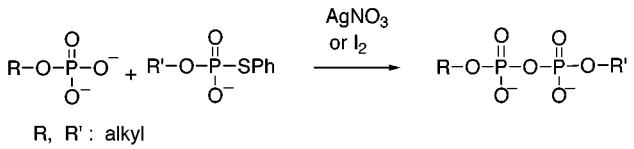
Scheme 1



Scheme 2



Scheme 3



Hata and co-workers found that the condensation between a phenylthiophosphate and a phosphomonoester was effectively promoted by I_2 or $AgNO_3$ to give the corresponding pyrophosphate compounds, as shown in Scheme 3.[9] They successfully synthesized the 5′-cap structure of mRNA using this method.[9d] We previously investigated Hata's method to form the intramolecular pyrophosphate linkage with phenylthiophosphate derivative **13** as a substrate in a synthetic study on cIDP-carbocyclic-ribose.[12] However, when phenylthiophosphate derivative **13**, derived from **10**, was treated with $AgNO_3$ in NMP/HMPA, the desired **12** was not obtained at all, while a cyclic dimer, **14**, was obtained as a major product (Scheme 1).[12] While it is unclear why such a cyclic dimer was produced, the reaction of **13** with $AgNO_3$ would proceed via intermediate **A**, in which the 5″-phosphate of the carbocyclic-ribose moiety was activated as a metaphosphate (Figure 2).[13] In this study, we designed phenylthiophosphate-type substrate **15** (Scheme 2), which is a regioisomer of **13**, as another substrate for the intramolecular condensation reaction. A metaphosphate intermediate, **B**, where the 5′-phosphate of the ribose moiety is activated, should be produced when **15** is treated by a promoter such as I_2 or $AgNO_3$. On the other hand, the condensation reaction of bisphosphate **11** with EDC might proceed via two kinds of intermediates, **A**′ (the 5″-phosphate is activated) and **B**′ (the 5′-phosphate is activated). It is possible that intermediates **A** and **B**, or **A**′ and **B**′, respectively, give different reaction products. The reaction of **15** with $AgNO_3$ or I_2 would make it

(12) Shuto, S.; Shirato, M.; Sumita, Y.; Ueno, Y.; Matsuda, A. *Tetrahedron Lett.* **1998**, *39*, 7341−7344.

(13) The generation of a highly active metaphosphate intermediate has been suggested when phenylthiophosphates are treated by a promoter such as Ag^+: see ref 9c.

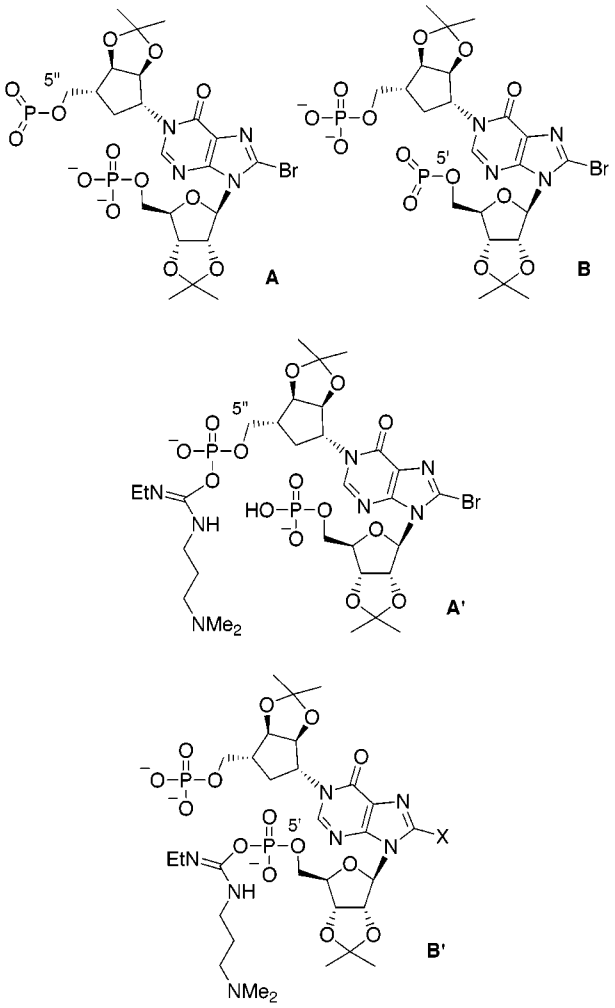


Figure 2.

clear whether intermediates **A** and **B** give different products, since intermediate **B** should only be produced when it is activated by the promoter. We also presumed that phenylthiophosphates **13** and **15** should be superior to bisphosphate **11** as a substrate for forming the intramolecular pyrophosphate linkage, since the electrostatic repulsion described above would be rather decreased in intermediates **A** and **B** due to their metaphosphate structure, compared with that in phosphodiester-type intermediates **A′** and **B′**, and the metaphosphate-type intermediates would be much more reactive than the phosphodiester-type intermediates.

We also planned to examine the intramolecular condensation reaction with 8-unsubstituted phenylthiophosphate-type substrate **16** (Scheme 2) to investigate whether the 8-bromo substitution facilitates intramolecular cyclization in the reaction system, which was observed previously when bisphosphate-type substrate **11** was condensed intramolecularly with EDC.[6]

On the other hand, Piccialli and co-workers recently reported an excellent method for preparing *N*-1-alkylinosines from *N*-1-(2,4-dinitrophenyl)inosine and alkylamines.[14] We planned to construct the *N*-1-carbocyclic-ribosyl structure from *N*-1-(2,4-dinitrophenyl)inosine derivative **17** and optically active carbocyclic amine **18** by this procedure.

Synthesis of the cIDP-Carbocyclic-Ribose and Its 8-Bromo Congener. The synthesis of **3** and **6** is summarized in Scheme 4. 2′,3′-*O*-Isopropylidene-5′-*O*-(monomethoxytrityl)inosine (**19**) was treated with 2,4-dinitrochlorobenzene and K_2CO_3 in DMF at 80 °C[14] to give *N*-1-(2,4-dinitrophenyl)inosine derivative **17** as a mixture of rotamers at the *N*-1-position[14] in 90% yield. Heating **17** with 10 equiv of **18**, which was prepared by Blackburn's method,[7b] at 50 °C in DMF gave the ring-cleaved product **20** as a mixture of stereoisomers due to a *cis*/*trans* geometry at the aminomethylene position in 74% yield.[15] When 1.5 equiv of **18** was used, the yield of **20** was decreased (42%). After the 5″-hydroxyl of **20** was protected with a TBS group, it was treated with *N*-bromoacetamide (NBA) in THF[16] to give 2-bromo derivative **22**. When **22** was heated in the presence of K_2CO_3 at 50 °C in DMF, the desired ring-closure product **23** was obtained in 98% yield. Similar treatment of **20** also gave 8-unsubstitued inosine derivative **28** in high yield. Thus, the *N*-1-carbocyclic inosine structure was efficiently constructed from *N*-1-(2,4-dinitrophenyl)inosine derivative **17** and a readily available optically active carbocyclic amine, **18**. This method is clearly superior to our previous method using S_N2-type condensation between an inosine derivative and an optically active carbocyclic unit.

The TBS group of **23** was removed with TBAF in THF, and a di(anilino)phosphoryl group was then introduced at the resulting 5″-primary hydroxyl by treating it with $(PhNH)_2POCl$[17] and tetrazole in pyridine[18] to give **25** in high yield. After the 5′-*O*-MMTr group of **25** was removed with aqueous AcOH, a bis(phenylthio)phosphoryl group was introduced at the primary hydroxyl of the ribose moiety with a cyclohexylammonium *S*,*S*-diphenylphosphorodithioate (PSS)/2,4,6-triisopropylbenzenesulfonyl chloride (TPSCl)/tetrazole/pyridine system[19] to give protected bisphosphate derivative **27**. Successive treatment of **27** with isoamyl nitrite in a mixed solvent of pyridine−AcOH−Ac_2O and H_3PO_2 in pyridine[20] gave **15**, the substrate for the intramolecular condensation reaction, in 88% yield as a triethylammonium salt. In a similar manner, the corresponding 8-unsubstituted substrate **16** was synthesized from **28**.

The intramolecular condensation reaction of **15** was investigated under various conditions, and the results are summarized in Table 1. HPLC charts of several reactions are also shown in Figure 3. Reactions were carried out by adding a solution of **15** slowly over 15 h, using a syringe pump, to a large excess of a promoter at room temperature, and monitored by HPLC. $AgNO_3$ or I_2 and *N*-methylpyrrolidone (NMP)/hexamethylphosphoramide (HMPA) or pyridine[9] were used as a promoter and a solvent, respectively. First, **15** was treated with $AgNO_3$ in NMP/HMPA to give the desired **12** in only 6% yield, along with cyclic dimer **14** and uncyclized hydrolysis

(14) De Napoli, L.; Messere, A.; Montesrchio, D.; Piccialli, G.; Varra, M. *J. Chem. Soc., Perkin Trans. 1* **1997**, 2079−2082: *N*-1-dinitrophenylinosine derivatives were obtained as a mixture of two rotamers at the *N*-1-position.

(15) The carbocyclic amine **18** was recovered in 87% yield after the reaction and was used repeatedly.

(16) Ivanovics, G. A.; Rousseau, R. J.; Kawana, M.; Srivastava, P. C.; Robins, R. K. *J. Org. Chem.* **1974**, *39*, 3651−3654.

