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Synthesis of 7-Deaza-cyclic Adenosine-5′-diphosphate-carbocyclic-ribose and Its 7-Bromo Derivative as Intracellular Ca^{2+}-Mobilizing Agents

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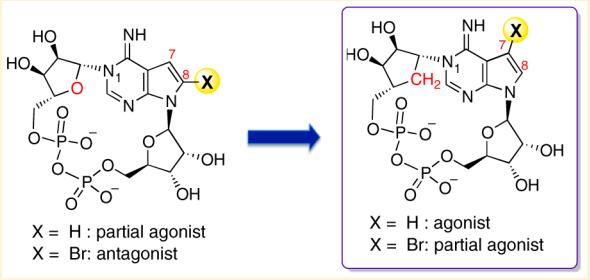
Supporting Information

ABSTRACT: Cyclic ADP-carbocyclic-ribose (cADPcR, **3**) is a biologically and chemically stable equivalent of cyclic ADP-ribose (cADPR, **1**), a Ca^{2+}-mobilizing second messenger. We became interested in the biological activity of the 7-deaza analogues of cADPcR, i.e., 7-deaza-cADPcR (**7**) and its 7-bromo derivative, i.e., 7-deaza-7-Br-cADPcR (**8**), because 7-deazaadenosine is an efficient bioisostere of adenosine. The synthesis of **7** and **8** required us to construct the key *N*1-carbocyclic-ribosyl-7-deazaadenosine structure. Therefore, we developed a general method for preparing N1-substituted 7-deazaadenosines by condensing a 2,3-disubstituted pyrrole nucleoside with amines. Using this method, we prepared the *N*1-carbocyclic ribosyl 7-deazaadenosine derivative **10a**, from which we then synthesized the target 7-deaza-cADPcR (**7**) via an Ag^{+}-promoted intramolecular condensation to construct the 18-membered pyrophosphate ring structure. The corresponding 7-bromo derivative **8**, which was the first analogue of cADPR with a substitution at the 7-position, was similarly synthesized. Biological evaluation for Ca^{2+}-mobilizing activity in the sea urchin egg homogenate system indicated that 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**) acted as a full agonist and a partial agonist, respectively.



INTRODUCTION

Much attention has been focused on 7-deazaadenosine, which is also known as the natural product tubercidin, and its naturally occurring derivatives such as toyocamycin or sangivamycin (Figure 1) due to their remarkable biological activities, particularly as antitumor and antiviral agents.[1] In recent years, 7-deazaadenosine and its derivatives have also been used as base-modified analogues of natural nucleosides in oligonucleotide chemistry and chemical biology studies.[2] The attractive potency of 7-deazaadenosine as an efficient bioisostere of adenosine led to our interest in the biological activity of 7-deaza congeners of cyclic ADP-ribose (cADPR, **1**, Figure 2), a Ca^{2+}-mobilizing second messenger.[3]

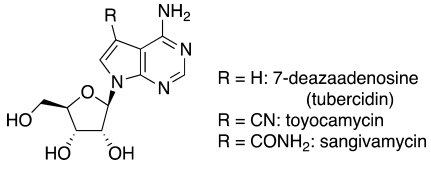


Figure 1. 7-Deazaadenosine (tubercidin) and related naturally occurring compounds.

cADPR mobilizes intracellular Ca^{2+} in various mammalian cells, such as pancreatic β-cells, smooth muscle and cardiac muscle cells, T-lymphocytes, and cerebellar neurons, and therefore cADPR is now considered as a general mediator of intracellular Ca^{2+} signaling.[4] Analogues of cADPR have been extensively designed and synthesized due to their potential usefulness for investigating the mechanism of cADPR-mediated Ca^{2+}release.[5−8] On the basis of the important physiologic roles of cADPR, cADPR analogues are also expected to be lead structures for the development of potential drug candidates.[4,8]

cADPR and its analogues (Figure 2) have been synthesized by enzymatic or chemo-enzymatic methods using ADP-ribosyl cyclase from *Aplysia californica*, which mediates the intramolecular ribosylation of NAD^{+} and some modified NAD^{+} (prepared chemically or enzymatically) at the N1-position of the purine moiety, to yield cADPR or the corresponding analogues.[5] These studies disclosed that some 8-substituted analogues of cADPR, such as 8-NH_2-cADPR (**2**), are antagonists of cADPR at its intracellular receptor,[5a] and these analogues are effective pharmacological tools for studying cADPR-modulated Ca^{2+} signaling pathways.[4]

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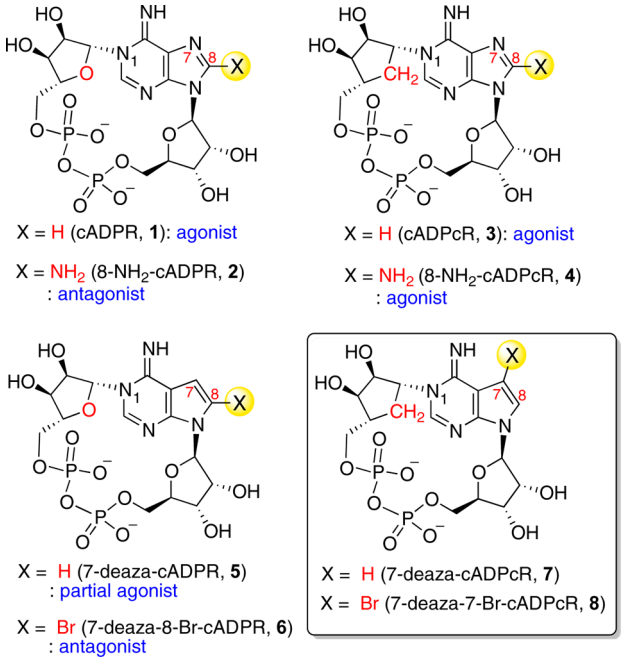

Figure 2. cADPR (**1**) and its analogues **2**–**8**.

Potter and co-workers performed the synthetic studies of 7-deaza-cADPR (**5**) and its analogues by enzymatic methods and identified 7-deaza-cADPR as a partial agonist.[7] They also synthesized its 8-bromo derivative, i.e., 7-deaza-8-Br-cADPR (**6**), and demonstrated that it is an antagonist of cADPR with membrane permeable properties.[8] Thus, the 7-deaza modification appears to enhance the antagonistic function of cADPR because a partial agonist can be an intermediate form between an agonist and antagonist.

However, cADPR is readily hydrolyzed at the unstable *N*1-ribosyl linkage to produce inactive ADP-ribose (ADPR), even in neutral aqueous solutions.[11] This is due to the fact that cADPR is in a zwitterionic form positively charged around the N1–C6–N^6 moiety (pK_a = 8.3), making the molecule unstable, where the charged adenine moiety attached to the anomeric carbon of the N1-linked ribose acts as an efficient leaving group. Under physiological conditions, cADPR is also hydrolyzed at the same N1-ribosyl linkage by cADPR hydrolase to give the inactive ADP-ribose.[11]

We previously designed and synthesized cyclic ADP-carbocyclic-ribose (cADPcR, **3**) as a stable mimic of cADPR, in which the oxygen in the *N*1-ribose ring of cADPR was replaced by a methylene group. cADPcR is both chemically and biologically stable, and effectively mobilizes intracellular Ca^{2+} in sea urchin eggs and neuronal cells.[9c,f,g] On the basis of these findings, we designed and synthesized the 8-modified analogues of cADPcR, e.g., 8-NH$_2$-cADPcR (**4**), expecting that they might be chemically and biologically stable potent cADPR antagonists. To our surprise, however, these analogues acted as agonists rather than antagonists.[9e]

The interesting finding mentioned above on the biological activity of the 7-deaza-cADPR derivatives by Potter and co-workers that the 7-deaza modification enhances the antagonistic function of cADPR led us to synthesize the carbocyclic analogue of 7-deaza-cADPR, i.e., 7-deaza-cADPcR (**7**) and its 7-bromo derivative 7-deaza-7-Br-cADPcR (**8**) as stable analogues of cADPR of biological interest. We focused our efforts on the biological activity of 7-deaza-7-Br-cADPcR because substitution at the 7-position was possible only in the 7-deazaadenine motif and not in the adenine itself, and there have been no 7-substituted cADPR analogues of this type reported to date. Here, we describe the synthesis of 7-deaza-cADPcR (**7**) and 7-Br-7-deaza-cADPcR (**8**) and their Ca^{2+}-mobilizing activity. We also developed the first general method for synthesizing N1-substituted 7-deazaadenosines.

