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Straightforward Synthesis of the Poly(ADP-ribose) Branched Core Structure

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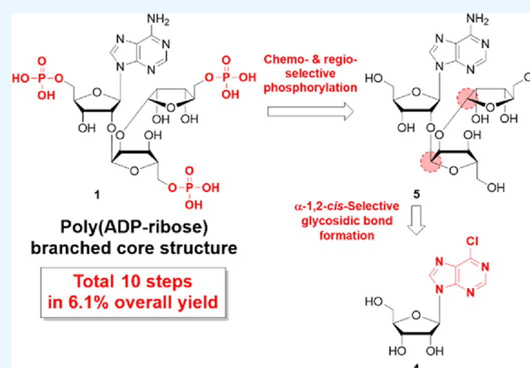
ABSTRACT: Poly(ADP-ribosyl)ation is a post-translational modification that produces poly(ADP-ribose) with a branched structure every 20−50 units; such branching structure has been previously suggested to be involved in regulating chromatin remodeling. To elucidate its detailed functions, we developed a straightforward method for the synthesis of the poly(ADP-ribose) branched core structure, α-D-ribofuranosyl-(1‴ → 2″)-α-D-ribofuranosyl-(1″ → 2′)-adenosine 5′,5″,5‴-trisphosphate **1**, from 6-chloropurine ribofuranoside **4** in 10 steps and 6.1% overall yield. The structure poses synthetic challenges for constructing iterative α-1,2-*cis*-glycosidic bonds in the presence of a purine base and the installation of three phosphate groups at primary hydroxyl groups. Iterative glycosidic bonds were formed by α-1,2-*cis*-selective ribofuranosylation using 2-*O*-(2-naphthylmethyl)-protected thioglycoside donor **6** and a thiophilic bismuth promoter. After the construction of diribofuranosyl adenosine **5** had been constructed, it was chemo- and regioselectively phosphorylated at a later stage. Subsequent deprotection provided the synthetic target **1**.



INTRODUCTION

ADP-ribosylation is a post-translational modification (PTM) in which the ADP-ribose moiety is transferred from nicotinamide adenine dinucleotide (NAD^+) to a specific amino acid residue, such as arginine, asparagine, aspartate, cysteine, glutamate, lysine, and serine.[1] This PTM plays a pivotal role in a variety of biological processes, including transcriptional regulation,[2] DNA repair,[3] cell differentiation,[4] stress response,[5] apoptosis,[6] proliferation of cancer cells,[7] and viral and bacterial infections[8] (including COVID-19[8c]). Poly(ADP-ribose)polymerase 1 (PARP1), one of the corresponding biosynthetic enzymes, catalyze elongation of the ADP-ribose chain at the 2′ position of the adenosine moiety in a terminal ADP-ribose residue through a 1,2-*cis*(α)-glycosidic linkage to provide a long-chain poly(ADP-ribose) (PAR) with up to approximately 200 units.[9] The PAR possesses a branched structure every 20−50 units that is linked to the 2″ position of the ribose moiety of an inner ADP-ribose through an α-glycosidic bond (Figure 1).[10]

Chen et al. showed that PARP2, the closet paralogue of PARP1, accelerates the branching formation and suggested that the PAR branching structure is involved in the regulation of chromatin remodeling by the recognition of DNA damage-specific histone chaperone APLF (aprataxin and PNKP like factor).[11] While more than 30 years has elapsed since the discover of the branched structure by Miwa and coworkers,[10a] its detailed functions at the molecular level have not been intensively investigated compared to those of the PAR liner structure. One major obstacle is the availability of homogeneous ADP-ribose molecules with branched structures, which are limited by the heterogeneity of naturally occurring PAR. Therefore, the development of chemical synthetic methods that facilitate the production of highly pure structurally well-defined molecules is a very important objective. Liu et al. reported the chemical synthesis of the PAR branched core structure, α-D-ribofuranosyl-(1‴ → 2″)-α-D-ribofuranosyl-(1″ → 2′)-adenosine 5′,5″,5‴-trisphosphate **1** (Scheme 1A).[12] While their synthetic route represents a pioneering chemical study of PAR, the long multistep synthesis and synthetic efficiency remain to be issues that need to be addressed. The longest linear sequence (LLS) in their route to **1** from the commercially available 1,3,5-tri-*O*-benzoyl-α-D-ribofuranose **2** involved 18 steps, with an overall yield of 4.1% and major drawbacks including several protecting group manipulations for *O*-glycosylation and phosphorylation, and the late-stage installation of the *N*-benzoyl-protected adenine base **3**. Therefore, we envisaged a straightforward route to shorten the synthesis of **1** and improve the overall yield. Herein, we

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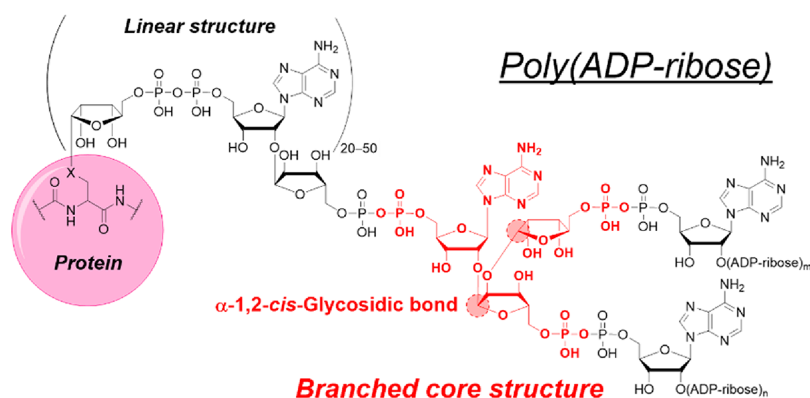
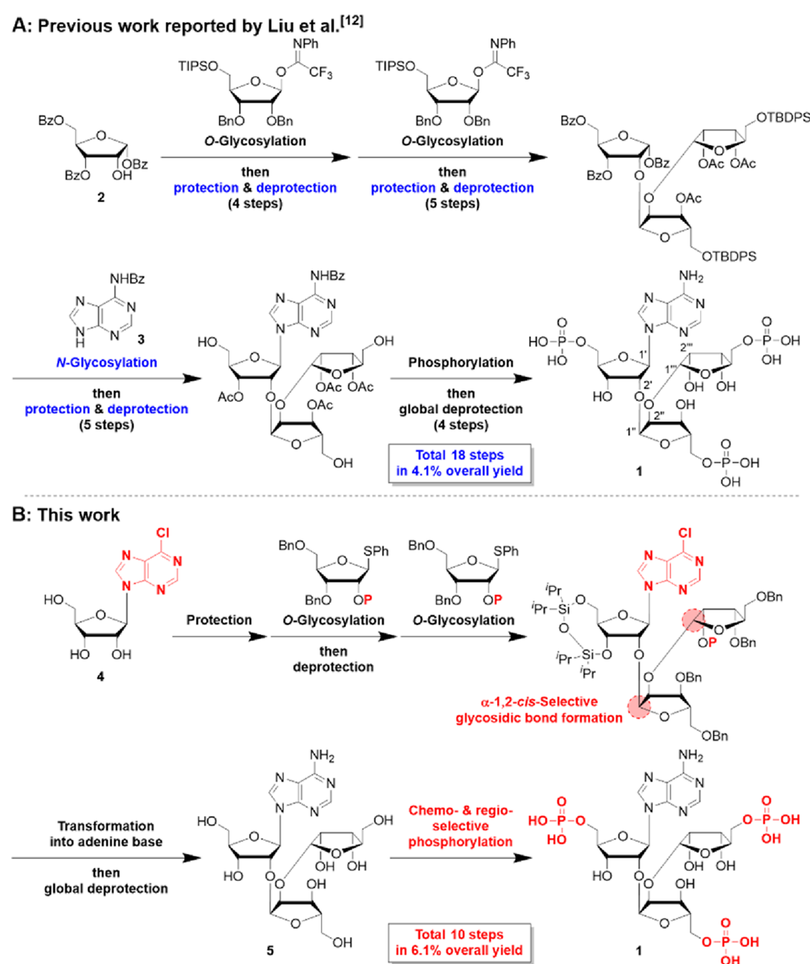