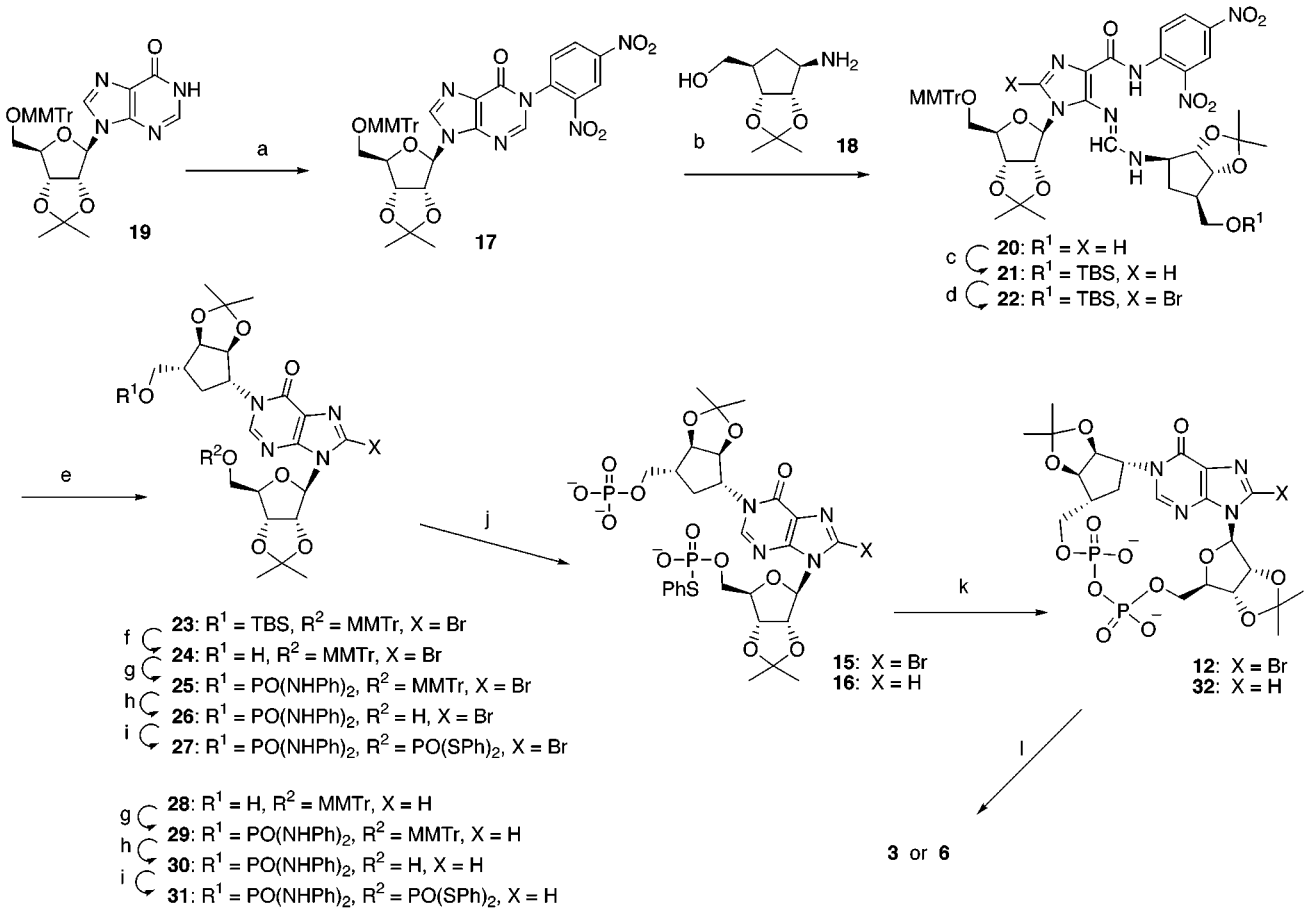
(17) Sasse, K. *Methoden der Organiches Chemie*; 1971; Vol. 12, No. 2, pp 444−450.

(18) Smrt, J. *Tetrahedron Lett.* **1973**, *47*, 4727−4728.

(19) Sekine, M.; Hamaoki, K.; Hata, T. *Bull. Chem. Soc. Jpn.* **1981**, *54*, 3815−3827.

(20) Hata, T.; Kamimura, T.; Urakami, K.; Kohno, K.; Sekine, M.; Kumagai, I.; Shinozaki, K.; Miura, K. *Chem. Lett.* **1987**, 117−120.

Scheme 4[a]



[a] Conditions: (a) 2,4-dinitrochlorobenzene, K_2CO_3, DMF, 80 °C; (b) **18**, DMF, 50 °C; (c) TBSCl, imidazole, rt; (d) NBA, THF, −10 °C; (e) K_2CO_3, DMF, 50 °C; (f) TBAF, THF; (g) $(PhNH)_2POCl$, tetrazole, py, rt; (h) aq 80% AcOH, rt; (i) PSS, tetrazole, TPSCl, py, rt; (j) (1) isoamyl nitrite, Ac_2O, AcOH, py, rt; (2) H_3PO_2, Et_3N, py, rt; (k) I_2, or $AgNO_3$, MS 3A, py; (l) 60% HCO_2H.

Table 1. Intramolecular Condensation Reactions of 15[a]

			products (yield, %)[b]		
entry	promoter	solvent	**12**	**14**	**11**
1	$AgNO_3$	NMP/HMPA	6	10	15
2	I_2	NMP/HMPA	6	5	29
3	I_2	pyridine	34	8	47
4	I_2/MS 3A	pyridine	100		
5	$AgNO_3$/MS 3A	pyridine	94		
6	I_2/MS 3A	NMP/HMPA	49	10	trace
7	$AgNO_3$/MS 3A	NMP/HMPA	42	6	trace

[a] To a mixture of $AgNO_3$ (30 equiv) or I_2 (20 equiv) [and MS 3A (500 mg, entries 4−6)] in NMP/HMPA (3:1, 8.0 mL) or pyridine (8.0 mL) was slowly added a solution of **15** (9.4 μmol) in the same solvent (8.0 mL) at room temperature over 15 h. [b] Entries 4 and 5, isolated yield; entries 1−3, 6, and 7, determined by HPLC.

product **11** in respective yields of 10% and 15% (entry 1). Compounds **12**, **14**, and **11** were identified by comparison with the authentic samples synthesized previously.[6,12] Treatment of **15** with I_2 instead of $AgNO_3$ gave a similar insufficient result (entry 2, Figure 3a). However, when pyridine was used as a solvent, the yield of **12** was clearly improved (34%), and hydrolysis product **11** was obtained in 47% yield (entry 3). Therefore, we performed the reaction in the presence of molecular sieves 3A (MS 3A) to remove water from the reaction system. Surprisingly, the reaction with I_2/MS 3A as a promoter gave the desired **12** as a sole product (Figure 3b), which was obtained quantitatively as a triethylammonium salt, after purification by C_{18} column chromatography (entry 4). A similar reaction with $AgNO_3$ instead of I_2 in the presence of MS 3A in pyridine also gave **12** in very high yield (entry 5). Use of NMP/HMPA as a solvent clearly decreased the yield of **12**, and the cyclic dimer **14** was formed (entries 6 and 7, Figure 3c).

We next examined the use of 8-unsubstituted substrate **16** in the intramolecular condensation reaction to investigate whether the 8-bromo substituent at the purine moiety facilitates intramolecular condensation to form a pyrophosphate linkage. Thus, **16** was treated under the same conditions as in entry 4 to give the desired cyclization product **32** in 81% isolated yield. An HPLC chart for the reaction is shown in Figure 3d. An increase of byproducts was observed by HPLC in the reaction of 8-unsubstitued **16** compared with that with 8-bromo substrate **15** (Figure 3b). These results suggest that the 8-bromo group facilitates the intramolecular condensation reaction to some extent, due to conformational restriction of the substrate in a *syn*-form around its glycosyl linkage.

The intramolecular condensation reaction of **13**, which is the regioisomeric substrate of **15**, was also examined. Compound **13** was readily prepared from **24** (Scheme 5). Treatment of **13** under the best conditions (Table 1, entry 4) gave the desired cyclized product **12** in 99% isolated yield. This clearly shows that both metaphosphate intermediates, i.e., 5″-phosphate-activated **A** and 5′-

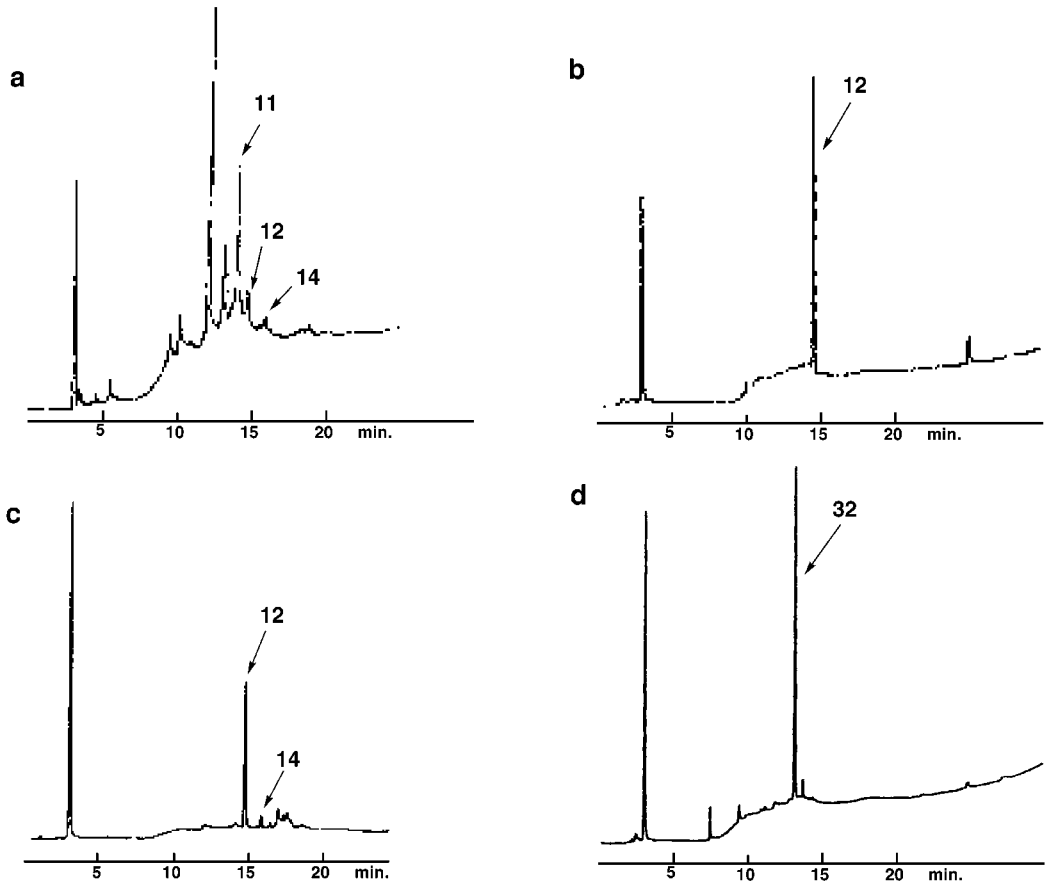
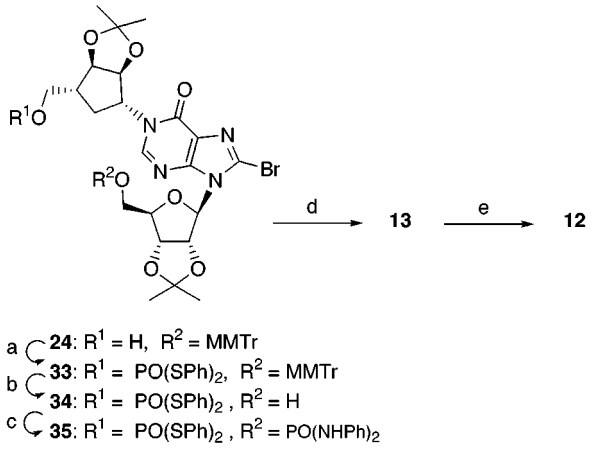