RESULTS AND DISCUSSION

Synthetic Plan. As described above, many cADPR analogues have been synthesized by enzymatic or chemoenzymatic methods with ADP-ribosyl cyclase from *Aplysia californica*, as described above.[5] The analogues obtained by these methods, however, are limited due to the substrate specificity of the enzyme.[10] Therefore, we have developed an efficient chemical method for synthesizing cADPR analogues, in which the key reaction was intramolecular condensation to form the 18-membered pyrophosphate ring by the Ag^+-promoted activation of phenylthiophosphate-type substrates.[9b] This is now a general method for synthesizing these types of biologically important cyclic nucleotides particularly those chemically modified in the *N*-1-linked ribose moiety, which are not expected to be accessible by an enzymatic route.

The retrosynthetic analysis of the target 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**) is shown in Figure 3. To

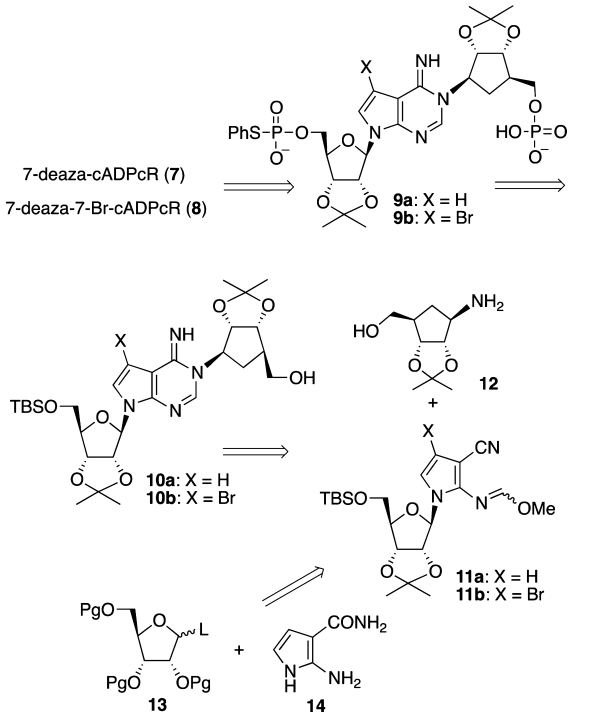

Figure 3. Retrosynthetic analysis of 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**).

synthesize the target compounds, although construction of the 18-membered pyrophosphate structure is an important step, the structure was likely to be constructed using the above-mentioned Ag^+-promoted intramolecular condensation reaction of an *S*-phenyl phosphorothioate-type substrates **9**.[5b,c] Subsequent deprotection of the cyclization product would furnish the desired **7** or **8**. The *S*-phenyl phosphorothioate-type substrates **9** would be obtained from the *N*1-*β*-carbocyclic ribosyl-7-deazaadenosines **10**, which we planned to construct

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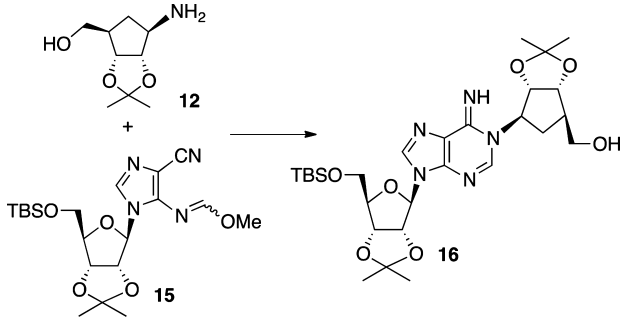
by condensation between a known β-carbocyclicribosylamine **12**[12] and the substituted pyrrole nucleosides **11**. Both of the pyrrole nucleosides **11a** and **11b** were unknown, but **11a** was expected to be prepared from a protected ribose **13** and a pyrrole derivative **14** via β-selective glycosidation. We planned to examine the bromination of the pyrrole nucleoside **11a** at 4-position.

Development of a General Procedure for Synthesizing N1-Substituted 7-Deazaadenosines. Construction of the *N*1-carbcyclic ribosyl-7-deazaadenosine structure was a challenge in the present study, because there are no reported procedures providing this type of nucleoside structure. Synthesis of 7-deazaadenosine and its derivatives has been studied extensively due to their biological importance, and the Lewis acid-promoted glycosidation between a silylated 6-chloro-7-deazaadenine base and a tetra-*O*-acyl ribose, developed by Seela and co-workers, is thought to be the best procedure in terms of synthetic usefulness, i.e., excellent yield and product scope.[13] This method, however, was not applicable to our synthesis of 7-deaza-cADPcR because substitution at the N1-position of 7-deazaadenosine derivatives is difficult. The only example of such an N1-substitution of 7-deazaadenosine derivatives reported to date is the N1-methylation of 7-deazaadenosine by treating 7-deazaadenosine with a large excess of MeI.[14]

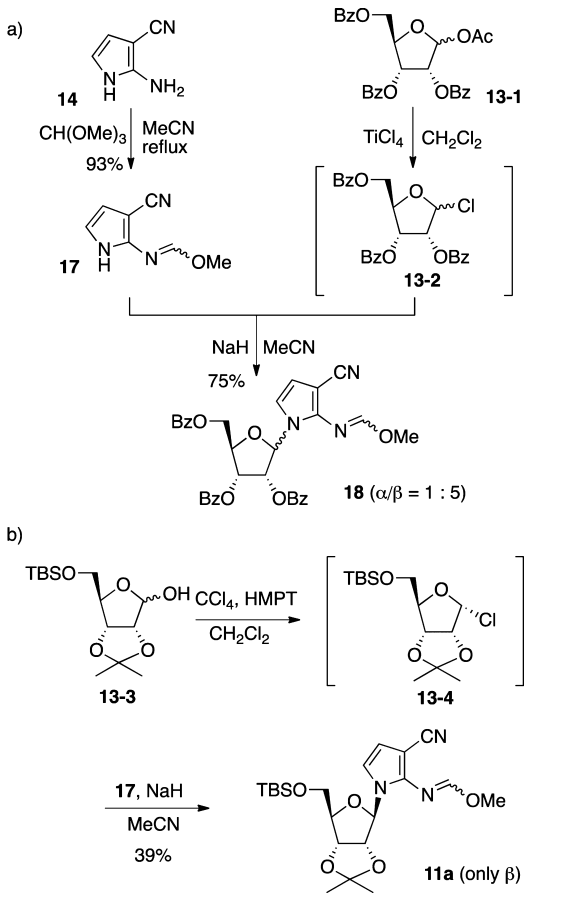
We planned to construct the key N1-carbocyclic ribosyl-7-deazaadenosine structure by condensation between the 2,3-disubstituted pyrrole nucleoside **11** and the carbocyclic ribosylamine **12** (Figure 3). Because condensation between **12** and an imidazole nucleoside **15** effectively produced the *N*1-carbocyclic-ribosyladenosine derivative **16** (Scheme 1),[9c] we

Scheme 1. Construction of the *N*1-Carbocyclic Adenosine Structure with the Imidazole Nucleoside 15 and Carbocyclic Amine 12



expected that similar condensation with the corresponding pyrrole nucleoside **11** instead of the imidazole nucleoside **15** would produce the desired *N*1-carbocyclic ribosyl-7-deazaadenosine derivatives. Thus, the pyrrole nucleoside **11** was needed, but this type of 2-nitrogen substituted 3-cyanopyrrole nucleoside was unknown. Although ribosylation of 2-amino-3-cyanopyrole (**14**), prepared by Townsend's procedure,[15] was examined under the Vorbrüggen glycosidation conditions with tetra-*O*-acyl ribose (**13−1**), the corresponding pyrrole nucleoside was not obtained. We then investigated glycosidation with the corresponding pyrrole anion (Scheme 2). After conversion of **14** into the corresponding methoxyimidate **17**, it was treated with NaH in MeCN in the presence of an anomeric mixture of tri-*O*-benzoyl-1-chlororibose (**13−2**),[16] which was prepared in situ from **13−1** (Scheme 2a). Although the reaction gave

Scheme 2. Synthesis of the 2,3-Disubstituted Pyrrole Nucleosides



pyrrole nucleoside **18**, it was an inseparable anomeric mixture (α/β = 1:5). We examined the reaction under other conditions by changing the solvent and temperature, but the desired pyrrole β-riboside was not obtained in a pure form. When a similar glycosidation was performed with an isopropylidene-protected chlororiboside **13−4**,[17] however, the desired pyrrole β-riboside **11a** was obtained in pure form. Although the yield was not high, **11a** had the 2,3-*O*-isopropylidene and 5-*O*-silyl protecting groups, which were very suitable for further transformation, and therefore, we proceeded to the next step, i.e., condensation of **11a** with the carbocyclic ribosylamine **12**.