Figure 1. Structure of naturally occurring poly(ADP-ribose).

Scheme 1. Synthetic Outline of Previous Work (A) and This Work (B) in the Synthesis of Diribofuranosyl Adenosine Trisphosphate 1



report the straightforward synthesis of **1** from 6-chloropurine ribofuranoside **4** in 10 steps and 6.1% overall yield (Scheme 1B). Our synthetic route features α-1,2-*cis*-selective *O*-glycosidic bond formation and late-stage chemo- and regioselective phosphorylation of α-D-ribofuranosyl-(1‴ → 2″)-α-D-ribofuranosyl-(1″ → 2′)-adenosine **5**.

■ RESULTS AND DISCUSSION

For the improvement of the synthetic efficiency and overall yield of **1**, we highlight the following three key points: (1) the use of 6-chloropurine ribofuranoside **4** as the starting material for the synthesis of diribofuranosyl adenosine **5**, which avoids the challenging late-stage *N*-glycosylation of a purine base with trisaccharide building block; (2) the use of a 2-naphthylmethyl (NAP)-protecting group at the 2-position of the ribofuranosyl thioglycoside donor, as Ito and Ishiwata have widely reported that 1,2-*cis*-selective *O*-glycosylation can be achieved via intramolecular aglycon delivery (IAD) using 2-*O*-NAP-protected glycosyl donors;[13] and (3) chemo- and regioselective phosphorylation of fully unprotected substrate **5** to dramatically decrease the number of synthetic steps.

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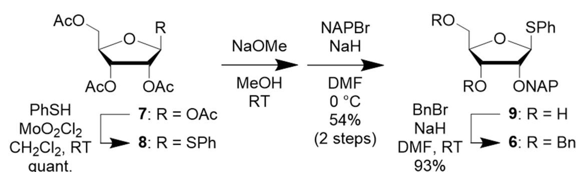
As depicted in Scheme 2, our efforts commenced with the preparation of the 2-*O*-NAP-protected ribofuranosyl thioglyco-

Scheme 2. Preparation of 2-*O*-NAP-Protected Ribofuranosyl Thioglycoside 6

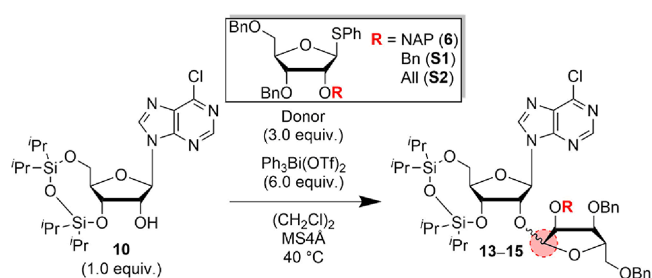


side donor **6**. Following the MoO_2Cl_2-catalyzed thioglycosylation method reported by Chen and coworkers,[14] phenyl *β*-thioglycoside **8** was obtained from commercially available tetra-acetate **7** in excellent yield. After deacetylation, the resulting triol was subjected to Williamson ether synthesis with NAP bromide and sodium hydride in *N*,*N*-dimethylformamide (DMF). The numbers of equivalents of reagents, reaction temperature, and methods to add the NAP bromide were optimized, which enabled regioselective monoalkylation at the 2-position to afford the 2-*O*-NAP ether **9** in a satisfactory yield of 54%, along with monoalkylated regioisomers and dialkylated compounds. A similar regioselective alkylation strategy has been reported for the secondary 2′-hydroxyl group of ribonucleosides in the presence of a primary 5′-hydroxyl group.[15] Benzylation of the remaining hydroxyl groups provided the desired product **6** in 93% yield.

We next attempted NAP-mediated IAD to *α*-1,2-*cis*-selectively ribofuranosylate 6-chloropurine ribofuranoside acceptor **10** with thioglycoside donor **6** (Scheme 3). 2′-Hydroxyl acceptor **10** was readily prepared by reacting commercially available compound **4** with 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane ($TIPDSCl_2$) in the presence of pyridine.[16] The corresponding mixed acetal **11** was formed from **10** and **6** in an acceptable yield of 66% as a single stereoisomer using the previous DDQ procedure.[13] Although the thioglycoside moiety is usually activated with methyl triflate, we used triphenylbismuth ditriflate ($Ph_3Bi(OTf)_2$),[17] a thiophilic promoter developed by Pohl's group, owing to differences in the thioglycoside moiety (methylthio versus phenylthio) and the presence of the purine base. While **11** was observed to be completely converted by TLC, the desired IAD product **12** was not obtained; the reaction provided **10** and a 1,2-*O*-naphthylidene acetal byproduct derived from the thioglycoside moiety of **11**. It is likely that thioglycoside activation using $Ph_3Bi(OTf)_2$ and/or the stereochemistry of the mixed acetal **11** is unfavorable for glycosidic bond formation by IAD.

Based on these results, we examined the direct ribofuranosylation of 3′,5′-TIPDS-protected acceptor **10** with thioglycoside donor **6** using $Ph_3Bi(OTf)_2$ (Table 1). The

Table 1. Screening for Direct Ribofuranosylation

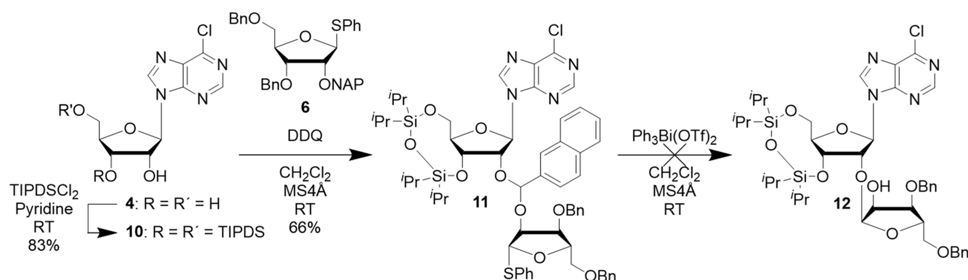


entry	donor	R	product	yield (%)	α/β ratio
1	**6**	NAP	**13**	63	6.1/1
2	**S1**	Bn	**14**	61[a]	3/1[b]
3	**S2**	all	**15**	36[a]	*α* only