Figure 3. HPLC analysis at 254 nm of the intramolecular condensation reactions of **15** (a, b, and c) and **16** (d) at 15 h: (a) reaction with **15**, entry 2 in Table 1; (b) reaction with **15**, entry 4 in Table 1; (c) reaction with **15**, entry 6 in Table 1; (d) reaction with **16**, the same conditions as in entry 4 in Table 1.

Scheme 5[a]

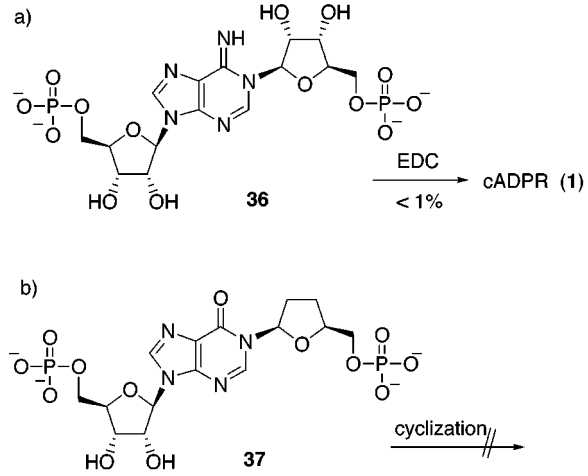


[a] Conditions: (a) PSS, tetrazole, TPSCl, py, rt; (b) aq 80% AcOH, rt; (c) $(PhNH)_2POCl$, tetrazole, py, rt; (d) (1) isoamyl nitrite, Ac_2O, AcOH, py, rt; (2) H_3PO_2, Et_3N, py, rt; (3) aq $NaHCO_3$; (e) I_2, MS 3A, py rt.

phosphate-activated **B**, readily cyclized to form **12** in high yield under these conditions.

It has been demonstrated that phenylthiophosphate-type substrates are very effective for a condensation reaction to form the intramolecular pyrophosphate linkage. This may be because the electrostatic repulsion between the two phosphate groups in the metaphosphate intermediates **A** and **B** is relatively decreased, as we expected. It may also be possible that the reactive

Scheme 6



intermediate is a pyridinium adduct which promotes the intramolecular cyclization via salt formation with the phosphate anion, since the use of pyridine as a solvent was key to the success of the intramolecular condensation. An attempt to prepare cADPR or its analogues by chemical intramolecular condensation was first reported by Gu and Sih.[8] They investigated condensation between the two phosphate groups of *N*-1-phosphoribosyl-AMP (**36**) with EDC, but were unsuccessful (yield <1%, Scheme 6a).[8] Later, ring closure of bisphosphate **37** through the formation of a pyrophosphate linkage was examined by Fortt and Potter, but they also failed.[7a] We

have also experienced the difficulty of conducting such an intramolecular condensation to prepare carbocyclic cADPR analogues.[6] Accordingly, our finding in this study with phenylthiophosphate-type substrates, i.e., that an I_2 or $AgNO_3$/MS 3A system very efficiently promotes the intramolecular condensation reaction between the phenylthiophosphate and phosphate groups to provide the desired cyclic pyrophosphate product in very high yield, is very important.

The cyclic pyrophosphates **32** and **12** were treated with aqueous HCO_2H at room temperature to give the targets cIDP-carbocyclic-ribose (**3**) and cyclic 8-bromo-IDP-carbocyclic-ribose (**6**), respectively.

Finally, we investigated whether the synthesized cIDP-carbocyclic-ribose (**3**) is stable in aqueous solution, as we hypothesized. When **3** was treated in 10 mM triethylammonium acetate buffer (pH 7.0) at 37 °C, none of its hydrolysis was observed by HPLC analysis after 3 days, whereas cADPR (**1**) was clearly hydrolyzed ($t_{1/2}$ = 60.5 h) under the same conditions.

Conclusion. We have developed a very efficient method for synthesizing cyclic IDP-carbocyclic-ribose and its 8-bromo derivative. The *N*-1-carbocyclic inosine structure was efficiently constructed from *N*-1-(2,4-dinitrophenyl)inosine derivative **17** and a readily available optically active carbocyclic amine, **18**. The key intramolecular cyclization reaction of the phenylthiophosphate-type substrates **13**, **15**, and **16** proceeded with I_2 or $AgNO_3$/MS 3A to give the products in very high yields. These results suggest that the construction of *N*-1-substituted inosine structures from *N*-1-(2,4-dinitrophenyl)inosine derivatives and the intramolecular condensation through the activation of a phenylthiophosphate group with I_2 or $AgNO_3$/MS 3A combined to provide a very efficient route for synthesizing analogues of cyclic ADP-ribose, such as **3** and **6**. This method may be applicable to the synthesis of cADPR analogues with an adenine base, including **2**. Thus, this may be an entry to a general method for synthesizing biologically important cyclic nucleotides of this type.

Experimental Section

Chemical shifts are reported in parts per million downfield from TMS (^{1}H and ^{13}C) or H_3PO_4 (^{31}P), and *J* values are given in hertz. The ^{1}H NMR assignments described are in agreement with COSY spectra. Thin-layer chromatography was done on Merck coated plate $60F_{254}$. Silica gel chromatography was done on Merck silica gel 5715. Reactions were carried out under an argon atmosphere.

5′-*O*-(Monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (19). A mixture of 2′,3′-*O*-isopropylideneinosine (12.9 g, 41.8 mmol) and MMTrCl (15.5 g, 50.2 mmol) in pyridine (400 mL) was stirred at room temperature for 40 h. After MeOH (10 mL) was added, the resulting solution was stirred at room temperature for 30 min and evaporated. The residue was partitioned between EtOAc and H_2O, and the organic layer was washed with brine (200 mL), dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 4% MeOH in $CHCl_3$) to give **19** (24.3 g, quant) as white solids: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 13.19 (br s, 1 H), 8.12 (s, 1 H), 7.91 (s, 1 H), 7.37−6.76 (m, 14 H), 6.12 (d, 1 H, J = 2.1), 5.35 (dd, 1 H, J = 2.1, 6.2), 4.94 (dd, 1 H, J = 2.7, 6.2), 4.53 (ddd, 1 H, J = 2.7, 4.4, 6.1), 3.75 (s, 3 H), 3.35 (dd, 1 H, J = 6.1, 10.2), 3.27 (dd, 1 H, J = 4.4, 10.2), 1.63, 1.39 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 159.2, 158.6, 148.2, 145.1, 143.9, 143.8, 139.3, 135.0, 130.2, 129.2, 128.3, 127.8, 127.8, 127.1, 127.0, 125.4, 114.3, 113.2, 113.1, 91.3, 86.7, 86.5, 84.5, 81.9, 63.9, 55.2, 27.1, 25.4, 25.2; HRMS (FAB, positive) calcd for $C_{33}H_{33}N_4O_6$ 581.2400 (MH^+), found 581.2396; UV (MeOH) λ_{max} 251, sh 259 nm. Anal. Calcd for $C_{33}H_{32}N_4O_6 \cdot {}^1/_4H_2O$: C, 67.74; H, 5.60; N, 9.57. Found C, 67.56; H, 5.64; N, 9.86.

***N*-1-(2,4-Dinitrophenyl)-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (17).** A mixture of **19** (11.5 g, 19.7 mmol), K_2CO_3 (6.8 g, 49.3 mmol), and 2,4-dinitrochlorobenzene (10.0 g, 49.3 mmol) was stirred in DMF (200 mL) at 80 °C for 2.5 h. After the mixture was cooled to room temperature, the insoluble materials were filtered and washed with EtOAc. The filtrates and washings were combined and evaporated, and the residue was partitioned between EtOAc and H_2O. The organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 60% EtOAc in hexane) to give a rotameric mixture of **17** (13.2 g, 90%) as brown solids: ^{1}H NMR ($CDCl_3$, 270 MHz) δ 9.03, 9.01 (each d, each 0.5 H, J = 2.6) 8.64 (dd, 0.5 H, J = 2.6, 8.6), 8.61 (dd, 0.5 H, J = 2.6, 8.6), 7.95, 7.94 (each s, each 0.5 H), 7.89, 7.81 (each s, each 0.5 H), 7.67 (d, 0.5 H, J = 8.6), 7.49 (d, 0.5 H, J = 8.6), 7.41−6.75 (m, 14 H), 6.14 (d, 0.5 H, J = 2.6), 6.13 (d, 0.5 H, J = 2.6), 5.34 (dd, 0.5 H, J = 2.6, 6.6), 5.25 (dd, 0.5 H, J = 2.6, 5.9), 4.97 (dd, 0.5 H, J = 3.3, 6.6), 4.93 (dd, 0.5 H, J = 2.6, 5.9), 4.51 (m, 1 H), 3.75, 3.75 (each s, each 1.5 H), 3.44−3.32 (m, 2 H), 1.64, 1.41 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 158.5, 158.5, 155.0, 154.9, 147.9, 146.8, 154.1, 144.9, 143.9, 143.7, 139.7, 135.3, 134.9,131.8, 131.7, 130.3, 130.2, 128.8, 128.7, 128.2, 128.1, 127.8, 127.7, 126.9,124.6, 121.0, 120.9, 114.5, 114.5, 113.1, 113.0, 91.1, 91.0, 86.8, 86.6, 86.1, 85.9, 84.7, 84.5, 81.4, 81.4, 77.3, 77.0, 76.7, 63.8, 63.5, 55.1, 27.1, 27.1, 25.3, 25.3; HRMS (FAB, positive) calcd for $C_{39}H_{35}N_6O_{10}$ 747.2414 (MH^+), found 747.2401; UV (MeOH) λ_{max} 264, 253 nm. Anal. Calcd for $C_{39}H_{34}N_6O_{10} \cdot {}^1/_2H_2O$: C, 61.98; H, 4.67; N, 11.12. Found C, 62.23; H, 4.75; N, 10.77.