We next investigated the bromination of the pyrrole β-riboside **11a**. In the reaction of pyrroles with electrophiles, an electrophile usually reacts selectively at the undesired 2-position (the position adjacent to the nitrogen), Dvarnikova and Trela, however, found that the 2/3-regioselectivity in pyrrole bromination could be controlled by changing the reaction conditions,[18] and we therefore applied these conditions to the pyrrole nucleoside in the present study (Table 1). Thus, consistent with their report, treatment of the pyrrole nucleoside **11a** with only NBS in THF gave 5-bromination product **19** exclusively, while supplementary use of PBr_3 as an additive successfully reversed the regioselectivity to selectively produce the desired 4-bromination product **11b**.

We examined the condensation of **11a** with various amines in our attempt to develop a general method for synthesizing 7-deazaadenosine and its N1-substituted derivatives (Table 2). Treatment of **11a** with NH_4OH in the presence of K_2CO_3 in MeOH successfully afforded the desired protected 7-

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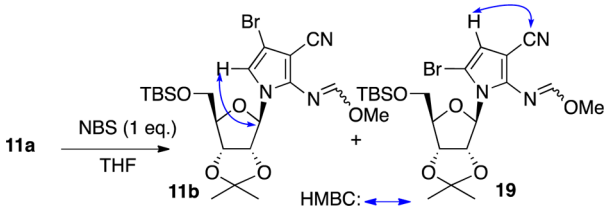
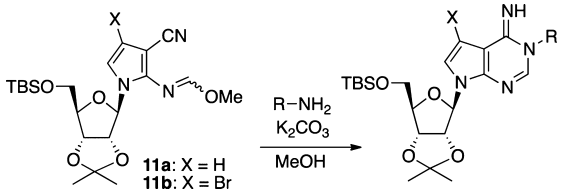
Table 1. Regioselective Bromination of the Pyrrole Nucleoside 11a



entry	additive	temp.	time	yield (**11b**:**19**)[a]
1		−78 °C	3 h	40% (0:1)
2		0 °C	5 min	89% (0:1)
3	PBr_3 (5 mol %)	−78 °C	3 h	72% (2,4:1)
4	PBr_3 (5 mol %)	−78 °C	1 h	85% (3:1)

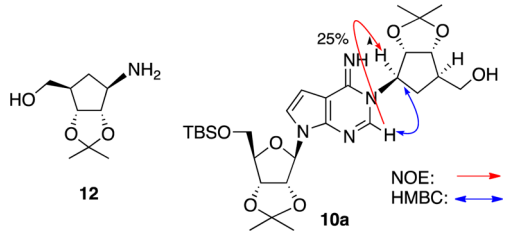
[a]The ratio was determined by ^{1}H NMR.

Table 2. General Method for Preparing N1-Substituted and Non-substituted 7-Deazaadenosines



entry	substrate	R−NH_2	yield
1	**11a**	NH_3[a]	84%
2	**11a**	$MeNH_2$	quant.
3	**11a**	$EtNH_2$	quant.
4	**11a**	allyl amine	57%
5	**11a**	cyclohexyl amine	80%
6	**11a**	**12**	82% (**10a**)
7	**11b**	**12**	79% (**10b**)

[a]Aqueous ammonia (28% in H_2O) was used.



deazaadenosine in 84% yield. Similar treatments with a variety of amines provided the corresponding N1-substituted 7-deazaadenosines, as summarized in Table 2 (entries 2−5). Thus, we successfully developed the first general method for synthesizing N1-substituted 7-deazaadenosine, which is also an alternative synthetic procedure for 7-deazaadenosine (tubercidin). This method can be applied to the synthesis of a variety of N1-substituted 7-deazaadenosines of biological interest.

When the carbocyclic ribosylamine **12**[12] was used as an amine in the condensation with **11a**, the *N*1-carbocyclicribosyl-7-deazaadenine ring was effectively constructed to afford the desired product **10a** in 82% yield, and its N1-carbocyclic structure was confirmed based on its NOE data and HMBC spectrum (Table 2). Similarly, the reaction of the 4-bromopyrrole nucleoside **11b** and **12** also afforded the corresponding 7-brominated product **10b** in 79% yield.

Synthesis of 7-Deaza-cADPcR (7) and 7-Deaza-7-Br-cADPcR (8). We next investigated the conversion of *N*1-*β*-carbocyclic ribosyl-7-deazaadenosine derivatives **10a** and **10b** into *S*-phenyl phosphorothioate-type substrates **9a** and **9b** (Scheme 3). The 5″-hydroxy group of **10a** was protected with a dimethoxytrityl group, and then the 5′-*O*-TBS group of the product was removed with TBAF to give **20a**. Treatment of **20a** with an *S*,*S*′-diphenylphosphorodithioate/2,4,6-triisopropylbenzenesulfonyl chloride/pyridine system,[19] followed by removal of the 5″-*O*-dimethoxytrityl group of the product with aqueous AcOH, gave the corresponding 5′-bis-*S*-(phenyl)-phosphorothioate, which was then treated sequentially with $MeOPOCl_2$/pyridine[20] and with H_3PO_2/Et_3N/triethylammonium acetate.[21] This three-step reaction gave *S*-phenyl phosphorothioate **9a**, which was the substrate for the next intramolecular condensation reaction. The corresponding 7-bromo *S*-phenyl phosphorothioate **9b** was similarly obtained.

With the *S*-phenyl phosphorothioates **9a** and **9b** in hand, we next investigated the Ag^+-promoted intramolecular cyclization reaction. Slow addition of a solution of **9a** or **9b** in pyridine to a mixture of a large excess of $AgNO_3$ and Et_3N in the presence of MS3A in pyridine at room temperature[9b,c] led to the corresponding cyclization products **21a** or **21b** in 48% or 75% yield, respectively. Finally, removing the isopropylidene group of **21a** and **21b** with aqueous HCO_2H produced the target 7-deaza-cADPcR (**7**) and 7-Br-7-deaza-cADPcR (**8**), respectively.

Ca^{2+}-Mobilizing Activity in Sea Urchin Egg Homogenate. We examined the Ca^{2+}-mobilizing ability of 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**) as well as cADPcR (**3**) by fluorometrically monitoring Ca^{2+} with *Hemicentrotus pulcherrimus* sea urchin egg homogenate (Figure 4).[22,23] cADPcR released Ca^{2+} from the homogenate in a concentration-dependent manner with an EC_{50} value of 54 nM.

7-Deaza-cADPcR was a full agonist, similar to cADPcR, but its potency (EC_{50} = 429 nM) was lower than that of cADPcR. 7-Deaza-7-Br-cADPcR, the first cADPR analogue with a substituent at the adenine-7-position, was therefore identified as a weak partial agonist.

It is interesting that 7-deaza-cADPR (**5**) acts as a partial agonist[7] but its carbocyclic congener 7-deaza-cADPcR (**7**) acts as a full agonist. Further, we previously demonstrated that although 8-NH_2-cADPR (**2**) acts as an antagonist, its carbocyclic congener 8-NH_2-cADPcR (**4**) acts as a full agonist.[9e] These findings suggest that replacement of the *N*1-ribose with the carbocyclic-ribose in cADPR and its analogues makes their function more agonistic. Therefore, in cADPR derivatives, the ring oxygen of the *N*1-ribose moiety appears to be essential for showing the antagonistic effect.

In summary, because 7-deazaadenosine is an efficient bioisostere of adenosine, we designed 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**), which were successfully synthesized via an Ag^+-promoted intramolecular condensation to construct the 18-membered pyrophosphate ring structure. We also developed the first general method for preparing N1-substituted 7-deazaadenosines by the condensation of a 2,3-disubstituted pyrrole nucleoside with amines, which was effectively used for the construction of the *N*1-carbcyclic-ribosyl 7-deazaadenosine and 7-bromo-7-deazaadenosine structures, the key structures for the synthesis of the targets compounds. Biological evaluation of the Ca^{2+}-mobilizing activity revealed that 7-deaza-cADPcR (**7**) and 7-deaza-7-Br-cADPcR (**8**) act as a full agonist and a partial agonist, respectively, in the sea urchin egg homogenate system. These findings provide important structural information on the