[a]Including impurities. [b]α/β ratio was determined based on ^{1}H NMR spectrum.

corresponding *α*-1,2-*cis* product **13** was successfully obtained in good yield and with high stereoselectivity (63%, α/β = 6.1:1) using 3 equivalents of **6** (entry 1). To determine the effect of the 2-*O*-NAP-protecting group on the coupling yield and stereoselectivity during ribofuranosylation, **10** was coupled with 3 equivalents of the 2-*O*-benzylated and allylated thioglycosides **S1**[18] and **S2**, respectively. Glycosylation of **10** with **S1** afforded the desired product **14** in good yield and stereoselectivity although lower than in entry 1 (61%, α/β = 3:1; entry 2), while the use of **S2** facilitated complete *α*-1,2-*cis*-selective ribofuranosylation to give **15** but with a dramatically lower coupling yield (36%, *α* only; entry 3). These results indicated that the presence of the 2-*O*-electron-rich alkyl substituent in the thioglycoside donor, such as the NAP or benzyl group, stabilizes the oxocarbenium intermediate, which facilitates glycosidic bond formation. However, the *α*-1,2-*cis*-directing effect of the 2-*O*-NAP protecting group remains questionable. One possible explanation for the *α*-1,2-*cis*-selectivity observed in each entry involves remote stereocontrol by the C3 benzyloxy group. Larsen et al. experimentally showed that a ribofuranosyl oxocarbenium ion prefers to be in a pseudoaxial E_3 conformation stabilized by a C3 alkoxy group, which assists *α*-1,2-*cis*-selective ribofuranosylation through nucleophilic attack from the inside face of the envelope intermediate.[19] Notably, the anomeric configurations of ribofuranosides **13**−**15** were easily determined by vicinal ^{1}H NMR coupling constants between the C1 and C2 protons

Scheme 3. *α*-1,2-*cis*-Selective Ribofuranosylation via NAP-Mediated IAD



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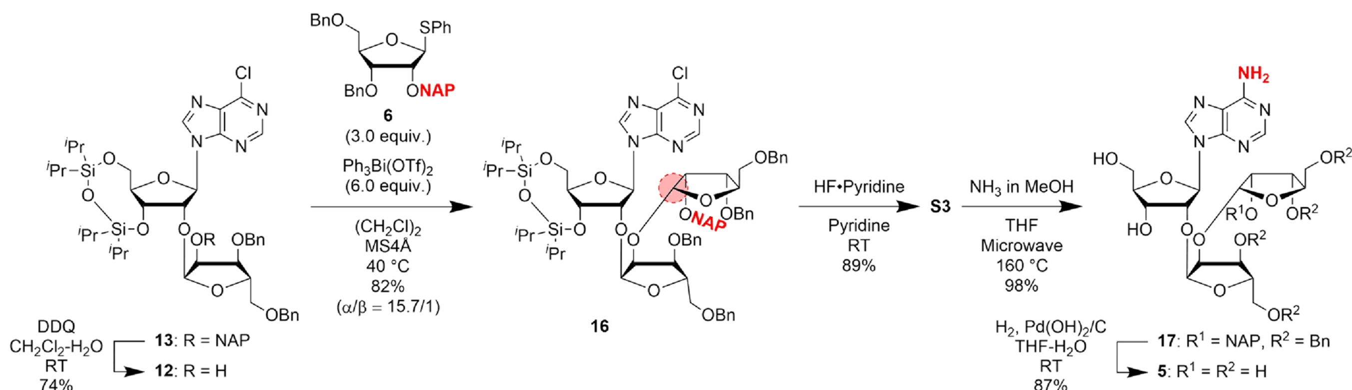
Scheme 4. Synthesis of Diribofuranosyl Adenosine 5

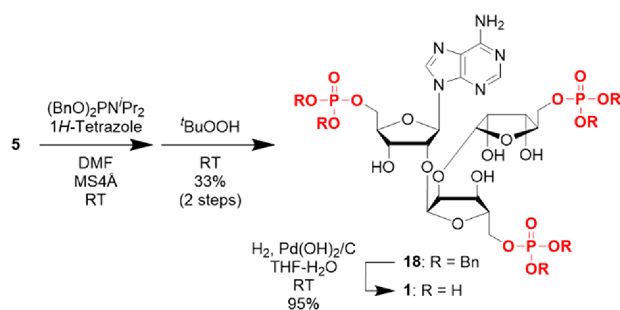


($^3J_{H1,H2}$) and the ^{13}C NMR chemical shift of the anomeric carbon.[20] The $^3J_{H1,H2}$ value of an α-1,2-*cis*-ribofuranosides (4−5 Hz) is larger than its β-1,2-*trans* stereoisomer (0−2 Hz). The carbon signal of the α-isomer typically appears between 100 and 105 ppm; in contrast, the corresponding β-isomers is downshifted by approximately 5 ppm.

With the appropriate reaction conditions for α-1,2-*cis*-selective ribofuranosidic bond formation with **6** in hand, we next focused on the synthesis of diribofuranosyl adenosine **5** (Scheme 4). The 2″-*O*-NAP group was oxidatively removed using DDQ to afford glycosyl acceptor **12** in 74% yield. The same ribofuranosylation conditions as shown in entry 1 of Table 1 produced the desired α-1,2-*cis* product **16** in high coupling yield and stereoselectivity (82%, α/β = 15.7:1), which is ascribable to the difference in acceptor reactivities. The 2″-hydroxyl group of **12** is expected to be more reactive than the 2′-hydroxyl group of **10** because the reactive site is far from the bulky and electron-withdrawing 6-chloropurine base. The use of the more reactive acceptor **12** predominately provided the kinetic α-product via the pseudoaxial E_3 conformation. Exposure to the hydrogen fluoride−pyridine complex deblocked the TIPDS group of **16** to give **S3** in 89% yield. The following microwave-assisted transformation of the 6-chloropurine base with ammonia−methanol solution proceeded efficiently to afford the diribofuranosyl adenosine structure **17** in 98% yield, after which **5** was synthesized in 87% yield by the hydrogenolysis of **17** using Pearlman's catalyst to cleave the remaining benzyl-type protecting groups. Notably, our synthetic approach (seven steps in 19.4% overall yield) is five steps shorter than the first reported synthesis[21] and afforded a more than 10-fold improved overall yield (12 steps in 1.6% overall yield).