5-[[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropyridenedioxy)-4-(hydroxymethyl)cyclopentyl]aminomethyleneamino]-1-[5-*O*-(monomethoxytrityl)-2,3-*O*-(isopropylidene)-*β*-D-ribofuranosyl]imidazole-4-(*N*-2,4-dinitrophenyl)carboxamide (20). A mixture of **17** (560 mg, 0.75 mmol) and the optically active carbocyclic amine **18** (1.40 g, 7.5 mmol) in DMF (1.5 mL) was stirred at 50 °C for 21 h. The mixture was evaporated, and the residue was partitioned between EtOAc and H_2O. The aqueous layer was evaporated, and the residue was coevaporated with toluene (4.0 mL × 3) to give the carbocyclic amine **18** (1.22 g, 87%), which can be used repeatedly. The organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 60% EtOAc in hexane) to give a *cis*/*trans* mixture of **20** (520 mg, 74%) as an orange foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 12.24 (s, 1 H), 9.14−9.11 (m, 2 H,), 8.72 (d, 1 H, J = 3.8), 8.34 (m, 1 H), 7.42−6.82 (m, 15 H), 6.87 (m, 1 H), 7.37 (s, 1 H), 6.02 (d, 0.8 H), 5.90 (m, 0.2 H), 5.08 (dd, 0.8 H, J = 2.5, 6.0), 4.98 (m, 0.2 H), 4.77 (dd, 1 H, J = 3.1, 6.0), 4.61 (m, 2 H), 4.51 (m, 1 H), 4.40 (ddd, 1 H, J = 3.1, 3.6, 6.0), 3.85 (dd, 1 H, J = 2.8, 10.0), 3.77 (s, 3 H), 3.68 (dd, 1 H, J = 2.8, 10.0), 3.37 (dd, 1 H, J = 6.0, 10.2), 3.33 (dd, 1 H, J = 3.6, 10.2), 2.52 (m, 1 H), 2.37 (m, 1 H), 1.60, 1.45, 1.35, 1.28 (each s, each 3 H), 1.51 (m, 1 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 162.6, 158.6, 155.1, 147.7, 143.9, 141.0, 140.4, 135.1, 134.7, 130.9, 130.3, 129.3, 128.3, 127.9, 127.9, 127.0, 122.3, 121.5, 118.7, 114.2, 113.2, 110.6, 90.6, 87.0, 86.8, 85.2, 84.9, 84.0, 81.3, 64.4, 64.1, 55.9, 55.2, 47.0, 32.9, 27.2, 26.9, 25.5, 24.4, 11.4; HRMS (FAB, positive) calcd for $C_{48}H_{52}N_7O_{13}$ 934.3623 (MH^+), found 934.3615; UV (MeOH) λ_{max} 270, sh 286 nm. Anal. Calcd for $C_{48}H_{51}N_7O_{13} \cdot {}^1/_2H_2O$: C, 61.14; H, 5.56; N, 10.40. Found C, 61.39; H, 5.78; N, 10.10.

5-[[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[[(*tert*-butyldimethylsilyl)oxy]methyl]cyclopentyl]aminomethyleneamino]-1-[5-*O*-(monomethoxytrityl)-2,3-*O*-(isopropylidene)-*β*-D-ribofuranosyl]imidazole-4-(*N*-2,4-dinitrophenyl)carboxamide (21). A mixture of **20** (520 mg, 0.56 mmol), imidazole (114 mg, 1.67 mmol), and TBSCl (126 mg, 0.84 mmol) in DMF (2.8 mL) was stirred at room temperature for 30 min. After MeOH (1.0 mL) was added, the solution was stirred at room temperature for 10 min and evaporated. The

residue was partitioned between EtOAc and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 50% EtOAc in hexane) to give a *cis*/*trans* mixture of **21** (508 mg, 87%) as an orange foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 12.28 (s, 1 H), 9.26 (m, 0.2 H), 9.23 (d, 0.8 H, J = 9.5), 9.15 (d, 1 H, J = 2.5), 8.74 (d, 1 H, J = 4.0) 8.40 (dd, 1 H, J = 2.5, 9.5), 7.43−6.82 (m, 14 H), 7.37 (s, 1 H), 6.68 (dd, 1 H, J = 4.0, 7.8), 6.03 (d, 0.8 H, J = 2.9), 6.02 (m, 0.2 H), 5.08 (dd, 0.8 H, J = 2.9, 6.3), 4.87 (m, 0.2 H), 4.76 (dd, 1 H, J = 3.2, 6.3), 4.61 (m, 1 H), 4.55 (m, 1 H), 4.47 (m, 1 H), 4.41 (ddd, 0.8 H, J = 3.2, 3.7, 6.0), 4.32 (m, 0.2 H), 3.87 (dd, 1 H, J = 3.7, 10.2), 3.77 (s, 0.6 H), 3.77 (s, 2.4 H), 3.72 (dd, 1 H, J = 2.8, 10.2), 3.38 (dd, 1 H, J = 6.0, 10.2), 3.32 (dd, 1 H, J = 3.7, 10.2), 2.55 (m, 1 H), 2.37 (m, 1 H), 1.60, 1.44, 1.35, 1.27 (each s, each 3 H), 1.54 (m, 1 H), 0.96 (s, 9 H), 0.17 (s, 6 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 162.5, 158.7, 155.0, 147.6, 144.0, 143.9, 141.3, 140.4, 135.1, 134.7, 130.9, 130.4, 129.4, 128.4, 127.9, 127.9, 127.0, 122.3, 121.5, 118.9, 114.2, 113.2, 110.6, 90.6, 86.8, 86.8, 85.2, 85.0, 83.7, 81.4, 65.5, 64.1, 55.7, 55.2, 47.7, 33.0, 27.2, 27.0, 26.1, 26.1, 25.5, 24.6, 18.5, 14.2, −5.3, −5.4, −5.5; HRMS (FAB, positive) calcd for $C_{54}H_{65}N_7O_{13}NaSi$ 1070.4307 (MNa^+), found 1070.4330; UV (MeOH) λ_{max} 271, sh 286 nm. Anal. Calcd for $C_{54}H_{65}N_7O_{13}Si$: C, 61.87; H, 6.25; N, 9.35. Found C, 61.68; H, 6.35; N, 9.35.