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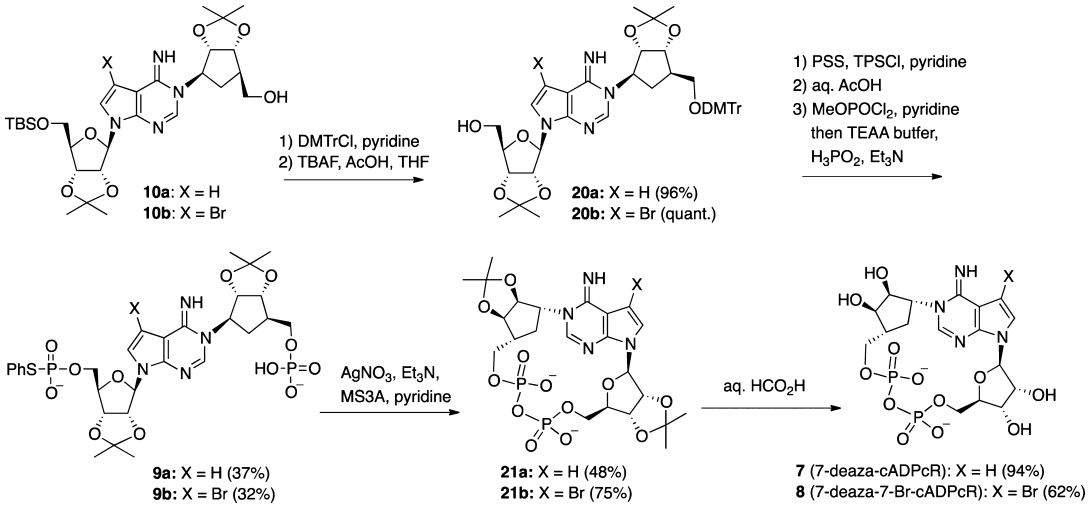
Scheme 3. Synthesis of 7-Deaza-cADPcR (7) and 7-Deaza-7-Br-cADPcR (8)



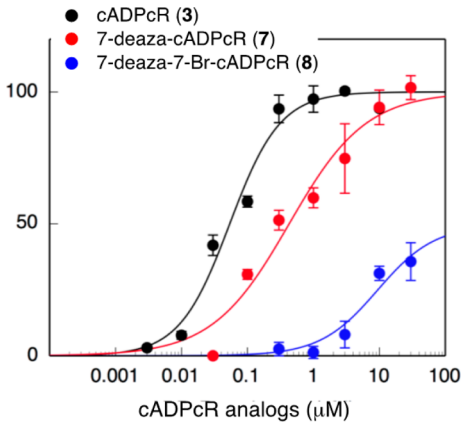


Figure 4. Concentration-dependent Ca^{2+}-mobilizing activity of cADPcR (**3**), 7-deaza-cADPcR (**7**), and 7-deaza-7-Br-cADPcR (**8**) in sea urchin egg homogenate. The Ca^{2+}-mobilizing activity of each compound is expressed as the percent change in the ratio of fura-2 fluorescence (F340/F380) relative to that of 3 μM cADPcR. Data are the mean ± SEM of 3 to 6 experiments.

agonist−antagonist switching of cADPR-related compounds, indicating that the ring oxygen of the *N*1-ribose moiety might be essential for the antagonistic effect.

EXPERIMENTAL SECTION

General Methods and Materials. ^{1}H NMR spectra were recorded in $CDCl_3$ at ambient temperature unless otherwise noted, at 400 or 500 MHz, with TMS as an internal standard. ^{13}C NMR spectra were recorded in $CDCl_3$ at ambient temperature at 100 or 125 MHz. ^{1}H NMR peak assignments were based on H−H COCY spectrum. Silica gel column chromatography was performed with silica gel 60 N (spherical, neutral, 63-210 μm). Flash column chromatography was performed with silica gel 60 N (spherical, neutral, 40−50 μm). Celite 545 was purchased from a chemical supplier. Analytical HPLC was performed with YMC J'sphere ODS-M80 (250 × 4.6 mm), A sol. 5% MeCN in 0.1 M triethylamomonium acetate buffer, and B sol. 80% MeCN in 0.1 M triethylamomonium acetate buffer; B conc. 0−100% (30 min), 1 mL/min.

*3-Cyano-2-(methoxymethyleneamino)pyrrole (**17**).* A solution of **14** (2.30 g, 21.5 mmol) and methyl orthoformate (7.06 mL, 64.5 mmol) in CH_3CN (200 mL) was stirred under reflux for 4 h and then evaporated to give **17** (2.98 g, 93%, brown solid): ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.59 (1 H, br), 8.35 (1 H, s), 6.64 (1 H, d, J = 3.1 Hz), 6.29 (1 H, d, J = 3.1 Hz) 3.82 (3 H, s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 158.9, 143.2, 117.4, 116.0, 109.9, 80.3, 54.0; HRMS (ESI-ion trap, negative) calcd for $C_7H_6N_3O$ 148.05164 [$(M - H)^-$], found 148.05128; mp 75.5−76.0 °C.

*3-Cyano-2-(methoxymethyleneamino)-1-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)pyrrole (**18**).* To a solution of **13−1** (504 mg, 1.0 mmol) in CH_2Cl_2 (16.7 mL), a solution of $TiCl_4$ (109 μL, 1.0 mmol) in CH_2Cl_2 (0.9 mL) was added slowly at room temperature, and the mixture was stirred at the same temperature for 1.5 h. After the addition of H_2O, the resulting mixture was partitioned, and the organic layer was dried (Na_2SO_4) and evaporated to give crude **13−2**. To a solution of **17** (74.6 mg, 0.50 mmol) in CH_3CN (2 mL) was added NaH (60% mineral oil, 28 mg, 0.70 mmol) at 0 °C, and the resulting mixture was stirred at the same temperature for 20 min. To the resulting mixture, a solution of crude **13−2** in CH_3CN (3 mL) was added at 0 °C, and the mixture was stirred at room temperature for 9 h and then evaporated. The residue was partitioned between AcOEt and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by flash column chromatography (silica gel, hexane/AcOEt = 5/1) to give **18** (α/β = 1:5, 219 mg, 75%, white amorphous solid): ^{1}H NMR (500 MHz, $CDCl_3$, α/β mixture) δ 7.99−7.18 (15 H, m), 6.63 (5/6 H, d, J = 3.4 Hz), 6.36 (1/6 H, d, J = 3.4 Hz), 6.26 (1/6 H, d, J = 3.4 Hz), 6.22 (5/6 H, d, J = 4.5 Hz), 6.21 (5/6 H, d, J = 3.4 Hz), 6.20 (1/6 H, d, J = 5.1 Hz), 5.47 (1/6 H, dd, J = 10.2, 5.1 Hz), 5.26 (5/6 H, dd, J = 9.0, 4.5 Hz), 5.08 (5/6 H, dd, J = 9.0, 5.1 Hz), 5.04 (1/6 H, dd, J = 10.2, 2.8 Hz), 4.71−4.68 (5/6 H, m), 4.63−4.60 (1/6 H, m), 4.46−4.43 (5/6 H, m), 4.41−4.39 (2/6 H, m), 4.06−4.03 (1/6 H, m), 3.49 (15/6 H, s), 3.14 (3/6 H, s); ^{13}C NMR (125 MHz, $CDCl_3$, α/β mixture) δ 166.0, 166.0, 165.5, 157.7, 157.0, 143.2, 138.7, 137.1, 134.0, 133.8, 133.3, 130.0, 129.9, 129.7, 129.7, 129.6, 129.4, 129.4, 128.7, 128.6, 128.6, 128.5, 128.4, 128.3, 128.2, 125.9, 125.2, 117.4, 117.2, 117.1, 116.9, 116.1, 115.2, 109.0, 106.5, 105.5, 81.7, 81.0, 80.2, 78.2, 73.1, 72.9, 63.0, 62.4, 60.4, 54.0, 53.6, 53.5, 29.7, 21.1, 14.2; HRMS (ESI-ion trap, positive) calcd for $C_{33}H_{27}N_3O_8Na$ 616.16904 [$(M + Na)^+$], found 616.16951

*3-Cyano-2-(methoxymethyleneamino)-1-[5-O-(tert-butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]pyrrole (**11a**).* To a solution of **13−3** (3.04 g, 10.0 mmol) and CCl_4 (1.16 mL, 12.0 mmol) in toluene (20 mL) was added HMPT (2.09 mL, 11.5 mmol) over 20 min at −15 °C, and then, the mixture was stirred at the same temperature for 1 h. After the addition of brine, the mixture was partitioned, and the organic layer was dried (Na_2SO_4) and evaporated to give crude **13−4**. To a solution of **17** (745 mg, 5.0 mmol) in CH_3CN (10 mL) was added NaH (60% mineral oil, 240 mg, 6.0 mmol) at 0 °C, and the resulting mixture was stirred at the same temperature for 1 h. A solution of the crude **13−4** in CH_3CN (30 mL) was added to the mixture at 0 °C, and the resulting mixture was stirred

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at room temperature for 24 h and then evaporated. The residue was partitioned between AcOEt and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by flash column chromatography (silica gel, hexane/AcOEt = 6/1) to give **11a** (849 mg, 39%, yellow oil): ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.43 (1 H, s), 6.78 (1 H, d, J = 3.2 Hz), 6.28 (1 H, d, J = 3.2 Hz), 6.03 (1 H, d, J = 3.6 Hz), 4.81 (1 H, dd, J = 5.8, 3.2 Hz), 4.68 (1 H, dd, J = 5.8, 3.6 Hz), 4.27−4.25 (1 H, m), 3.92 (1 H, s), 3.87−3.77 (2 H, m), 1.58, 1.35 (each 3 H, each s), 0.91 (9 H, s), 0.09, 0.08 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 158.7, 142.5, 117.6, 115.4, 114.0, 110.7, 89.9, 85.6, 85.4, 80.6, 79.0, 63.4, 54.3, 27.5, 26.0, 25.6, 18.5; HRMS (ESI-ion trap, positive) calcd for $C_{21}H_{33}N_3O_5NaSi$ 458.20817 [(M + Na)$^+$], found 458.20849.