To complete the synthesis of **1**, we chemo- and regioselectively phosphorylated the fully unprotected substrate **5** at its primary hydroxyl groups (Scheme 5). Although several regioselective phosphorylation methods using P(V) reagents[22] have been reported, the phosphoramidite method that uses a P(III) reagent,[23] which is widely used in nucleic acid syntheses, was chosen. We hypothesized that the acid used to activate the phosphoramidite reagent protonates the free amino group, thereby decreasing its reactivity. In addition, the secondary hydroxyl groups adjacent or close to the *N*- and *O*-glycosidic bonds are expected to be less reactive. According to Haché et al.,[24] the primary alcohol in a 1,2-diol substrate can be regioselectively phosphorylated by phosphitylation with a di(*tert*-butyl)phosphoramidite reagent ((tBuO)$_2$PNiPr$_2$) and subsequent oxidation. Accordingly, **5** was first phosphitylated

Scheme 5. Chemo- and Regioselective Phosphorylation of 5 to Synthesize 1



using an excess amount of (tBuO)$_2$PNiPr$_2$; while the reaction proceeded only slightly, the predominant introduction of a single phosphite group was confirmed by mass spectrometry. Therefore, we chose to use the dibenzylphosphoramidite reagent ((BnO)$_2$PNiPr$_2$), which is less sterically hindered and more reactive than (tBuO)$_2$PNiPr$_2$; indeed, **5** was phosphitylated slowly when (BnO)$_2$PNiPr$_2$ was added. The addition of 16.5 equivalents of (BnO)$_2$PNiPr$_2$ mainly resulted in triphosphitylation along with some diphosphitylation. The desired 5′,5″,5‴-phosphorylated product **18** was isolated in an acceptable yield of 33% over two steps after oxidation and purification. Finally, hydrogenolysis using Pearlman's catalyst led to debenzylation and the target PAR branched core structure **1** in an excellent yield of 95%. The ^{1}H NMR spectrum of the synthesized **1** is consistent with that of the naturally occurring **1** reported by Miwa and coworkers.[10b] Although only a slightly upfield shift of 0.1 ppm was observed, this difference is account for the presence or absence of sodium 2,2-dimethyl-2-silapentane-5-sulfonate.

■ CONCLUSIONS

We have successfully developed a straightforward method for the synthesis of the PAR branched core structure **1**. In this method, α-1,2-*cis*-selective *O*-ribofuranosidic bond formation and late-stage chemo- and regioselective phosphorylation of **5** were achieved. In terms of both the number of steps and overall yield, our route to **1** is significantly more efficient compared to that previously reported.[12] Recently, Filippov and coworker have synthesized a branched tri(ADP-ribose) for the first time.[25] Further synthetic efforts on such molecules lead to the detailed elucidation of the functions of the branched structure at the molecular level. Application of this method to

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synthesis of branched oligo(ADP-ribose) molecules is currently underway in our laboratory.

■ EXPERIMENTAL SECTION

General Methods. All chemicals were purchased from commercial suppliers and used without further purification. Dry solvents for reaction media (DMF, MeOH, pyridine, THF, and toluene) were purchased from Kanto Chemical Co., Inc. (Tokyo, Japan) and used without purification. Microwave irradiation experiments were carried out using Initiator+ Eight (Biotage, Uppsala, Sweden) in sealed reaction vials without pressure. 6-Chloropurine ribofuranoside **4** and tetra-*O*-acetyl-*β*-D-ribofuranose **7** were purchased from Tokyo Chemical Industry Co., Ltd. (Tokyo, Japan). Phenyl 2,3,5-tri-*O*-acetyl-1-thio-*β*-D-ribofuranoside **8** and 6-chloropurine 3′,5′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-*β*-D-ribofuranoside **10** were prepared using previously reported methods.[16] Phenyl 2,3,5-tri-*O*-benzyl-1-thio-*β*-D-ribofuranoside **S1** and phenyl 2-*O*-allyl-3,5-tri-*O*-benzyl-1-thio-*β*-D-ribofuranoside **S2** were prepared using conventional methods (see the Supporting Information). TLC analyses were performed using TLC plates (silica gel 60F254 on a glass plate; Merck KGaA, Darmstadt, Germany). Compound detection was carried out either by exposure to UV light (2536 Å) or by soaking in H_2SO_4 solution (10% in EtOH) or phosphomolybdic acid solution (20% in EtOH) followed by heating. Flash column chromatography separations were performed by using silica gel (80 mesh and 300 mesh; Fuji Silysia Co., Aichi, Japan) or Biotage Isolera equipped with Biotage SNAP Ultra Silica Cartridges (10, 25, 50, 100, and 340 g). The quantity of silica gel was typically 100 to 200 times the weight of the crude sample. Sephadex LH-20 (Cytiva, Marlborough, MA, USA) was used for size-exclusion chromatography. Solvent systems for chromatography were specified in v/v ratios. ^{1}H and ^{13}C NMR spectra were recorded using an Avance III 500 spectrometer (Bruker, Billerica, MA, USA). Chemical shifts in ^{1}H NMR spectra were expressed in ppm (δ) relative to the Me_4Si signal (0.00 ppm). Chemical shifts in ^{13}C NMR spectra were reported relative to the residual solvent signals ($CDCl_3$, 77.16 ppm; CD_3OD, 49.00 ppm). The data are presented as follows: chemical shift multiplicity (s = singlet, d = doublet, dd = doublet of doublets, td = triplet of doublets, ddd = doublet of doublet of doublets, t = triplet, q = quartet, m = multiplet), coupling constant (Hz), and integration. Structural assignments were performed using additional information from the 2D NMR (COSY) experiment. High-resolution mass spectrometry (ESI-TOF MS) was performed using a Bruker micrOTOF mass spectrometer (Bruker, Billerica, MA, USA). Optical rotations were measured with a SEPA-500 automatic polarimeter (Horiba, Kyoto, Japan).

Experimental Details. ***α*-D-Ribofuranosyl-(1‴ → 2″)-*α*-D-ribofuranosyl-(1″ → 2′)-adenosine 5′,5′′,5‴-trisphosphate (1).** To a solution of **18** (13.1 mg, 9.98 μmol) in THF/H_2O (2:1, 0.50 mL), $Pd(OH)_2/C$ (20% loading: 13.5 mg) was added at room temperature. After stirring for 1.5 h at room temperature under a H_2 gas atmosphere, the reaction mixture was filtered through a membrane filter and lyophilized to yield **1** (7.3 mg, 95%) as white powder. The physical characteristics of **1** was consistent with the reported values.[10b,12]

***α*-D-Ribofuranosyl-(1‴ → 2″)-*α*-D-ribofuranosyl-(1″ → 2′)-adenosine (5).** To a solution of **17** (143 mg, 0.139 mmol) in THF/H_2O (2:1, 4.2 mL), $Pd(OH)_2/C$ (20% loading: 188 mg) was added at room temperature. The reaction was monitored by TLC ($CHCl_3$/MeOH = 10:1 and $CHCl_3/MeOH/H_2O$ = 5:6:2). After stirring for 5 h at room temperature under a H_2 gas atmosphere, $Pd(OH)_2/C$ (20% loading: 94.0 mg) was re-added to the reaction mixture. After stirring for additional 12 h, $Pd(OH)_2/C$ (20% loading: 94.0 mg) was added again. After stirring for additional 13 h, the reaction mixture was filtered through a pad of Celite, and the pad was rinsed with MeOH. The combined filtrate was concentrated in vacuo. The resulting residue was purified using reverse-phase silica gel column chromatography (H_2O only) to yield **5** (63.0 mg, 87%) as white powder. The physical characteristics of **5** was consistent with the reported values.[21]