2-Bromo-5-[[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(*tert*-butyldimethylsilyl)oxy]methyl]cyclopentyl]-aminomethyleneamino]-1-[5-*O*-(monomethoxytrityl)-2,3-*O*-(isopropylidene)-β-D-ribofuranosyl]imidazole-4-(*N*-2,4-dinitrophenyl)carboxamide (22). To a solution of **21** (1.23 g, 1.17 mmol) in THF (6.0 mL) was added NBA (194 mg, 1.36 mmol) in THF (6.0 mL) at −10 °C, and the mixture was stirred at the same temperature for 30 min. The resulting mixture was evaporated, and the residue was partitioned between EtOAc and H_2O. The organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 20% EtOAc in hexane) to give a *cis*/*trans* mixture of **22** (1.16 g, 88%) as an orange foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 12.15 (s, 0.8 H), 12.14 (s, 0.2 H), 9.22−9.16 (m, 2 H), 8.40 (dd, 1 H, J = 2.7, 9.5), 8.36 (d, 1 H, J = 4.1) 7.42−6.75 (m, 14 H), 6.80 (m, 1 H), 6.19 (d, 0.2 H, J = 2.5), 6.14 (d, 0.8 H, J = 2.6), 5.54 (dd, 0.8 H, J = 2.6, 6.8), 5.22 (dd, 0.2 H, J = 2.5, 6.8), 4.83 (dd, 0.8 H, J = 4.5, 6.8), 4.72 (m, 0.2 H), 4.52 (m, 1 H), 4.41 (m, 1 H), 4.34 (ddd, 0.8 H, J = 3.7, 4.5, 8.7), 4.32 (m, 1 H), 4.17 (m, 0.2 H), 3.85 (dd, 1 H, J = 3.4, 10.3), 3.74 (s, 3 H), 3.72 (dd, 1 H, J = 2.8, 10.3), 3.47 (dd, 1 H, J = 8.7, 9.9), 3.17 (dd, 1 H, J = 3.7, 9.9), 2.44 (m, 1 H), 2.37 (m, 1 H), 1.59, 1.39, 1.34, 1.24 (each s, each 3 H), 1.41 (m, 1 H), 0.98 (s, 9 H), 0.18 (s, 6 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 161.5, 158.5, 155.2, 150.0, 144.2, 144.2, 141.0, 140.6, 135.4, 134.8, 130.3, 129.4, 128.5, 128.4, 127.7, 127.7, 127.0, 126.8, 126.8, 122.3, 121.6, 119.5, 114.5, 113.1, 110.5, 91.3, 87.0, 86.4, 85.8, 84.1, 82.7, 81.6, 65.7, 64.8, 56.1, 55.1, 47.8, 32.7, 27.1, 26.9, 26.1, 25.3, 24.4, 18.5, 14.2, −5.4; HRMS (FAB, positive) calcd for $C_{54}H_{64}BrN_7O_{13}NaSi$ 1148.3413 (MNa^+), found 1148.3400; UV (MeOH) λ_{max} 269, sh 285 nm. Anal. Calcd for $C_{54}H_{64}BrN_7O_{13}Si$: C, 57.54; H, 5.72; N, 8.70. Found C, 57.53; H, 5.84; N, 8.93.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(*tert*-butyldimethylsilyl)oxy]methyl]cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (23). A mixture of **22** (1.16 g, 1.03 mmol) and K_2CO_3 (356 mg, 2.6 mmol) in DMF (10 mL) was stirred at 50 °C for 2.5 h. The mixture was evaporated, and the residue was partitioned between EtOAc and H_2O. The organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 25% EtOAc in hexane) to give **23** (953 mg, 98%) as a yellow foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.56 (s, 1 H, H-2), 7.38−6.74 (m, 14 H, Ar-H), 6.17 (d, 1 H, H-1′, J = 1.8), 5.46 (dd, 1 H, H-2′, J = 1.8, 6.4), 5.02 (dd, 1 H, H-3′, J = 3.7, 6.4), 4.87 (dd, 1 H, H-2″, J = 4.9, 6.5), 4.61−4.59 (m, 2 H, H-1″, H-3″), 4.46 (ddd, 1 H, H-4′, J = 3.7, 4.8, 7.1), 3.82 (dd, 1 H, H-5″a, J = 3.3, 10.0), 3.78 (s, 3 H, OCH_3), 3.72 (dd, 1 H, H-5″b, J = 5.4, 10.0), 3.31 (dd, 1 H, H-5′a, J = 7.1, 9.8), 3.23 (dd, 1 H, H-5′b, J = 4.8, 9.8), 2.31−2.27 (m, 3 H, H-4″, H-6″), 1.62, 1.54, 1.37, 1.28 (each s, each 3 H, *i*-PrMe), 0.91 (s, 9 H, *t*-Bu), 0.08 (s, 6 H, SiMe); NOE ($CDCl_3$, 400 MHz) irradiated H-2, observed H-1″ (9.7%), H-2″ (4.5%); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 158.5, 155.1, 147.5, 146.2, 144.3, 143.9, 135.3, 130.3, 128.5, 128.3, 127.6, 126.9, 126.8, 126.1, 125.5, 114.4, 113.1, 113.0, 91.4, 87.0, 86.3, 83.5, 82.9, 82.1, 81.0, 77.3, 77.0, 76.7, 65.0, 64.0, 63.7, 55.2, 46.7, 33.3, 31.9, 29.7, 29.6, 29.3, 29.3, 27.7, 27.2, 25.9, 25.4, 25.3, 22.6, 18.3, −5.4; HRMS (FAB, positive) calcd for $C_{48}H_{59}BrN_4NaO_9Si$ 965.3133 (MNa^+), found 965.3119; UV (MeOH) λ_{max} 258, 252, sh 271 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-(hydroxymethyl)cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (24). A mixture of **23** (944 mg, 1.0 mmol) and TBAF (1.0 M in THF, 3.4 mL, 3.4 mmol) in THF (3.4 mL) was stirred at room temperature for 1.5 h. The resulting solution was evaporated, and the residue was purified by column chromatography (SiO_2, 70% EtOAc in hexane) to give **24** (748 mg, 90%) as a yellow foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.54 (s, 1 H, H-2), 7.38−6.73 (m, 14 H, Ar-H), 6.17 (d, 1 H, H-1′, J = 1.6), 5.46 (dd, 1 H, H-2′, J = 1.6, 6.2), 5.03 (dd, 1 H, H-3′, J = 3.6, 6.2), 4.94 (m, 1 H, H-2″), 4.71 (m, 1 H, H-3″), 4.47 (m, 1 H, H-1″), 4.45 (ddd, 1 H, H-4′, J = 3.6, 4.9, 7.0), 3.82 (m, 2 H, H-5″), 3.78 (s, 3 H, OCH_3), 3.31 (dd, 1 H, H-5′a, J = 7.0, 9.8), 3.25 (dd, 1 H, H-5′b, J = 4.9, 9.8), 2.40−2.29 (m, 3 H, H-4″, H-6″), 1.62, 1.55, 1.37, 1.29 (each s, each 3 H, *i*-PrMe); NOE ($CDCl_3$, 400 MHz) irradiated H-2, observed H-1″ (10.4%), H-2″ (1.7%); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 158.5, 155.2, 147.7, 146.4, 144.3, 144.0, 135.3, 130.3, 128.4, 127.6, 126.9, 126.4, 125.6, 114.4, 113.1, 113.0, 91.4, 86.9, 86.3, 83.4, 83.2, 82.0, 77.3, 77.0, 76.7, 66.4, 64.1, 63.9, 55.3, 46.6, 32.4, 27.7, 27.2, 25.4, 25.2, 11.4 ; HRMS (FAB, positive) calcd for $C_{42}H_{45}BrN_4NaO_9$ 851.2268 (MNa^+), found 851.2236; UV (MeOH) λ_{max} 261 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (25). A mixture of **24** (166 mg, 200 μmol), tetrazole (56 mg, 800 μmol), and dianilinophosphorochloridate (213 mg, 800 μmol) in pyridine (2.0 mL) was stirred at room temperature for 47 h. Aqueous sodium acetate (2 M, 3.0 mL) was added, and the resulting solution was stirred at room temperature for 1.0 h. Water and $CHCl_3$ were added, and the resulting mixture was partitioned. The organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 15% acetone in $CHCl_3$) to give **25** (209 mg, 98%) as a yellow foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.46 (s, 1 H), 7.37−6.73 (m, 24 H), 6.17 (d, 1 H, J = 2.1), 6.02, 5.90 (each br s, each 1 H), 5.45 (dd, 1 H, J = 2.1, 6.4), 5.03 (dd, 1 H, J = 3.7, 6.4), 4.98 (m, 1 H), 4.80 (m, 1 H), 4.45 (ddd, 1 H, J = 3.7, 5.0, 6.8), 4.37−4.28 (m, 3 H), 3.77 (s, 3 H), 3.32 (dd, 1 H, J = 6.8, 9.9), 3.23 (dd, 1 H, J = 5.0, 9.9), 2.53−2.24 (m, 3 H), 1.62, 1.53, 1.37, 1.25 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 158.2, 155.0, 147.6, 146.3, 144.1, 143.8, 139.4, 135.2, 130.2, 129.0, 128.2, 128.2, 127.7, 127.5, 127.0, 126.7, 126.3, 125.5, 121.7, 121.6, 117.9, 117.8, 114.3, 113.1, 113.0, 112.8, 91.3, 86.8, 86.2, 83.4, 82.8, 81.9, 81.8, 66.5, 66.1, 63.8, 55.2, 45.4, 45.3, 31.9, 29.2, 27.7, 27.2, 25.4, 25.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.41 (s); HRMS (FAB, positive) calcd for $C_{54}H_{56}BrN_6NaO_{10}P$ 1081.2877 (MNa^+), found 1081.2840; UV (MeOH) λ_{max} 267, 262, 233 nm. Anal. Calcd for $C_{54}H_{56}BrN_6O_{10}P\cdot{}^1/_5CHCl_3$: C, 60.06; H, 5.23; N, 7.75. Found C, 60.10; H, 5.35; N, 7.69.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-2′,3′-*O*-isopropylideneinosine (26). A solution of **25** (376 mg, 355 μmol) in 80% aqueous AcOH (10 mL) was stirred at room temperature for 3 h. The resulting mixture was evaporated, and the residue was purified by column chromatography (SiO_2, 3% MeOH in $CHCl_3$) to give **26** (225 mg, 80%) as a yellow foam: ^{1}H NMR ($CDCl_3$, 270 MHz) δ 8.00 (s, 1 H), 7.22−6.87 (m, 10 H), 6.13 (d, 1 H, J = 8.6), 6.08 (d, 1 H, J = 5.3), 6.00 (d, 1 H, J = 8.6), 5.16 (dd, 1 H, J = 5.3, 5.9), 5.04 (dd, 1 H, J = 1.6, 5.9), 5.00 (dd, 1 H, J = 4.3, 6.6), 4.93 (br, 1 H), 4.78 (dd, 1 H, J = 2.0, 6.6), 4.57 (ddd, 1 H, J = 4.3, 5.3, 5.9), 4.47 (m, 1