General Procedure of the Bromination of Pyrrole Nucleoside 11a. To a solution of **11a** (1 equiv) in THF (0.1 M) was added NBS (1 equiv) at −78 or 0 °C, then the mixture was stirred at the same temperature for 3 h or 5 min. After the addition of aqueous saturated $NaHCO_3$, the resulting mixture was partitioned between AcOEt and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel).

General Procedure of the Bromination of Pyrrole Nucleoside 11a Using PBr_3. To a solution of **11a** (54 mg, 0.12 mol) in THF (1.5 mL) was added NBS (1 equiv) and BBr_3 (5 mo%) at −78 °C, then the mixture was stirred at the same temperature for 3 or 1 h. After the addition of Et_3N, the resulting mixture was partitioned between AcOEt and H_2O, and the organic layer was washed with aqueous saturated $NaHCO_3$ and brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel).

*4-Bromo-3-cyano-2-(methoxymethyleneamino)-1-[5-O-(tert-butyldimethylsilyl)-2,3-O-isopropylidene-β-D-ribofuranosyl]pyrrole (**11b**, Table 1/Entry 4).* To a solution of **11a** (54 mg, 0.12 mmol) in THF (1.5 mL) was added NBS (22 mg, 0.12 mmol) and PBr_3 (10 μL, 0.06 mmol) at −78 °C, then the mixture was stirred at the same temperature for 1 h. After the addition of Et_3N, the resulting mixture was partitioned between AcOEt and H_2O, and the organic layer was washed with aqueous saturated $NaHCO_3$ and brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel, hexane/AcOEt = 6/1) to give **11b** (34 mg, 63%, yellow oil) along with **19** (12 mg, 21%, yellow oil): ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.43 (1 H, s, N=C<u>H</u>), 6.94 (1 H, s, H-5), 6.02 (1 H, d, J = 3.1 Hz, H-1′), 4.77 (1 H, dd, J = 5.8, 2.2 Hz, H-3′), 4.63 (1 H, dd, J = 5.8, 3.1 Hz, H-2′), 4.32 (1 H, d, J = 2.2 Hz, H-4′), 3.92 (3 H, s, OC<u>H</u>$_3$), 3.81 (1 H, dd, J = 11.3, 2.7 Hz, H-5′a), 3.78 (1 H, dd, J = 11.3, 2.7 Hz, H-5′b), 1.57, 1.34 (each 3 H, each s, isopropyl C<u>H</u>$_3$), 0.92 (9 H, s, *tert*-butyl), 0.12, 0.12 (each 3 H, each s, dimethyl); ^{13}C NMR (125 MHz, $CDCl_3$) δ 159.1, 142.3, 115.5, 115.2, 113.7, 97.9, 90.4, 85.9, 85.7, 82.6, 80.6, 63.4, 54.4, 27.4, 25.9, 25.9, 25.8, 25.4, 18.4, 6.4; HRMS (ESI-ion trap, positive) calcd for $C_{21}H_{32}N_3O_5BrNaSi$ 536.11868 [(M + Na)$^+$], found 536.11902.

5-Bromo-3-cyano-2-(methoxymethyleneamino)-1-[5-O-(tert-butyldimethylsilyl)-2,3-O-isopropylidene-β-D-ribofuranosyl]pyrrole (19, Table 1/Entry 2). To a solution of **11a** (54 mg, 0.12 mol) in THF (1.5 mL) was added NBS (22 mg, 0.12 mmol) at −0 °C, then the mixture was stirred at the same temperature for 5 min. After the addition of Et_3N, the resulting mixture was partitioned between AcOEt and H_2O, and the organic layer was washed with aqueous saturated $NaHCO_3$ and brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel, hexane/AcOEt = 6/1) to give **19** (57 mg, 89%, yellow oil): ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.32 (1 H, s, N=C<u>H</u>), 6.35 (1 H, s, H-4), 6.07 (1 H, d, J = 3.6 Hz, H-1′), 5.29 (1 H, dd, J = 6.7, 3.6 Hz, H-2′), 4.79 (1 H, dd, J = 6.7, 4.9 Hz, H-3′), 3.99−3.95 (2 H, m, H-4′, H-5′a), 3.81 (1 H, dd, J = 5.8, 2.7 Hz, H-5′b), 1.58, 1.36 (each 3 H, each s, isopropyl C<u>H</u>$_3$), 0.89 (9 H, s, *tert*-butyl), 0.07, 0.06 (each 3 H, each s, dimethyl); ^{13}C NMR (125 MHz, $CDCl_3$) δ 159.5, 144.6, 116.0, 115.2, 113.1, 99.3, 89.7, 85.1, 82.8, 80.4, 63.1, 54.8, 29.5, 27.4, 25.9, 25.5, 18.5, 6.4; HRMS (ESI-ion trap, positive) calcd for $C_{21}H_{32}N_3O_5BrNaSi$ 536.11868 [(M + Na)$^+$], found 536.11874.

General Procedure for the Synthesis of 7-Deazaadenosines (Table 2). A mixture of **11a** or **11b** (0.10 mmol), an amine (entries 1−5, 5 equiv; entries 6 and 7, 2.5 equiv), and K_2CO_3 (0.15 equiv) in MeOH (0.5 mL, entries 1−6) or MeOH/THF (1:1, 0.5 mL, entry 7) was stirred at room temperature for 20 h and then evaporated. The residue was partitioned between $CHCl_3$ and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel, hexane/AcOEt = 4/1 to 1/1).

9-[5-O-(tert-Butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]-7-deazaadenosine (Entry 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.33 (1 H, s), 7.23 (1 H, d, J = 4.0 Hz), 6.37 (1 H, d, J = 4.0 Hz), 6.34 (1 H, d, J = 2.7 Hz), 5.12−5.10 (3 H, m), 4.97 (1 H, dd, J = 6.2, 3.6 Hz), 4.30−4.29 (1 H, m), 3.87 (1 H, dd, J = 10.7, 3.6 Hz), 3.78 (1 H, dd, J = 10.7, 4.1 Hz), 1.63, 1.38 (each 3 H, each s), 0.90 (9 H, s), 0.05, 0.04 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 156.5, 152.1, 150.5, 122.8, 114.1, 103.7, 98.6, 90.2, 85.9, 84.8, 80.9, 63.3, 27.3, 25.9, 25.5, 18.4, 6.4; HRMS (ESI-ion trap, positive) calcd for $C_{20}H_{33}N_4O_4Si$ 421.22656 [(M + H)$^+$], found 421.22690; UV (MeOH) λ_{max} = 273 nm.

9-[5-O-(tert-Butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]-1-methyl-7-deazaadenosine (Entry 2). ^{1}H NMR (500 MHz, CD_3OD) δ 7.62 (1 H, s, H-2), 7.00 (1 H, d, J = 3.6 Hz, H-8), 6.44 (1 H, d, J = 3.6 Hz, H-7), 6.19 (1 H, d, J = 3.1 Hz, H-1′), 4.95 (1 H, dd, J = 6.7, 3.1 Hz, H-2′), 4.90 (1 H, dd, J = 6.7 Hz, 2.7 Hz, H-3′), 4.27−4.24 (1 H, m, H-4′), 3.84−3.78 (2 H, m, H-5′a, H-5′b), 3.52 (1 H, s, N−C<u>H</u>$_3$), 1.61, 1.37 (each 3 H, each s, isopropyl C<u>H</u>$_3$), 0.91 (9 H, s, *tert*-butyl), 0.07, 0.06 (each 3 H, each s, dimethyl); ^{13}C NMR (125 MHz, $CDCl_3$) δ 156.4, 145.8, 142.5, 119.9, 114.1, 107.0, 102.0, 89.8, 85.7, 85.1, 80.8, 63.3, 35.4, 27.3, 25.9, 25.5, 18.4; HRMS (ESI-ion trap, positive) calcd for $C_{21}H_{35}N_4O_4Si$ 435.24221 [(M + H)$^+$], found 435.24207; UV (MeOH) λ_{max} = 273 nm.