Phenyl 3,5-Di-O-benzyl-2-O-(2-naphthylmethyl)-1-thio-β-D-ribofuranoside (6). To a solution of **9** (2.44 g, 6.38 mmol) in DMF (25.5 mL), NaH (2.28 g, 19.1 mmol) was added at 0 °C. After stirring for 30 min, BnBr (0.760 mL, 19.1 mmol) was added at 0 °C. The reaction was monitored by TLC (*n*-hexane/EtOAc = 1:1). After stirring for 17 h at room temperature, the reaction was quenched with MeOH. The reaction mixture was diluted with $CHCl_3$ and washed with water and brine. The organic layer was dried over Na_2SO_4, filtered off, and co-evaporated with toluene. The resulting residue was purified using flash silica gel column chromatography (*n*-hexane/EtOAc = 17:3) to yield **6** (3.34 g, 93%) as a colorless syrup: $[\alpha]_D^{25}$−23.4 (c 0.9, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.76−7.05 (m, 22 H, Ar), 5.47 (d, 1 H, $J_{1,2}$ = 4.0 Hz, H-1), 4.78 (d, 1 H, J_{gem} = 11.0 Hz, CH_2Ar), 4.70 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.57−4.47 (m, 4 H, CH_2Ar), 4.32 (dd, 1 H, $J_{3,4}$ = 4.5 Hz, $J_{4,5}$ = 10.0 Hz, H-4), 3.95 (m, 2 H, H-2, H-3), 3.54 (m, 2 H, H-5a, H-5b); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 138.2137.7, 135.0, 133.6, 133.3, 133.2, 132.4, 128.9, 128.4, 128.4, 128.3, 128.1, 128.0, 128.0, 127.8, 127.7, 127.6, 127.6, 127.0, 126.2, 126.1, 88.9, 82.0, 80.4, 77.6, 77.4, 76.9, 73.4, 72.3, 70.3; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{34}H_{36}O_4S$, 585.2070; found [M + Na]$^+$, 585.2068.

Phenyl 2-O-(2-Naphthylmethyl)-1-thio-β-D-ribofuranoside (9). To a solution of **8** (12.9 g, 35.0 mmol) in MeOH (70.0 mL), NaOMe (1 M solution in MeOH: 3.5 mL) was added at room temperature. The reaction was monitored by TLC (*n*-hexane/EtOAc = 1:1). After stirring for 1 h at room temperature, the reaction was neutralized with Muromac (H$^+$ form) at 0 °C. The reaction mixture was evaporated in vacuo and exposed to high vacuum for 2 h. The resulting residue was dissolved in DMF (70.0 mL). To the solution, NaH (1.40 g, 35.0 mmol) was added at 0 °C. After stirring for 30 min, NAPBr (7.70 g, 35.0 mmol) was added at 0 °C. The reaction was monitored by TLC (*n*-hexane/EtOAc = 1:1). After stirring for 17 h at 0 °C, the reaction was quenched with MeOH. The reaction mixture was diluted with $CHCl_3$ and washed with water and brine. The organic layer was dried over Na_2SO_4, filtered off, and co-evaporated with toluene. The resulting residue was purified using flash silica gel column chromatography (*n*-hexane/EtOAc = 9:1) to yield **9** (6.78 g, 54%) as a colorless syrup: $[\alpha]_D^{25}$−23.9 (c 1.0, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 7.86−7.25 (m, 12 H, Ar), 5.42 (d, 1 H, $J_{1,2}$ = 4.5 Hz, H-1), 4.90 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.80 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.20 (m, 1 H, H-3), 4.07 (m, 2 H, H-2, H-4), 3.81 (m, 1 H, H-5a), 3.63 (m, 1 H, H-5b), 2.65 (d, 1 H, $J_{3\text{-}OH,3}$ = 6.5 Hz, 3-OH), 1.75 (dd, 1 H, $J_{5\text{-}OH,5a}$ = $J_{5\text{-}OH,5b}$ = 4.4 Hz, 5-OH); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 134.1, 133.2, 132.9, 132.4, 132.3, 132.1, 131.9, 131.6, 129.1, 128.9, 128.6, 128.0, 127.8, 127.2, 126.4, 126.3, 125.8, 88.4, 85.9, 82.4,

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73.1, 70.6, 62.3; HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{22}H_{22}O_4S$, 405.1131; found [M + Na]$^+$, 405.1132.

*2-Naphthaldehyde [6-Chloropurine 3″,5″-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosid-2″-yl] (Phenyl 3′,5′-di-O-benzyl-1-thio-β-D-ribofuranoside) Acetal (**11**).* To a solution of **6** (117 mg, 0.210 mmol) and **10** (84.0 mg, 0.160 mmol) in CH_2Cl_2 (3.2 mL), MS4Å (320 mg) was added at room temperature. After stirring for 30 min at room temperature, DDQ (50 mg, 0.220 mmol) was added at 0 °C. The reaction was monitored by TLC (Toluene/EtOAc = 3:1). After stirring for 8 h at room temperature, the reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The combined filtrate was washed with sat. aq. $NaHCO_3$, water, and brine. The organic layer was dried over Na_2SO_4, filtered off, and concentrated in vacuo. The resulting residue was purified using size-exclusion chromatography on Sephadex LH-20 ($CHCl_3$/MeOH = 1:1) to yield **11** (single isomer: 115 mg, 66%) as a colorless syrup: $[\alpha]_D^{25}$−60.2 (c 1.3, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.38, 8.20 (2 s, 2 H, H-2, H-8), 7.90−7.07 (m, 22 H, Ar), 6.25 (s, 1 H, ArC*H*<), 6.16 (s, 1 H, H-1′), 5.87 (d, 1 H, $J_{1'',2''}$ = 3.0 Hz, H-1″), 4.71 (t, 1 H, $J_{2'',3''}$ = 4.2 Hz, H-2″), 4.60 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.53 (m, 2 H, CH_2Ar), 4.45 (m, 2 H, H-2′, H-3′), 4.40 (d, 1 H, $J_{3',4'}$ = 4.5 Hz, H-4′), 4.34 (td, 1 H, $J_{4'',5''a}$ = $J_{4'',5''b}$ = 4.5 Hz, $J_{3'',4''}$ = 5.7 Hz, H-4″), 4.27 (d, H, J_{gem} = 9.0 Hz, CH_2Ar), 4.21 (d, 1 H, J_{gem} = 13.5 Hz, H-5′a), 4.11 (t, 1 H, H-3″), 3.99 (dd, 1 H, $J_{4',5'b}$ = 1.9 Hz, H-5′b), 3.66−3.58 (m, 2 H, H-5″a, H-5″b), 1.15−0.73 (m, 28 H, ^{i}Pr); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 151.6, 150.7, 150.3143.5, 138.2, 137.4, 134.1, 133.8, 132.8, 132.2, 131.3, 129.0, 128.5, 128.4, 128.3, 127.8, 127.7, 127.6, 127.3, 126.7, 126.3, 124.7, 103.6, 90.0, 89.1, 81.9, 81.8, 80.2, 78.3, 76.2, 73.4, 72.3, 70.4, 68.5, 59.6, 17.5, 17.3, 17.1, 16.9, 13.3, 13.0, 12.8; HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{58}H_{69}ClN_4O_4SSi_2$, 1111.3905; found [M + Na]$^+$, 1111.3908.