H,), 4.39−4.22 (m, 2 H), 3.96−3.74 (m, 2 H), 2.54−2.24 (m, 3 H), 1.67, 1.52, 1.39, 1.25 (each s, each 3 H); ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.62 (s); HRMS (FAB, positive) calcd for $C_{34}H_{40}BrN_6NaO_9P$ 809.1676 (MNa^+), found 809.1646; UV (MeOH) λ_{max} 257, 233, sh 273 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-[bis(phenylthio)phosphoryl]-2′,3′-*O*-isopropylideneinosine (27). A mixture of **26** (110 mg, 130 μmol), tetrazole (27 mg, 390 μmol), PSS (149 mg, 390 μmol), and TPSCl (79 mg, 260 μmol) in pyridine (1.3 mL) was stirred at room temperature for 40 h. After ice-cooled H_2O (2.0 mL) was added, the resulting solution was stirred at room temperature for 30 min, and then H_2O and $CHCl_3$ were added. The resulting mixture was partitioned, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (SiO_2, 90% EtOAc in hexane) to give **27** (99 mg, 72%) as a yellow foam: ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.85 (s, 1 H), 7.47−6.87 (m, 20 H), 6.43, 6.41 (each d, each 1 H, J = 8.5, 8.5), 6.18 (d, 1 H, J = 1.6), 5.45 (dd, 1 H, J = 1.6, 6.2), 5.11 (dd, 1 H, J = 3.5, 6.2), 5.00 (dd, 1 H, J = 4.5, 6.3), 4.73 (m, 1 H), 4.55 (m, 1 H), 4.42−4.27 (m, 5 H), 2.48 (m, 1 H), 2.33 (m, 1 H), 2.23 (m, 1 H), 1.62, 1.50, 1.39, 1.21 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 155.2, 147.8, 147.2, 140.0, 135.3, 135.3, 135.0, 135.0, 129.7, 129.4, 129.2, 129.1, 126.1, 125.7, 125.7, 125.6, 121.8, 121.7, 118.1, 118.1, 114.7, 113.2, 91.3, 85.5, 85.4, 83.6, 83.1, 81.8, 81.2, 66.2, 66.1, 66.0, 65.9, 45.1, 32.2, 27.6, 27.2, 25.4, 25.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.38 (s), 50.9 (s); HRMS (FAB, positive) calcd for $C_{46}H_{50}BrN_6O_{10}P_2S_2$ 1051.1689 (MH^+), found 1051.1690; UV (MeOH) λ_{max} 249, 230, sh 265 nm. Anal. Calcd for $C_{46}H_{49}BrN_6O_{10}P_2S_2$: C, 52.52; H, 4.70; Br, 7.60; N, 7.90; S, 6.10. Found C, 52.68; H, 5.01; Br, 7.42; N, 7.83; S, 5.71.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(phosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-[(phenylthio)phosphoryl]-2′,3′-*O*-isopropylideneinosine (15). A mixture of **27** (105 mg, 100 μmol) and isoamyl nitrite (202 μL, 1.5 mmol) in pyridine−AcOH−Ac_2O (2:1:1, v/v, 3.0 mL) was stirred at room temperature for 8 h. After the reaction mixture was evaporated (at <30 °C), the residue was dissolved in a mixture of H_3PO_2 (102 μL, 2.0 mmol), Et_3N (139 μL, 1.0 mmol), and pyridine (2.5 mL), and the resulting solution was stirred at room temperature for 11 h. After the mixture was evaporated (at <30 °C), the residue was partitioned between $CHCl_3$ and H_2O. After pyridine (5 mL) was added to the aqueous layer, the resulting solution was evaporated (at <30 °C), and the residue was dissolved in TEAA buffer (0.1 M, pH 7.0, 10 mL). The solution was applied to a C_{18} reversed phase column (1.8 × 15 cm), and the column was developed using a linear gradient of 0−40% CH_3CN in TEAA buffer (0.1 M, pH 7.0, 400 mL). Appropriate fractions were evaporated, and excess TEAA was removed by C_{18} reversed phase column chromatography (1.8 × 13 cm, eluted with 20% aqueous CH_3CN) to give **15** (89 mg, 88%) as a triethylammonium salt: ^{1}H NMR (D_2O, 500 MHz) δ 8.44 (s, 1 H, H-2), 7.24−7.09 (m, 5 H, Ar-H), 6.35 (s, 1 H, H-1′), 5.80 (d, 1 H, H-2′, J = 6.4), 5.28 (dd, 1 H, H-3′, J = 2.7, 6.4), 5.07−5.01 (m, 2 H, H-1″, H-2″), 4.79 (m, 1 H, H-3″), 4.68 (m, 1 H, H-4′), 4.44 (m, 1 H, H-5′a), 4.11 (m, 1 H, H-5′b), 3.97 (m, 2 H, H-5″), 3.19 (q, 12 H, $-CH_2N$, J = 7.3), 2.51 (m, 1 H, H-4″), 2.42 (m, 1 H, H-6″a), 2.15 (m, 1 H, H-6″b), 1.66, 1.60, 1.44, 1.38 (each s, each 3 H, *i*-PrMe), 1.27 (t, 18 H, CH_3CH_2N, J = 7.3); ^{13}C NMR (D_2O, 125 MHz) δ 159.0, 150.5, 150.2, 133.6, 133.6, 132.5, 132.5, 131.6, 130.6, 130.0, 126.8, 117.6, 117.1, 94.6, 89.8, 89.7, 86.5, 86.2, 84.2, 84.0, 68.5, 68.3, 64.8, 49.5, 47.2, 47.1, 35.7, 29.1, 28.6, 27.1, 26.9, 11.0; ^{31}P NMR (D_2O, 125 MHz, decoupled with ^{1}H) δ 1.81 (s), 17.9 (s); HRMS (FAB, negative) calcd for $C_{28}H_{34}BrN_4O_{13}P_2S$ 807.0502 [(M − H)$^-$], found 807.0516; UV (H_2O) λ_{max} 246, sh 259, 284 nm.

Synthesis of Cyclic 8-Bromo-IDP-Carbocyclic-Ribose Diacetonide (12). Typical Procedure A (Entry 4). A solution of **15** (9.5 mg, 9.4 μmol, 142 OD_{260} units) in pyridine (8.0 mL) was added slowly over 15 h, using a syringe pump, to a mixture of I_2 (50 mg, 190 μmol) and MS 3A (500 mg) in pyridine (8.0 mL) at room temperature in the dark. The MS 3A was filtered off with Celite and washed with H_2O. To the combined filtrate and washing was added TEAA buffer (2 M, pH 7.0, 1.0 mL), and the resulting solution was evaporated. The residue was partitioned between $CHCl_3$ and H_2O. The aqueous layer was evaporated, and the residue was dissolved in 0.1 M TEAA buffer (5.0 mL), which was applied to a C_{18} reversed phase column (1.1 × 11 cm). The column was developed using a linear gradient of 0−40% CH_3CN in TEAA buffer (0.1 M, pH 7.0, 200 mL). Appropriate fractions were evaporated under reduced pressure, and excess TEAA was removed by C_{18} reversed phase column chromatography (1.1 × 11 cm, eluted with 20% aqueous CH_3CN) to give **12** (8.5 mg, 113 OD_{260} units, quant) as a triethylammonium salt, the spectra data of which were in accord with those reported previously.[6]

Typical Procedure B (Entry 7). A solution of **15** (9.5 mg, 9.4 μmol, 142 OD_{260} units) in NMP/HMPA (3:1, 8.0 mL) was added slowly over 15 h, using a syringe pump, to a mixture of $AgNO_3$ (48 mg, 280 μmol) and molecular sieves 3A (500 mg) in the same solvent (8.0 mL) at room temperature in the dark. The mixture was cooled with an ice bath, into which H_2S for 10 min and then argon for 30 min were bubbled, and the resulting mixture was evaporated. The insoluble materials were removed by centrifugation, and the supernatant and washings (NMP, 2.0 mL) were combined and partitioned between $CHCl_3$ and H_2O. The aqueous layer was evaporated, dissolved in water (100 mL), and applied to a DEAE-Sephadex A-25 column (HCO_3^- form, 1.8 × 15 cm). The column was developed using a linear gradient of 0−0.5 M TEAB buffer (0.5 M, pH 8.6, 200 mL). Appropriate fractions were evaporated under reduced pressure, and excess TEAB was coevaporated with H_2O. The residue was dissolved in H_2O (3.0 mL), and applied to a C_{18} reversed phase column (1.1 × 11 cm). The column was developed using a linear gradient of 0−40% CH_3CN in TEAA buffer (0.1 M, pH 7.0, 100 mL). Appropriate fractions were evaporated under reduced pressure, and excess TEAA was removed by C_{18} reversed phase column chromatography (1.1 × 11 cm, eluted with 20% aqueous CH_3CN) to give **12** (2.5 mg, 33 OD_{260} units, 29%) as a triethylammonium salt.

HPLC Analysis of the Reactions. To the reaction mixture (30 μL) was added TEAA buffer (2 M, pH 7.0, 10 μL), and the resulting solution was evaporated. The residue was coevaporated with H_2O and partitioned between H_2O and $CHCl_3$. The aqueous layer was filtered with a syringe filter (cellulose acetate) and analyzed by HPLC [column YMS-ODS-M-80, 4.6 × 150 mm; 5−80% MeCN/0.1 M TEAA buffer (pH 7.0), 1.0 mL/min, 30 min; 254 nm]. Compounds **11**, **12**, and **14** were eluted at 14.1, 14.7, and 15.8 min, respectively, which were identified with the authentic samples synthesized previously,[6,12] and a peak observed at 3.1 min was unknown nonnucleosidic compound due to its UV spectrum.

***N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[(hydroxymethyl)cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (28).** Compound **28** (240 mg, quant) was obtained from **20** (300 mg, 320 μmol) as described for the synthesis of **23** after purification by column chromatography (SiO_2, 10% acetone in EtOAc): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.85 (s, 1 H, H-8), 7.77 (s, 1 H, H-2), 7.38−6.76 (m, 14 H, Ar-H), 6.06 (d, 1 H, H-1′, J = 2.5), 5.23 (dd, 1 H, H-2′, J = 2.5, 6.2), 5.04 (dd, 1 H, H-2″, J = 4.3, 6.6), 4.90 (dd, 1 H, H-3′, J = 3.0, 6.2), 4.75 (dd, 1 H, H-3″, J = 3.3, 6.6), 4.56 (ddd, 1 H, H-1″, J = 4.3, 9.2, 9.2), 4.48 (ddd, 1 H, H-4′, J = 3.0, 4.4, 5.9), 3.83 (m, 2 H, H-5″), 3.79 (s, 3 H, OCH_3), 3.35 (dd, 1 H, H-5′a, J = 5.9, 10.3), 3.30 (dd, 1 H, H-5′b, J = 4.4, 10.3), 2.41 (m, 1 H, H-4″), 2.35 (m, 2 H, H-6″), 1.61, 1.55, 1.37, 1.30 (each s, each 3 H, *i*-PrMe); NOE ($CDCl_3$, 400 MHz) irradiated H-2, observed H-1″ (5.9%), H-2″ (1.4%); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 158.6, 156.6, 146.7, 144.1, 143.9, 139.2, 135.1, 130.3, 128.3, 127.8, 127.0, 125.6, 114.5, 113.2, 113.0, 91.0, 86.7, 86.2, 84.6, 83.5, 82.5, 81.8, 66.1, 64.1, 63.9, 55.3, 46.8, 32.7, 30.9, 27.8, 27.2, 25.4, 25.3, 11.5; HRMS (FAB, positive) calcd for $C_{42}H_{47}N_4O_9$ 751.3342 (MH^+), found 751.3373; UV (MeOH) λ_{max} 270 nm. Anal. Calcd for $C_{42}H_{46}N_4O_9 \cdot {}^1/_2H_2O$: C, 66.39; H, 6.23; N, 7.37. Found C, 66.60; H, 6.36; N, 7.29.