9-[5-O-(tert-Butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]-1-ethyl-7-deazaadenosine (Entry 3). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.62 (1 H, s), 6.99 (1 H, d, J = 3.4 Hz), 6.43 (1 H, d, J = 3.4 Hz), 6.18 (1 H, d, J = 2.9 Hz), 4.97 (1 H, dd, J = 6.2, 2.8 Hz), 4.91 (1 H, dd, J = 6.2, 3.4 Hz), 4.26−4.24 (1 H, m), 4.06 (2 H, q, J = 6.8 Hz), 3.84−3.76 (2 H, m), 1.62, (3 H, s), 1.38 (3 H, t, J = 6.8 Hz), 1.37 (3 H, s), 0.91 (9 H, s), 0.07, 0.06 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 155.1, 145.4, 142.3, 119.8, 114.1, 107.3, 102.1, 89.9, 85.7, 85.0, 63.3, 42.5, 27.3, 25.9, 25.5, 18.4, 14.5; HRMS (ESI-ion trap, positive) calcd for $C_{22}H_{37}N_4O_4Si$ 449.25786 [(M + H)$^+$], found 449.25772; UV (MeOH) λ_{max} = 273 nm.

9-[5-O-(tert-Butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]-1-allyl-7-deazaadenosine (Entry 4). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.56 (1 H, s), 6.98 (1 H, d, J = 3.6 Hz), 6.53 (1 H, d, J = 3.6 Hz), 6.14 (1 H, d, J = 3.1 Hz), 6.00−5.91 (1 H, m), 5.20−5.14 (2 H, m), 4.90 (1 H, dd, J = 6.2, 2.6 Hz), 4.83 (1 H, dd, J = 6.2), 4.66−4.64 (2 H, m), 4.22−4.19 (1 H, m), 3.79−3.69 (2 H, m), 1.55, 1.30 (each 3 H, each s), 0.83 (9 H, s), −0.01, −0.07 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 154.9, 145.3, 142.5, 132.2, 120.5, 117.7, 114.1, 106.7, 102.7, 89.9, 85.8, 85.1, 80.8, 63.3, 49.4, 27.3, 25.9, 25.5, 18.4; HRMS (ESI-ion trap, positive) calcd for $C_{23}H_{37}N_4O_4Si$ 461.25786 [(M + H)$^+$], found 461.25797; UV (MeOH) λ_{max} = 273 nm.

9-[5-O-(tert-Butyldimethylsilyl)-2,3-O-(isopropylidene)-β-D-ribofuranosyl]-1-cyclohexyl-7-deazaadenosine (Entry 5). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.74 (1 H, s), 6.99 (1 H, d, J = 3.6 Hz), 6.43 (1 H, d, J = 3.6 Hz), 6.18 (1 H, d, J = 3.2 Hz), 5.00−4.98 (2 H, m), 4.91 (1 H, dd, J = 6.4, 2.8 Hz), 4.28−4.26 (1 H, m), 3.85−3.76 (2 H, m), 2.07−2.05 (2 H, m), 1.90−1.89 (2 H, m), 1.79−1.76 (1 H, m), 1.62 (3 H, s), 1.57−1.49 (4 H, m), 1.38 (3 H, s), 1.25−1.22 (1 H, m), 0.91 (9 H, s), 0.07, 0.06 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 155.6, 142.9, 141.7, 119.7, 114.0, 106.9, 102.2, 89.9, 85.7, 85.0, 80.8, 63.3, 52.6, 33.0, 33.0, 27.3, 26.0, 25.9, 25.5, 25.5, 18.4, 6.4, −5.4, −5.5; HRMS (ESI-ion trap, positive) calcd for $C_{26}H_{43}N_4O_4Si$ 503.30481 [(M + H)$^+$], found 503.30478; UV (MeOH) λ_{max} = 273 nm.

*Compound **10a** (Entry 6).* ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.61 (1 H, s, H-2), 7.04 (1 H, d, J = 3.6 Hz, H-8), 6.45 (1 H, d, J = 3.6 Hz, H-7), 6.16 (1 H, d, J = 3.1 Hz, H-1′), 5.34 (1 H, dd, J = 10.4, 5.4 Hz, H-2″), 4.93 (1 H, dd, J = 6.2, 2.7 Hz, H-3′), 4.89 (1 H, dd, J = 6.2, 3.6

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Hz, H-2′), 4.77 (1 H, dd, J = 10.4, 5.8 Hz, H- 3″), 4.50 (1 H, m, H-1″), 4.27 (1 H, dd, J = 6.6, 3.6 Hz), 3.85−3.71 (4 H, m, H-5′a, H-5′b, H-5″a, H-5″b), 2.58−2.49 (3 H, m, H-4″, H-6″a, H-6″b), 1.61, 1.57, 1.37, 1.32 (each 3 H, each s, isopropyl C$\underline{H}_3$), 0.90 (9 H, s, *tert*-butyl), 0.07, −0.06 (each 3 H, each s, dimethyl); ^{13}C NMR (125 MHz, $CDCl_3$) δ 154.5, 145.9, 141.9, 120.7, 114.1, 111.5, 107.1, 101.9, 90.0, 85.7, 85.1, 83.8, 82.4, 80.8, 70.8, 64.7, 63.3, 44.7, 30.4, 28.1, 27.3, 25.9, 25.4, 25.3, 18.3; HRMS (ESI-ion trap, positive) calcd for $C_{29}H_{47}N_4O_7Si$ 591.3208 [(M + H)$^+$], found 591.3215; UV (MeOH) λ_{max} = 273 nm.

*Compound **10b** (Entry 7).* ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.56 (1 H, s, H-2), 7.10 (1 H, s, H-8), 6.16 (1 H, d, J = 2.8 Hz, H-1′), 5.34 (1 H, dd, J = 10.8, 5.6 Hz, H-2″), 4.85 (1 H, dd, J = 5.6, 2.8 Hz, H-2′), 4.80 (1 H, dd, J = 5.6, 3.4 Hz, H-3′), 4.77 (1 H, dd, J = 5.7, 2.8 Hz, H-3″), 4.47−4.43 (1 H, m, H-1″), 4.30 (1 H, dd, J = 5.6, 2.8 Hz, H-4′), 3.88−3.71 (4 H, m, H-5′a, H-5′b, H-5″a, H-5″b), 2.63−2.58 (2 H, m, H-6″a, H-6″b), 2.43−2.41 (1 H, m, H-4″), 1.60, 1.55, 1.35, 1.31 (each 3 H, each s, isopropyl C$\underline{H}_3$), 0.92 (9 H, s, *tert*-butyl), 0.10, 0.10 (each 3 H, each s, dimethyl); ^{13}C NMR (125 MHz, $CDCl_3$) δ 153.8, 147.0, 142.0, 119.4, 114.0, 111.5, 103.6, 91.4, 90.0, 85.9, 85.6, 83.6, 82.4, 80.7, 70.4, 64.8, 63.4, 45.0, 30.4, 28.0, 27.3, 25.9, 25.8, 25.4, 25.3, 18.4; HRMS (ESI-ion trap, positive) calcd for $C_{29}H_{46}N_4O_7BrSi$ 669.23137 [(M + H)$^+$], found 669.23124; UV (MeOH) λ_{max} = 277 nm.

*N-1-[(1R,2S,3R,4R)-2,3-(Isopropylidenedioxy)-4-[(5-dimethoxytrityl)oxymethyl]cyclopentyl]-5′-O-(tert-butyldimethylsilyl)-2′,3′-O-isopropylidene-7-deazadenosine (**20a**).* A solution of **10a** (336 mg, 0.570 mmol) and DMTrCl (290 mg, 0.855 mmol) in pyridine (3.8 mL) was stirred at room temperature for 11 h. After the addition of MeOH, the resulting mixture was partitioned between AcOEt and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel, hexane/AcOEt = 1/3) to give the 5″-O-DMTr product (491 mg, 96%, pale yellow amorphous solid). A solution of the product (486 mg, 0.540 mmol), TBAF (1.0 M in THF, 1.63 mL, 1.63 mmol), and AcOH (30 μL, 0.52 mmol) in THF (5.4 mL) was stirred at room temperature for 2.5 h and then evaporated. The residue was purified by column chromatography (silica gel, $CHCl_3$/MeOH = 1/0 to 9/1) to give **20a** (420 mg, quant., white amorphous solid): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.64 (1 H, s), 7.43−6.81 (14 H, m), 6.42 (1 H, d, J = 4.1 Hz), 5.08−5.03 (3 H, m), 4.98−4.96 (1 H, m), 4.52−4.48 (1 H, m), 4.43−4.41 (1 H, m), 3.96−3.93 (1 H, m), 3.78 (3 H, s), 3.78 (3 H, s), 3.76−3.74 (1 H, m), 3.34−3.32 (1 H, m), 3.15−3.12 (1 H, m), 2.45−2.41 (2 H, m), 2.30−2.27 (1 H, m), 1.61, 1.53, 1.35, 1.26 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 158.3, 154.8, 145.0, 144.0, 139.8, 136.2, 136.1, 130.0, 130.0, 128.1, 127.7, 126.7, 123.1, 114.0, 113.4, 113.0, 109.1, 101.7, 95.5, 85.8, 85.2, 83.7, 83.0, 81.5, 81.2, 64.2, 63.2, 63.0, 55.1, 44.7, 34.0, 27.6, 27.5, 25.2; HRMS (ESI-ion trap, positive) calcd for $C_{44}H_{51}N_4O_9$ 779.3650 [(M + H)$^+$], found 779.3648; UV (MeOH) λ_{max} = 273 nm.