*6-Chloropurine 3″,5″-Di-O-benzyl-α-D-ribofuranosyl-(1″ → 2′)-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranoside (**12**).* To a solution of **13α** (164 mg, 0.170 mmol) in CH_2Cl_2/H_2O (20:1, 3.3 mL), DDQ (75.0 mg, 0.330 mmol) was added at 0 °C. The reaction was monitored by TLC (*n*-hexane/EtOAc = 1:1). After stirring for 3.5 h at room temperature, the reaction mixture was diluted with $CHCl_3$ and washed with sat. aq. $NaHCO_3$ and brine. The organic layer was dried over Na_2SO_4, filtered off, and concentrated in vacuo. The resulting residue was purified using flash silica gel column chromatography (*n*-hexane/EtOAc = 7:3) to yield **12** (107 mg, 74%) as a colorless syrup: $[\alpha]_D^{25}$ + 39.5 (c 1.1, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.71, 8.50 (2 s, 2 H, H-2, H-8), 7.36−7.24 (m, 10 H, Ar), 6.17 (s, 1 H, H-1′), 5.51 (d, 1 H, $J_{2'',3''}$ = 4.5 Hz, H-1″), 4.65 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.60−4.50 (m, 4 H, H-2′, H−4′, CH_2Ar), 4.43 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.37 (dd, 1 H, $J_{3'',4''}$ = 4.0 Hz, $J_{4'',5''a}$ = $J_{4'',5''b}$ = 7.5 Hz, H-4″), 4.29 (d, 1 H, J_{gem} = 13.5 Hz, H-5′a), 4.26−4.21 (m, 2 H, H-2″, H-3′), 4.02 (dd, 1 H, $J_{4',5'b}$ = 2.5 Hz, H-5′b), 3.89 (m, 1 H, H-3″), 3.46−3.40 (m, 2H, H-5″a, H-5″b), 3.18 (d, 1 H, $J_{2''\text{-OH},H\text{-}2''}$ = 11.5 Hz, 2″-OH) 1.15−0.83 (m, 28 H, ^{i}Pr); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 152.0, 151.0, 150.5, 143.0, 138.0, 137.9, 132.5, 128.4, 128.3, 127.8, 127.7, 127.6, 101.2 (<u>C-1″</u>), 89.2, 82.3, 81.9, 79.1, 75.9, 73.5, 72.8, 72.1, 69.7, 67.6, 59.4, 17.5, 17.4, 17.3, 17.0, 16.9, 14.1, 13.5, 12.9, 12.7; HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{44}H_{61}ClN_4O_9Si_2$, 863.3245; found [M + Na]$^+$, 863.3243.

*6-Chloropurine 3″,5″-Di-O-benzyl-2″-O-(2-naphthylmethyl)-D-ribofuranosyl-(1″ → 2′)-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranoside (**13**).* To a solution of **6** (2.08 g, 3.70 mmol) and **10** (648 mg, 1.23 mmol) in $(CH_2Cl)_2$ (74.0 mL), MS4Å (7.40 g) and $Ph_3Bi(OTf)_2$ (5.45 g, 7.38 mmol) were added at room temperature. The reaction was monitored by TLC (*n*-hexane/EtOAc = 3:1). After stirring for 24 h at 40 °C, the reaction was quenched with NEt_3. The reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The combined filtrate was concentrated in vacuo. The resulting residue was purified using size-exclusion chromatography on Sephadex LH-20 ($CHCl_3$/MeOH = 1:1) and flash silica gel column chromatography (*n*-hexane/EtOAc = 7:3) to yield **13α** (648 mg, 54%) as a yellow syrup along with **13β** (113 mg, 9%) as a yellow syrup.

13α: $[\alpha]_D^{25}$ + 39.8 (c 1.1, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.57, 8.26 (2 s, 2 H, H-2, H-8), 7.79−7.16 (m, 22 H, Ar), 6.01 (s, 1 H, H-1′), 5.44 (d, 1 H, $J_{1'',2''}$ = 2.5 Hz, H-1″), 4.89 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.80 (d, 1 H, CH_2Ar), 4.69 (d, 1 H, J_{gem} = 13.0 Hz, CH_2Ar), 4.63−4.55 (m, 3 H, CH_2Ar, H-2′, H-4′), 4.47−4.32 (m, 4 H, CH_2Ar, H-3′, H-4″), 4.23 (d, 1 H, J_{gem} = 13.5 Hz, H-5′a), 4.01−3.95 (m, 3 H, H-2″, H-3″, H-5′b), 3.43−3.36 (m, 2 H, H-5a″, H-5b″), 1.13−0.88 (m, 28 H, ^{i}Pr); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$) δ 151.9, 150.9, 150.5, 143.6, 138.6, 138.0, 135.3, 133.3, 133.2, 132.5, 128.5, 128.5, 128.4, 128.4, 128.1, 127.9, 127.8, 127.8, 127.7, 127.7, 127.0, 127.0, 126.4, 126.3, 126.0, 101.0 (<u>C-1″</u>), 89.6, 82.3, 81.8, 79.6, 78.7, 75.0, 73.6, 73.2, 72.2, 69.9, 67.9, 59.6, 31.1, 17.6, 17.5, 17.2, 17.2, 17.1, 17.1, 13.6, 13.1, 13.0, 12.9; HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{44}H_{61}ClN_4O_9Si_2$, 1003.3871; found [M + Na]$^+$, 1003.3869.

13β: $[\alpha]_D^{25}$−13.9 (c 1.6, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.59, 7.97 (2 s, 2 H, H-2, H-8), 7.84−7.18 (m, 22 H, Ar), 6.01 (s, 1 H, H-1′), 5.45 (s, 1 H, H-1″), 4.95 (dd, 1 H, $J_{2',3'}$ = 5.1 Hz, $J_{3',4'}$ = 5.2 Hz, H-3′), 4.76 (d, 1 H, J_{gem} = 12.0 Hz, CH_2Ar), 4.70 (d, 1 H, H-2′), 4.63 (d, 1 H, J_{gem} = 13.0 Hz, CH_2Ar), 4.47 (m, 3 H, CH_2Ar), 4.34 (q, 1 H, $J_{3'',4''}$ = $J_{4'',5''a}$ = $J_{4'',5''b}$ = 3.5 Hz, H-4″), 4.27 (td, 1 H, $J_{4',5'a}$ = $J_{4',5'b}$ = 4.5 Hz, H-4′), 4.11 (m, 2 H, H-3″, H-5′a), 4.00 (m, 2 H, H-2″, H-5′b), 3.74 (dd, 1 H, J_{gem} = 10.9 Hz, H-5″a), 3.58 (dd, 1 H, H-5″b), 1.10−0.88 (m, 28 H, ^{i}Pr); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 151.9, 151.2, 150.8, 144.7, 138.0, 135.3, 133.4, 133.2, 132.6, 128.6, 128.5, 128.5, 128.4, 128.1, 128.0, 127.9, 127.9, 127.9, 127.8, 127.6, 126.8, 126.3, 126.2, 125.9, 106.2 (<u>C-1″</u>), 89.6, 81.7, 81.1, 79.6, 78.8, 77.7, 73.3, 72.7, 72.3, 70.1, 69.9, 60.1, 17.6, 17.4, 17.4, 17.3, 17.3, 17.2, 13.5, 13.1, 12.9; HRMS (ESI) m/z [M + Na]$^+$ calcd for $C_{44}H_{61}ClN_4O_9Si_2$, 1003.3871; found [M + Na]$^+$, 1003.3874.