***N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (29).** Compound **29** (245 mg, 85%) was obtained from **28** (220 mg, 293 μmol) as described for the synthesis of **25** after purification by column chromatography (SiO_2, 10% acetone in EtOAc): ^{1}H NMR ($CDCl_3$, 270 MHz) δ 7.87 (s, 1 H), 7.71 (s, 1 H), 7.38−6.75 (m, 24 H), 6.06 (d, 1 H, J = 2.6), 5.91, 5.77 (each d, each 1 H, J = 7.9), 5.22 (dd, 1 H, J = 2.6, 6.6), 5.08 (dd, 1 H, J = 3.3, 6.6), 4.91−4.84 (m, 2 H), 4.49−4.31 (m, 4 H), 3.77 (s, 3 H), 3.33 (m, 1 H), 2.56−2.50 (m, 2 H), 2.29 (m, 1 H), 1.62, 1.54, 1.37, 1.28 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 158.4, 156.4, 146.6, 146.5, 143.8, 143.6, 139.5, 139.1, 134.8, 130.2, 129.0, 129.0, 128.1, 128.1, 127.7, 127.6, 126.9, 126.8, 125.5, 121.6, 121.5, 117.9, 117.8, 114.3, 113.0, 113.0, 90.8, 86.6, 86.1, 84.5, 83.2, 81.9, 81.6,66.2, 66.1, 63.7, 55.2, 53.7, 45.5, 45.5, 32.1, 30.9, 29.2, 27.7, 27.2, 25.4, 25.2, 14.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.68 (s); HRMS (FAB, positive) calcd for $C_{54}H_{58}N_6O_{10}P$ 981.3951 (MH^+), found 981.3895; UV (MeOH) λ_{max} 271, 236 nm. Anal. Calcd for $C_{54}H_{57}N_6O_{10}P\cdot{}^1/_2H_2O$: C, 65.51; H, 5.90; N, 8.49. Found C, 65.49; H, 6.14; N, 8.14.

***N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-2′,3′-*O*-isopropylideneinosine (30).** Compound **30** (118 mg, 94%) was obtained from **29** (175 mg, 178 μmol) as described for the synthesis of **26** after purification by column chromatography (SiO_2, 4% MeOH in $CHCl_3$): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 8.01 (s, 1 H), 7.94 (s, 1 H), 7.18−6.87 (m, 10 H), 6.40 (d, 1 H, J = 8.7), 6.28 (d, 1 H, J = 8.7), 5.90 (d, 1 H, J = 2.1), 5.09−5.00 (m, 4 H), 4.58 (m, 1 H), 4.49 (m, 1 H), 4.35−4.23 (m, 2 H), 3.96 (dd, 1 H, J = 1.4, 12.5), 3.80 (m, 1 H), 2.52 (m, 1 H), 2.39 (m, 1 H), 2.26 (m, 1 H), 1.64, 1.51, 1.37, 1.24 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 156.4, 147.1, 146.2, 139.9, 139.6, 129.2, 129.2, 126.2, 121.9, 121.8, 118.2, 118.1, 118.1, 114.3, 113.3, 93.3, 86.2, 83.9, 83.2, 81.8, 81.3, 65.9, 63.0, 32.3, 27.6, 27.5, 25.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.81 (s); HRMS (FAB, positive) calcd for $C_{34}H_{42}N_6O_9P$ 709.2750 (MH^+), found 709.2743; UV (MeOH) λ_{max} 274, 252 nm.

***N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[[(dianilinophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-[bis(phenylthio)phosphoryl]-2′,3′-*O*-isopropylideneinosine (31).** Compound **31** (107 mg, 80%) was obtained from **30** (98 mg, 138 μmol) as described for the synthesis of **27** after purification by column chromatography (SiO_2, 90% EtOAc in hexane): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.91 (s, 1 H), 7.84 (s, 1 H), 7.50−6.85 (m, 20 H), 6.20, 6.06 (each d, each 1 H, J = 8.5, 8.5), 6.05 (d, 1 H, J = 2.6), 5.07 (m, 2 H), 4.92 (dd, 1 H, J = 2.6, 6.2), 4.82 (m, 1 H), 4.53 (m, 1 H), 4.45−4.40 (m, 3 H), 4.37−4.25 (m, 2 H), 2.52 (m, 1 H), 2.43 (m, 1 H), 2.26 (m, 1 H), 1.61, 1.52, 1.36, 1.20 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 156.4, 147.0, 146.7, 139.7, 139.1, 135.2, 135.2, 135.0, 135.0, 129.7, 129.6, 129.4, 129.4, 129.0, 129.0, 125.6, 125.6, 125.5, 125.5, 121.6, 121.5, 118.0, 118.0, 114.7, 113.1, 90.5, 84.6, 84.5, 84.3, 83.2, 81.8, 80.9, 66.2, 65.5, 45.2, 45.2, 32.3, 27.6, 27.1, 25.2, 25.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.89 (s), 51.2 (s); HRMS (FAB, positive) calcd for $C_{46}H_{51}N_6O_{10}P_2S_2$ 973.2583 (MH^+), found 973.2547; UV (MeOH) λ_{max} 267, 255, 233 nm.

***N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(Isopropylidenedioxy)-4-[[(phosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-[(phenylthio)phosphoryl]-2′,3′-*O*-isopropylideneinosine (16).** Using **31** (86 mg, 88 μmol) as a substrate, the reaction and purification were carried out as described above for the synthesis of **15** to give a mixture of **16** and a byproduct.[21] The mixture was dissolved in aqueous $NaHCO_3$ (2%, 5 mL), and the solution was stirred at 37 °C for 15 h. TEAA buffer (0.1 M, pH 7.0, 5.0 mL) was added, and the resulting solution was applied to a C_{18} reversed phase column (1.1 × 11 cm). The column was developed using a linear gradient of 0−40% CH_3CN in TEAA buffer (0.1 M, pH 7.0, 300 mL). Appropriate fractions were evaporated under reduced pressure, and excess TEAA was removed by C_{18} reversed phase column chromatography (1.1 × 11 cm, 20% aqueous CH_3CN) to give pure **16** (53 mg, 65%) as a triethylammonium salt: ^{1}H NMR (D_2O, 500 MHz) δ 8.38 (s, 1 H, H-8), 8.20 (s, 1 H, H-2), 7.21−7.06 (m, 5 H, Ar-H), 6.29 (d, 1 H, H-1′, J = 2.0), 5.45 (dd, 1 H, H-2′, J = 2.0, 6.1), 5.10 (dd, 1 H, H-3′, J = 2.0, 6.1), 5.04 (ddd, 1 H, H-1″, J = 1.6, 6.8, 7.6), 4.99(dd, 1 H, H-2″, J = 1.6, 6.9), 4.77 (dd, 1 H, H-3″, J = 1.9, 6.9), 4.68 (m, 1 H, H-4′), 4.27 (m, 1 H, H-5′a), 4.12 (m, 1 H, H-5′b), 3.98 (m, 2 H, H-5″), 3.19 (q, 12 H, $-CH_2N$, J = 7.3), 2.51 (m, 1 H, H-4″), 2.39 (m, 1 H, H-6″a), 2.16 (m, 1 H, H-6″b), 1.64, 1.58, 1.42, 1.235 (each s, each 3 H, *i*-PrMe), 1.27 (t, 18 H, CH_3CH_2N, J = 7.3); ^{13}C NMR (D_2O, 125 MHz) δ 160.2, 150.1, 149.9, 134.9, 131.6, 130.2, 125.9, 117.5, 117.0, 93.3, 88.8, 86.6, 84.3, 76.2, 75.0, 68.2, 68.1, 64.9, 49.4, 47.1, 35.5, 29.2, 28.7, 27.0, 11.0; ^{31}P NMR (D_2O, 125 MHz, decoupled with ^{1}H) δ 1.17 (s), 18.0 (s); HRMS (FAB, negative) calcd for $C_{28}H_{35}N_4O_{13}P_2S$ 729.1396 [(M − H)$^-$], found 729.1417; UV (H_2O) λ_{max} 244, sh 270 nm.