*N-1-[(1R,2S,3R,4R)-2,3-(Isopropylidenedioxy)-4-[(5-dimethoxytrityl)oxymethyl]cyclopentyl]-5′-O-(tert-butyldimethylsilyl)-2′,3′-O-isopropylidene-7-bromo-7-deazadenosine (**20b**).* Title compound **20b** (209 mg, quant., white amorphous solid) was prepared from **10b** (163 mg, 0.244 mmol) according to the procedure described for the preparation of **20a**: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.61 (1 H, s), 7.61−7.19 (9 H, m), 6.83−6.81 (5 H, m), 5.59 (1 H, d, J = 4.8 Hz), 5.04−5.00 (4 H, m), 4.53−4.50 (1 H, m), 4.42−4.40 (1 H, m), 3.90 (1 H, dd, J = 12.6, 1.8 Hz), 3.78 (3 H, s), 3.78 (3 H, s), 3.76 (1 H, d, J = 12.6, 2.2 Hz), 3.32 (1 H, dd, J = 9.4, 4.5 Hz), 3.3 (1 H, dd, J = 9.4, 5.8 Hz), 2.43−2.38 (3 H, m), 1.60, 1.52, 1.35, 1.27 (each 3 H, each s); ^{13}C NMR (125 MHz, $CDCl_3$) δ 158.3, 153.7, 145.6, 145.0, 139.3, 136.2, 136.2, 130.3, 128.1, 127.7, 126.6, 121.9, 114.1, 113.2, 113.0, 105.7, 95.5, 90.9, 85.8, 85.3, 83.6, 82.8, 81.6, 81.1, 64.1, 63.1, 55.2, 44.9, 33.7, 27.7, 27.5, 25.3, 25.2; HRMS (ESI-ion trap, positive) calcd for $C_{44}H_{50}N_4O_9Br$ 857.2755 [(M + H)$^+$], found 857.27577; UV (MeOH) λ_{max} = 277 nm.

*N-1-[(1R,2S,3R,4R)-2,3-(Isopropylidenedioxy)-4-(phosphonoxymethyl)cyclopentyl]-5′-O- [(phenylthio)phosphoryl]-2′,3′-O-isopropylidene-7-deazaadenosine (**9a**).* A solution of PSS (402 mg, 1.05 mmol) and TPSCl (318 mg, 1.05 mmol) in pyridine (2.5 mL) was added to **20a** (274 mg, 0.352 mmol) at 0 °C, and the mixture was stirred at the same temperature for 30 min and then at room temperature for 10.5 h. After the addition of MeOH, the resulting mixture was evaporated and partitioned between AcOEt and H_2O, and the organic layer was washed with brine, dried (Na_2SO_4), and evaporated. The residue was purified by column chromatography (silica gel, $CHCl_3$/MeOH = 1/0 to 9/1) to give the phosphorothioate product (colorless amorphous solid). A solution of the product in aqueous 60% AcOH (2.8 mL) was stirred at room temperature for 20 min, evaporated, and azotropically dried with MeOH. The residue was purified by column chromatography (silica gel, $CHCl_3$/MeOH = 1/0 to 4/1) to give the 5″-*O*-DMTr-removed product (white amorphous solid). A solution of $MeOPOCl_2$ (63 μL, 0.63 mmol) in pyridine (0.4 mL) was stirred at −30 °C for 20 min. To the solution was added a solution of the 5″-*O*-DMTr-removed product in pyridine (2.2 mL), and the mixture was stirred at the same temperature for 3 h. To the resulting solution was added triethylammonium acetate (TEAA) buffer (2.0 M, pH 7.0, 2 mL) then H_3PO_2 (213 μL, 4.2 mmol) and Et_3N (293 μL, 2.1 mmol), and the mixture was stirred at room temperature for 3.5 h and then evaporated. The residue was purified by column chromatography (ODS, CH_3CN/H_2O = 1/1). The product was lyophilized to give **9a** (108 mg, 37% for 3 steps, white powder) as a triethylammonium salt: ^{1}H NMR (500 MHz, D_2O) δ 8.28 (1 H, s), 7.30 (1 H, d, J = 3.4 Hz), 7.10−6.95 (5 H, m), 6.59 (1 H, d, J = 3.4 Hz), 6.20 (1 H, d, J = 2.3 Hz), 5.18 (1 H, dd, J = 6.3, 2.3 Hz), 4.87 (1 H, dd, J = 6.3, 5.7 Hz), 4.71−4.68 (3 H, m), 4.45 (1 H, m), 4.02−3.95 (2 H, m), 3.88−3.86 (2 H, m), 3.03 (6 H, q, J = 7.4 Hz), 2.49−2.38 (2 H, m), 2.29−2.21 (1 H, m), 1.48, 1.46, 1.24, 1.22 (each 3 H, each s), 1.10 (9 H, t, J = 7.4 Hz); ^{13}C NMR (125 MHz, D_2O) δ 152.6, 145.8, 142.4, 133.1, 130.0, 129.5, 128.2, 127.3, 116.0, 115.3, 104.1, 103.3, 91.2, 85.8, 84.4, 84.1, 81.8, 81.1, 66.5, 65.8, 64.8, 47.2, 43.9, 33.1, 26.5, 24.9, 24.5, 8.6; ^{31}P NMR (202 MHz, D_2O) δ 17.62, 0.94; HRMS (ESI-ion trap, negative) calcd for $C_{29}H_{37}N_4O_{12}P_2S$ 727.16094 [(M − H)$^-$], found 727.16397; UV (H_2O) λ_{max} = 273 nm; HPLC purity; column A, retention time 13.75 min, 90.2%.

*N-1-[(1R,2S,3R,4R)-2,3-(Isopropylidenedioxy)-4-(phosphonoxymethyl)cyclopentyl]-5′-O-[(phenylthio)phosphoryl]-2′,3′-O-isopropylidene-7-bromo-7-deazaadenosine (**9b**).* Title compound **9b** (22 mg, 32% for 3 steps, white powder) was prepared from **20b** (64 mg, 0.074 mmol) according to the procedure described for the synthesis of **9a**: ^{1}H NMR (400 MHz, D_2O) δ 8.30 (1 H, s), 7.45 (1 H, s), 7.04−6.88 (5 H, m), 6.14 (1 H, d, J = 2.7 Hz), 5.08 (1 H, dd, J = 6.3, 2.7 Hz), 4.70 (1 H, dd, J = 13.5, 6.3 Hz), 4.69−4.50 (3 H, m), 4.45−4.43 (1 H, m), 4.02 (1 H, dd, J = 10.2, 5.4 Hz), 3.93−3.82 (3 H, m), 3.00 (6 H, q, J = 7.2 Hz), 2.46−2.39 (2 H, m), 2.33−2.26 (1 H, m), 1.45, 1.44, 1.21, 1.21 (each 3 H, each s), 1.07 (9 H, t, J = 7.2 Hz); ^{13}C NMR (125 MHz, D_2O) δ 135.7, 128.6, 126.6, 115.9, 113.5, 113.4, 112.7, 111.2, 110.5, 99.5, 98.6, 85.1, 74.8, 74.3, 69.5, 67.9, 67.2, 65.1, 64.4, 49.6, 48.9, 48.5, 30.5, 27.1, 16.3, 9.8, 8.1, 7.8; ^{31}P NMR (202 MHz, D_2O) δ 17.44, 1.21; HRMS (ESI-ion trap, negative) calcd for $C_{29}H_{36}N_4O_{12}BrP_2S$ 805.07145 [(M − H)$^-$], found 805.07145; UV (H_2O) λ_{max} = 277 nm.