*6-Chloropurine 2″,3″,5″-Tri-O-benzyl-D-ribofuranosyl-(1″ → 2′)-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranoside (**14**).* To a solution of **S1** (168 mg, 0.324 mmol) and **10** (57.1 mg, 0.108 mmol) in $(CH_2Cl)_2$ (6.5 mL), MS4Å (648 mg) and $Ph_3Bi(OTf)_2$ (478 mg, 0.648 mmol) were added at room temperature. The reaction was monitored by TLC (*n*-hexane/EtOAc = 3:1). After stirring for 12 h at 40 °C, the reaction was quenched with NEt_3. The reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The combined filtrate was concentrated in vacuo. The resulting residue was purified using size-exclusion chromatography on Sephadex LH-20 ($CHCl_3$/MeOH = 1:1) and flash silica gel column chromatography (*n*-hexane/EtOAc = 4:1) to yield a mixture of **14α** and **14β** (67.4 mg containing

impurities, 61%, α/β = 3:1) as a colorless syrup. **14α** and **14β** were partially separated by flash silica gel column chromatography (*n*-hexane/EtOAc = 5:1) in pure form.

14α: $[\alpha]_D^{25}$ + 5.32 (c 0.5, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$): δ 8.66, 8.27 (2 s, 2 H, H-2, H-8), 7.32−7.19 (m, 15 H, Ar), 6.09 (s, 1 H, H-1′), 5.48 (d, 1 H, $J_{1'',2''}$ = 3.9 Hz, H-1″), 4.69−4.58 (m, 6 H, H-2′, H-3′, CH_2Ar), 4.48 (d, 1 H, J_{gem} = 12.1 Hz, CH_2Ar), 4.44 (dd, 1H, $J_{3'',4''}$ = 4.0 Hz, H-4″), 4.40 (d, 1 H, J_{gem} = 12.1 Hz, CH_2Ar), 4.30 (d, 1 H, $J_{3',4'}$ = 9.2 Hz, H-4′), 4.22 (d, 1 H, J_{gem} = 13.4 Hz, H-5′a), 4.00 (dd, 1 H, $J_{4',5'b}$ = 2.5 Hz, H-5′b), 3.92 (m, 2 H, H-2″, H-3″), 3.45−3.38 (m, 2 H, H-5″a, H-5″b), 1.10−0.98 (m, 28 H, ^{i}Pr); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 152.0, 151.1, 150.6, 143.7, 138.6, 138.0, 137.8, 132.6, 128.5, 128.5, 128.4, 128.1, 128.1, 127.9, 127.8, 127.8, 127.7, 101.1 (C-1″), 89.7, 82.5, 81.8, 79.6, 78.8, 77.7, 75.0, 73.6, 72.8, 72.3, 70.0, 68.1, 59.7, 17.6, 17.5, 17.5, 17.3, 17.2, 17.2, 17.1, 13.6, 13.1, 13.0, 12.9; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{48}H_{63}ClN_4O_9Si_2$, 953.3714; found [M + Na]$^+$, 953.3715.

14β: $[\alpha]_D^{25}$−13.2 (c 0.6, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$): δ 8.60, 7.98 (2 s, 2 H, H-2, H-8), 7.36−7.19 (m, 15 H, Ar), 6.01 (s, 1 H, H-1′), 5.41 (s, 1 H, H-1″), 4.95 (dd, 1 H, $J_{3',4'}$ = 4.2 Hz, $J_{2',3'}$ = 5.2 Hz, H-3′), 4.74 (d, 1 H, J_{gem} = 12.1 Hz, CH_2Ar), 4.69 (d, 1 H, H-2′), 4.62−4.45 (m, 5 H, CH_2Ar), 4.30 (ddd, 1 H, $J_{4'',5''a}$ = 3.5 Hz, $J_{4'',5''b}$ = 4.0 Hz, $J_{3'',4''}$ = 4.5 Hz, H-4″), 4.25 (ddd, $J_{4',5'a}$ = 4.6 Hz, 1H, H-4′), 4.11 (near d, 1 H, J_{gem} = 13.2 Hz, H-5′a), 4.05 (d, 1 H, H-3″), 4.00 (m, 2 H, H-2″, H-5′b), 3.73 (dd, 1 H, J_{gem} = 10.9 Hz, H-5″a), 3.58 (dd, 1 H, H-5″b), 1.12−0.97 (m, 28 H, ^{i}Pr); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 151.9, 151.2, 150.9, 144.7, 138.0, 138.0, 137.8, 132.6, 128.6, 128.1, 128.0, 127.9, 127.9, 127.6, 106.1 (C-1″), 89.6, 81.7, 81.1, 79.6, 78.8, 77.6, 73.3, 72.6, 72.1, 70.1, 69.6, 60.1, 17.6, 17.4, 17.4, 17.3, 17.2, 13.5, 13.1, 12.9; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{48}H_{63}ClN_4O_9Si_2$, 953.3714; found [M + Na]$^+$, 953.3713.

*6-Chloropurine 3″,5″-Di-O-benzyl-2″-O-allyl-α-D-ribofuranosyl-(1″ → 2′)-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranoside (**15**).* To a solution of **S2** (142 mg, 0.307 mmol) and **10** (54.1 mg, 0.102 mmol) in $(CH_2Cl)_2$ (6.1 mL), MS4Å (610 mg) and $Ph_3Bi(OTf)_2$ (453 mg, 0.614 mmol) were added at room temperature. The reaction was monitored by TLC (*n*-hexane/EtOAc = 3:1). After stirring for 4 h at 40 °C, the reaction was quenched with NEt_3. The reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The filtrate was concentrated in vacuo. The resulting residue was purified using size-exclusion chromatography on Sephadex LH-20 ($CHCl_3$/MeOH = 1:1) and flash silica gel column chromatography (*n*-hexane/EtOAc = 9:2) to yield **15** (45.7 mg containing impurities, 36%, α only) as a colorless syrup. **15** was partially separated by flash silica gel column chromatography (*n*-hexane/EtOAc = 4:1) in pure form: $[\alpha]_D^{25}$ + 1.62 (c 1.1, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$): δ 8.69, 8.36 (2 s, 2 H, H-2, H-8), 7.34−7.23 (m, 10 H, Ar), 6.18 (s, 1 H, H-1′), 5.92−5.86 (m, 1 H, CH_2CH=CH_2), 5.52 (d, 1 H, $J_{1'',2''}$ = 4.5 Hz, H-1″), 5.25 (near d, 1 H, CH_2CH=CH_2), 5.16 (near d, 1 H, CH_2CH=CH_2), 4.67−4.49 (m, 5 H, H-2′, H-4′, CH_2Ar), 4.42 (m, 2 H, H-4″, CH_2Ar), 4.32 (d, 1 H, $J_{3',4'}$ = 9.0 Hz, H-3′), 4.24 (d, 1 H, J_{gem} = 13.5 Hz, H-5′a), 4.17−4.10 (m, 2 H, CH=CH_2), 4.02−3.88 (m, 3 H, H-2″, H-3″, H-5′), 3.45−3.37 (m, 2 H, H-5″), 1.25−0.91 (m, 28 H, ^{i}Pr); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 151.9, 151.0, 150.6, 143.5, 138.4, 137.9, 134.5, 132.5, 128.4, 128.3, 128.0, 127.7, 127.6, 101.1 (C-1″), 89.6, 82.3, 81.7, 79.7, 78.8, 74.8, 73.5, 72.1, 72.0, 69.8, 67.8, 59.5, 17.5, 17.4, 17.3, 17.1, 17.0, 16.9, 13.5, 13.0, 12.9, 12.7; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{44}H_{61}ClN_4O_9Si_2$, 903.3558; found [M + Na]$^+$, 903.3559.