Cyclic IDP-Carbocyclic-Ribose Diacetonide (32). Compound **16** (4.4 mg, 4.7 μmol) was treated under the same conditons as in entry 4 in Table 1 to give **32** (2.7 mg, 81%) after purification as described for the synthesis of **12** (procedure A): ^{1}H NMR (D_2O, 500 MHz) δ 8.52 (s, 1 H, H-8), 8.16 (s, 1 H, H-2), 6.32 (s, 1 H, H-1′), 5.72 (d, 1 H, H-2′, J = 6.0), 5.47 (dd, 1 H, H-3′, J = 6.0), 4.88 (m, 1 H, H-1″), 4.74 (m, 2 H, H-2″, H-3″), 4.56 (m, 1 H, H-4′), 4.14 (m, 1 H, H-5″a), 4.04 (m, 2 H, H-5′), 3.74 (m, 1 H, H-5″b), 3.19 (q, 12 H, $-CH_2N$, J = 7.3), 2.88−2.80 (m, 2 H, H-4″, H-6″a), 2.62 (m, 1 H, H-6″b), 1.64, 1.58, 1.45, 1.37 (each s, each 3 H, *i*-PrMe), 1.27 (t, 18 H, CH_3CH_2N, J = 7.3); ^{13}C NMR (D_2O, 125 MHz) δ 161.3, 149.5, 148.1, 145.0, 126.8, 117.2, 113.7, 94.2, 90.5, 89.0, 86.6, 85.6, 84.7, 71.2, 68.1, 67.9, 67.1, 49.4, 47.5, 29.7, 28.6, 27.0, 26.3, 22.8, 11.0; ^{31}P NMR (D_2O, 125 MHz, decoupled with ^{1}H) δ −10.20 (d, J = 15.3), −10.60 (d, J = 15.3); HRMS (FAB, negative) calcd for $C_{22}H_{29}N_4O_{13}P_2$ 619.1206 [(M − H)$^-$], found 619.1201; UV (H_2O) λ_{max} 250, sh 273 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(bisphenylthiophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-(monomethoxytrityl)-2′,3′-*O*-isopropylideneinosine (33). Compound **33** (165 mg, 75%) was obtained from **24** (166 mg, 200 μmol) as described for the synthesis of **27** after purification by column chromatography (SiO_2, 60% EtOAc in hexane): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.58−6.75 (m, 24 H), 7.48 (s, 1 H), 6.18 (d, 1 H, J = 2.2), 5.47 (dd, 1 H, J = 2.2, 6.4), 5.03 (dd, 1 H, J = 3.7, 6.4), 4.87 (dd, 1 H, J = 4.4, 6.7), 4.55 (m, 1 H), 4.49−4.40 (m, 3 H), 4.32 (m, 1 H), 3.32 (dd, 1 H, J = 6.9, 9.9), 3.25 (dd, 1 H, J = 4.9, 9.9), 2.47 (m, 1 H), 2.27−2.21 (m, 2 H), 1.62, 1.54, 1.37, 1.27 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 158.5, 155.0, 147.7, 146.2, 144.3, 144.0, 137.8, 135.4, 135.4, 135.3, 130.3, 129.5, 129.4, 129.4, 129.0, 128.4, 128.3, 128.2, 127.7, 126.9, 126.9, 126.3, 126.2, 125.6, 125.3, 114.5, 113.5, 113.0, 91.4, 86.9, 86.3, 83.5, 82.8, 81.9, 80.9, 68.3, 68.3, 65.2, 63.9, 55.2, 45.3, 45.2, 32.8, 27.6, 27.2, 25.4, 25.3, 21.4; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 50.0 (s); HRMS (FAB, positive) calcd for $C_{54}H_{54}BrN_4NaO_{10}PS_2$ 1115.2100 (MNa^+), found 1115.2070; UV (MeOH) λ_{max} 272, 249, sh 280 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(bisphenylthiophosphoryl)oxy]methyl]cyclopentyl]-2′,3′-*O*-isopropylideneinosine (34). Compound **34** (98 mg, 87%) was obtained from **33** (150 mg, 137 μmol) as described for the synthesis of **26** after purification by column chromatography (SiO_2, 20% acetone in $CHCl_3$): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.97 (s, 1 H), 7.58−7.33 (m, 10 H), 6.09 (d, 1 H, J = 5.0), 5.18 (dd, 1 H, J = 5.0, 6.1), 5.04 (dd, 1 H, J = 1.1, 6.1), 4.93 (dd, 1 H, J = 4.9, 6.4), 4.84 (m, 1 H), 4.64 (m, 1 H), 4.55 (m, 1 H), 4.47 (m, 1 H), 4.40−4.30 (m, 2 H), 3.93−3.77 (m, 2 H), 2.48−2.45 (m, 1 H), 2.27−2.21 (m, 2 H), 1.66, 1.52, 1.38, 1.27 (each s, each 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 154.7, 147.2, 146.8, 135.3, 135.3, 129.5, 129.4, 126.2, 126.1, 126.1, 125.9, 114.4, 113.5, 93.1, 85.5, 82.8, 82.6, 81.0, 80.6, 68.0, 68.0, 65.0, 62.9, 53.8, 44.9, 44.9, 32.8, 29.2, 27.5, 27.5, 25.3, 25.2; ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 49.9 (s);

(21) The HPLC analysis showed it included a byproduct, which might be an acetylated product of **13** at the 5′-phospate moiety, since treatment of the mixture with aqueous $NaHCO_3$ gave the desired 5′-phosphate **13** as a sole product.

HRMS (FAB, positive) calcd for $C_{34}H_{39}BrN_4O_9PS_2$ 821.1080 (MH^+), found 821.1064; UV (MeOH) λ_{max} 250, sh 274 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(bisphenylthiophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-(dianilinophosphoryl)-2′,3′-*O*-isopropylideneinosine (35). Compound **35** (90 mg, 80%) was obtained from **34** (88 mg, 107 μmol) as described for the synthesis of **25** after purification by column chromatography (SiO_2, 5% MeOH in $CHCl_3$): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 7.71 (s, 1 H), 7.59−6.76 (m, 20 H), 6.14 (d, 1 H, J = 1.6), 5.29 (dd, 1 H, J = 1.6, 5.3), 5.14 (dd, 1 H, J = 3.4, 5.3), 4.93 (dd, 1 H, J = 4.8, 6.7), 4.52 (m, 1 H), 4.47−4.45 (m, 2 H), 4.38−4.33 (m, 3 H), 4.23 (dd, 1 H, J = 3.3, 10.4), 2.36 (m, 1 H), 2.05 (m, 2 H), 1.56, 1.53, 1.29, 1.19 (each s, each 3 H); ^{31}P NMR ($CDCl_3$, 125 MHz, decoupled with ^{1}H) δ 2.94 (s), 50.1 (s); HRMS (FAB, positive) calcd for $C_{46}H_{50}BrN_6O_{10}P_2S_2$ 1051.1688 (MH^+), found 1051.1600; UV (MeOH) λ_{max} 232, sh 275, 257 nm.

8-Bromo-*N*-1-[(1*R*,2*S*,3*R*,4*R*)-2,3-(isopropylidenedioxy)-4-[[(phenylthiophosphoryl)oxy]methyl]cyclopentyl]-5′-*O*-(phosphoryl)-2′,3′-*O*-isopropylideneinosine (13). Compound **13** (47 mg, 75%) was obtained from **35** (90 mg, 86 μmol) as described for the synthesis of **16**: ^{1}H NMR (D_2O, 500 MHz) δ 8.39 (s, 1 H, H-2), 7.57−7.20 (m, 5 H, Ar-H), 6.32 (d, 1 H, H-1′, J = 1.7), 5.70 (d, 1 H, H-2′, J = 1.7, 6.5), 5.31 (dd, 1 H, H-3′, J = 4.0, 6.5),4.98 (m, 1 H, H-2″), 4.95 (m, 1 H, H-1″), 4.59 (m, 1 H, H-3″), 4.48 (m, 1 H, H-4′), 4.07−3.97 (m, 4 H, H-5′, H-5″), 3.18 (q, 12 H, $-CH_2N$, J = 7.3), 2.41 (m, 1 H, H-4″), 2.26−1.93 (m, 2 H, H-6″), 1.65, 1.54, 1.44, 1.29 (each s, each 3 H, *i*-PrMe), 1.26 (t, 18 H, CH_3CH_2N, J = 7.3); ^{13}C NMR (D_2O, 125 MHz) δ 159.6, 150.7, 149.4, 136.2, 131.8, 130.4, 126.8, 117.9, 116.7, 93.2, 88.9, 85.8, 83.8, 76.9, 74.6, 69.3, 67.2, 65.3, 63.5, 49.3, 46.6, 45.4, 35.2, 32.9, 30.8, 29.1, 28.7, 27.1, 10.9; ^{31}P NMR (D_2O, 125 MHz, decoupled with ^{1}H) δ 0.62 (s), 17.5 (s); HRMS (FAB, negative) calcd for $C_{28}H_{34}BrN_4O_{13}P_2S$ 809.0481 [$(M - H)^-$], found 809.0475; UV (H_2O) λ_{max} 249, sh 282 nm.

Cyclic 8-Bromo-IDP-Carbocyclic-Ribose (6). A solution of **12** (48 OD_{254} units) in 60% aqueous HCO_2H (1.0 mL) was stirred at room temperature for 3.5 h. After the solvent was evaporated, the residue was dissolved in H_2O (50 mL) and applied to a DEAE-Sephadex A-25 column (HCO_3^- form, 1.1 × 15 cm). The column was developed using a linear gradient of 0−0.5 M TEAB buffer (pH 8.6, 200 mL). Appropriate fractions were evaporated, and excess TEAB was coevaporated with water. The residue was freeze-dried to give **6** (18 OD_{254} units, 37%) as a triethylammonium salt: ^{1}H NMR (D_2O, 500 MHz) δ 9.00 (s, 1 H, H-2), 6.17 (d, 1 H, H-1′, J = 6.6), 5.24 (m, 1 H, H-2′), 5.17 (m, 1 H, H-1″), 4.65 (m, 1 H, H-2″), 4.60 (m, 1 H, H-3′), 4.41 (m, 1 H, H-4′), 4.26−4.07 (m, 5 H, H-3″, H-5′, H-5″), 3.18 (q, 12 H, $-CH_2N$, J = 7.3), 2.91 (m, 1 H, H-6″a), 2.46 (m, 1 H, H-4″), 2.20 (m, 1 H, H-6″b), 1.27 (t, 18 H, CH_3CH_2N, J = 7.3); ^{13}C NMR (D_2O, 125 MHz) δ 160.0, 151.4, 151.2, 130.8, 126.6, 93.6, 87.9, 87.8, 81.3, 75.7, 75.2, 73.6, 67.7, 61.7, 49.4, 44.9, 28.4, 11.0; ^{31}P NMR (D_2O, 125 MHz, decoupled with ^{1}H) δ -8.94 (d, J = 12.0), −10.28 (d, J = 12.0); HRMS (FAB, negative) calcd for $C_{16}H_{20}BrN_4O_{13}P_2$ 616.9686 [$(M - H)^-$], found 616.9716; UV (MeOH) λ_{max} 256, sh 278 nm.

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Supporting Information Available: ^{1}H NMR spectral charts of **6**, **13**, **15**, **16**, **23**, **24**, **26**, **30**, **31**, **32**, **33**, **34**, and **35** and HPLC charts of **3**, **6**, **11**, **12**, **13**, **14**, **15**, **16**, and **32**. This material is available free of charge via the Internet at http://pubs.acs.org.

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