*7-Deaza-cyclic ADP-Carbocyclic-ribose Diacetonaide (**21a**).* To a mixture of $AgNO_3$ (96 mg, 57 μmol), Et_3N (79 μL, 57 μmol), and MS 3A (powder, 1.0 g) in pyridine (20 mL), a solution of **9a** (22 mg, 27 μmol) in pyridine (18 mL) was added slowly over 15 h, using a syringe-pump, at room temperature under stirring in the dark. To the mixture was added TEAA buffer (2.0 M, pH 7.0, 2 mL), and the resulting mixture was filtered with Celite, and the filtrate was evaporated. The residue was partitioned between AcOEt and H_2O, and the aqueous layer was evaporated. The residue was purified by column chromatography (ODS, 0−40% CH_3CN/0.1 M TEAA buffer (0.1 M, pH 7.0, 400 mL), linear gradient). The excess TEAA included in the residue was removed by column chromatography (ODS, CH_3CN/H_2O = 1/1). The product was lyophilized to give **21a** (9 mg, 48%, white powder) as a triethylammonium salt: ^{1}H NMR (500 MHz, D_2O) δ 8.49 (1 H, s), 7.30 (1 H, d, J = 4.0 Hz), 6.71 (1 H, d, J = 4.0 Hz), 6.05 (1 H, d, J = 1.7 Hz), 5.61 (1 H, dd, J = 6.2, 1.7 Hz), 5.32 (1 H, dd, J = 6.2, 3.4 Hz), 4.68−4.65 (3 H, m), 4.37 (1 H, m), 4.05−3.96 (2 H, m), 3.86−3.82 (2 H, m), 3.03 (6 H, q, J = 7.4 Hz), 2.95−2.94 (1

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H, m), 2.75−2.62 (2 H, m), 1.48, 1.47, 1.28, 1.26 (each 3 H, each s), 1.11 (9 H, t, J = 7.4 Hz); ^{13}C NMR (125 MHz, D_2O) δ 162.5, 155.9, 151.7, 140.1, 125.1, 123.1, 114.9, 112.1, 104.0, 97.9, 96.5, 95.0, 94.6, 92.1, 79.3, 77.1, 75.2, 57.3, 54.4, 39.0, 36.8, 35.1, 18.9; ^{31}P NMR (202 MHz, D_2O) δ −10.37 (d), −11.08 (d); HRMS (ESI-ion trap, negative) calcd for $C_{23}H_{31}N_4O_{12}P_2$ 617.14192 [(M − H)$^-$], found 617.14292; UV (H_2O) λ_{max} = 273 nm.

*7-Bromo-7-deaza-cyclic ADP-Carbocyclic-ribose Diacetonaide (**21b**).* Title compound **21b** (10 mg, 75%, brown powder) was prepared from **9b** (15 mg, 0.017 mmol) according to the procedure described for the synthesis of **21a**: ^{1}H NMR (500 MHz, D_2O) δ 8.52 (1 H, s), 7.43 (1 H, s), 5.98 (1 H, d, J = 1.7 Hz), 5.57 (1 H, dd, J = 5.7, 1.7 Hz), 5.28 (1 H, dd, J = 5.7, 2.8 Hz), 4.67−4.61 (3 H, m), 4.37−4.34 (1 H, m), 3.96−3.95 (1 H, m), 3.94−3.92 (1 H, m), 3.89−3.85 (1 H, m), 3.81−3.77 (1 H, m), 3.01 (6 H, q, J = 7.4 Hz), 2.95−2.92 (1 H, m), 2.74−2.72 (1 H, m), 2.61−2.58 (1 H, m) 1.46, 1.45, 1.26, 1.24 (each 3 H), 1.09 (9 H, t, J = 7.4 Hz); ^{13}C NMR (125 MHz, D_2O) δ 152.5, 145.4, 142.6, 129.5, 115.1, 113.0, 102.8, 93.8, 89.5, 87.6, 86.7, 85.1, 84.4, 81.9, 69.4, 67.2, 65.0, 47.2, 44.4, 28.9. 26.7, 24.9, 24.7, 8.8; ^{31}P NMR (202 MHz, D_2O) δ −10.46 (d), −11.00 (d); HRMS (ESI-ion trap, negative) calcd for $C_{23}H_{30}N_4O_{12}BrP_2$ 695.05243 [(M − H)$^-$], found 695.05343; UV (H_2O) λ_{max} = 277 nm; HPLC purity; column A, retention time 13.63 min, 98.4%.

*7-Deaza-cyclic ADP-Carbocyclic-ribose (**7**).* A solution of **21a** (6.6 mg, 9.1 μmol) in aqueous 60% HCO_2H (1.0 mL) was stirred at room temperature for 4 h and then evaporated. After coevaporation with H_2O, the residue was purified by column chromatography (ODS, H_2O). The eluent was evaporated and lyophilized to give **7** (5.5 mg, 94%, white powder) as a triethylammonium salt: ^{1}H NMR (500 MHz, D_2O) δ 8.32 (1 H, s, H-2), 7.27 (1 H, d, J = 3.6 Hz, H-8), 6.66 (1 H, d, J = 3.6 Hz, H-7), 5.72 (1 H, d, J = 6.7 Hz, H-1′), 5.09 (1 H, dd, J = 6.2, 4.9 Hz, H-2′), 4.46−4.42 (2 H, m, H-3′, H-5′a), 4.22−4.21 (2 H, m, H-2″, H-4′), 4.08−4.04 (3 H, m, H-3″, H-5″a, H-5″b), 3.98−3.95 (1 H, m, H-5′b), 3.03 (6 H, q, J = 7.4 Hz, $(CH_3C\underline{H}_2)_3N$), 2.92−2.82 (1 H, m, H-6″a), 2.41−2.37 (1 H, m, H-4″), 2.26−2.22 (1 H, m, 6″b), 1.11 (9 H, t, J = 7.4 Hz, $(C\underline{H}_3CH_2)_3N$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 152.6, 145.8, 142.3, 130.2, 105.1, 101.8, 100.6, 93.0, 85.1, 79.5, 74.5, 73.7, 71.4, 65.5, 63.6, 47.4, 28.8, 8.8; ^{31}P NMR (202 MHz, D_2O) δ −9.29 (d), −10.33 (d); HRMS (ESI-ion trap, negative) calcd for $C_{17}H_{23}N_4O_{12}P_2$ 537.07932 [(M − H)$^-$], found 537.08012; UV (H_2O) λ_{max} = 275 nm; HPLC purity 97.2% (retention time 3.2 min).

*7-Bromo-7-deaza-cyclic ADP-Carbocyclic-ribose (**8**).* Title compound **8** (37.0 OD_{277} units, white powder) was prepared from **21b** (59.7 OD_{277} units) according to the procedure described for **7**: ^{1}H NMR (500 MHz, D_2O) δ 8.84 (1 H, s, H-2), 7.42 (1 H, s, H-8), 5.66 (1 H, d, J = 6.3 Hz, H-1′), 5.02 (1 H, dd, J = 6.3, 2.2 Hz, H-2′), 4.82−4.78 (1 H, m, H-1″), 4.44 (1 H, dd, J = 5.0, 2.2 Hz, H-3′), 4.39−4.36 (1 H, m, H-5′a), 4.27 (1 H, dd, J = 9.6, 4.5 Hz), 4.21−4.19 (1 H, m, H-4′), 4.07 (1 H, dd, J = 9.6, 4.0 Hz, H-3″), 4.02−4.01 (2 H, m, H-5″a, H-5″b), 3.95−3.92 (1 H, m, H-5′b), 3.03 (6 H, q, J = 7.4 Hz, $(CH_3C\underline{H}_2)_3N$), 2.88−2.83 (1 H, m, H-6″a), 2.38 (1 H, ddd, J = 9.0, 6.8, 2.8 Hz, H-4″), 2.19 (1 H, ddd, J = 15.8, 6.8, 3.4 Hz, H-6″b), 1.10 (9 H, t, J = 7.4 Hz, $(C\underline{H}_3CH_2)_3N$); ^{13}C NMR (125 MHz, D_2O) δ 153.0, 145.5, 143.2, 129.6, 102.9, 93.3, 89.3, 85.2, 80.3, 79.5, 75.0, 71.2, 66.2, 65.5, 63.9, 47.2, 43.6, 28.9, 8.8; ^{31}P NMR (202 MHz, D_2O) δ −9.31 (d), −10.34 (d); HRMS (ESI-ion trap, negative) calcd for $C_{17}H_{22}N_4O_{12}BrP_2$ 614.98983 [(M − H)$^-$], found 614.99138; UV (H_2O) λ_{max} = 277 nm (ε = 8440, based on the total phosphate analysis); HPLC purity 99.9% (retention time 5.6 min).

■ ASSOCIATED CONTENT

Supporting Information

^{1}H NMR, ^{13}C NMR, and ^{31}P NMR charts of compounds. The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.joc.5b00723.

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Notes

The authors declare no competing financial interest.

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