*6-Chloropurine 3‴,5‴-Di-O-benzyl-2‴-O-(2-naphthylmethyl)-D-ribofuranosyl-(1‴ → 2″)-3″,5″-di-O-benzyl-α-D-ribofuranosyl-(1″ → 2′)-3′,5′-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranoside (**16**).* To a solution of **6** (523 mg, 0.930 mmol) and **12** (265 mg, 0.310 mmol) in $(CH_2Cl)_2$ (6.2 mL), MS4Å (620 mg) and $Ph_3Bi(OTf)_2$ (1.44 g, 1.86 mmol) were added at room temperature. The reaction was monitored by TLC (*n*-hexane/EtOAc = 2:1). After stirring for 9 h at 40 °C, the reaction was quenched with NEt_3. The reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The combined filtrate was concentrated in vacuo. The resulting residue was purified using size-exclusion chromatography on Sephadex LH-20 ($CHCl_3$/MeOH = 1:1) and flash silica gel column chromatography (*n*-hexane/EtOAc = 7:3) to yield **16α** (310 mg, 77%) as a yellow syrup along with **16β** (21.0 mg, 5%) as a yellow syrup.

16α: $[\alpha]_D^{25}$ + 66.6 (c 1.3, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$): δ 8.64, 8.41 (2 s, 2 H, H-2, H-8), 7.78−7.14 (m, 27 H, Ar), 6.09 (s, 1 H, H-1′), 5.69 (d, 1 H, $J_{1'',2''}$ = 4.1 Hz, H-1″), 5.56 (d, 1 H, $J_{1''',2'''}$ = 4.2 Hz, H-1‴), 5.06−5.00 (m, 2 H, CH_2Ar), 4.74 (m, 2 H, CH_2Ar), 4.60−4.35 (m, 11 H, H-2′, H-2″, H-4′, H-4″, H-4‴, CH_2Ar), 4.12 (m, 1 H, H-2‴), 4.04 (m, 2 H, H-3″, H3‴), 3.80 (m, 1 H, H-3′), 3.67−3.61 (m, 2 H, H-5′a, H-5‴a), 3.54 (dd, 1 H, $J_{4',5'b}$ = 4.2 Hz, J_{gem} = 10.6 Hz, H-5′b), 3.74 (m, 2 H, H-5″a, H-5″b), 3.36 (m, 1 H, $J_{4''',5'''b}$ = 2.3 Hz, J_{gem} = 13.7 Hz, H-5‴b), 1.02−0.81 (m, 28 H, ^{i}Pr); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 152.0, 150.9, 150.4, 143.1, 139.6, 138.6, 138.2, 138.0, 136.0, 133.2, 133.0, 132.5, 128.5, 128.5, 128.4, 128.0, 128.0, 127.9, 127.8, 127.7, 127.7, 127.6, 127.0, 126.3, 126.3, 126.1, 125.9, 100.6 (C-1″), 100.5 (C-1‴), 89.4, 82.6, 82.1, 81.8, 79.9, 78.7, 76.5, 76.0, 75.5, 73.7, 73.6, 72.8, 72.5, 72.4, 70.1, 69.9, 67.3, 59.1, 17.6, 17.5, 17.4, 17.2, 17.1, 17.1, 17.0, 13.5, 13.0, 12.9, 12.9; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{62}H_{80}ClN_4O_{12}Si_2$, 1315.5232; found [M + Na]$^+$, 1315.5235.

16β: $[\alpha]_D^{25}$ + 35.9 (c 1.0, $CHCl_3$); ^1H NMR (500 MHz, $CDCl_3$): δ 8.52, 8.37 (2 s, 2 H, H-2, H-8), 7.79−7.11 (m, 27 H, Ar), 6.02 (s, 1 H, H-1′), 5.39 (d, 1 H, $J_{1'',2''}$ = 4.3 Hz, H-1″), 5.25 (s, 1 H, H-1‴), 4.73 (m, 2 H, CH_2Ar), 4.57−4.37 (m, 12 H, H-2′, H-4′, H-4″, H-4‴, CH_2Ar), 4.27 (near d, 1 H, $J_{2',3'}$ = 9.4 Hz, H-3′), 4.21−4.15 (m, 2 H, H-2″, H-5′a), 4.00−3.93 (m, 4 H, H-2‴, H-3″, H-3‴, H-5′b), 3.58 (m, 2 H, H-5″a, H-5″b), 3.41 (m, 2 H, H-5‴a, H-5‴b), 1.10−0.87 (m, 28 H, ^{i}Pr); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 152.0, 150.9, 150.5, 143.3, 138.7, 138.1, 138.0, 137.8, 135.5, 133.4, 133.2, 132.6, 128.5, 128.4, 128.3, 128.3, 128.3, 128.0, 127.8, 127.8, 127.7, 127.6, 127.3, 126.7, 126.3, 126.0, 125.9, 105.7 (C-1‴), 101.7 (C-1″), 89.2, 82.4, 81.6, 80.7, 80.1, 78.1, 77.7, 76.4, 74.6, 73.7, 73.1, 72.5, 72.4, 72.0, 71.5, 67.7, 59.6, 17.7, 17.5, 17.5, 17.2, 17.1, 17.0, 13.6, 13.1, 13.0, 12.8; HRMS (ESI) *m/z* [M + Na]$^+$ calcd for $C_{62}H_{80}ClN_4O_{12}Si_2$, 1315.5232; found [M + Na]$^+$, 1315.5236.

*6-Chloropurine 3‴,5‴-Di-O-benzyl-2‴-O-(2-naphthylmethyl)-α-D-ribofuranosyl-(1‴ → 2″)-3″,5″-di-O-benzyl-α-D-ribofuranosyl-(1″ → 2′)-β-D-ribofuranoside (**S3**).* To a solution of **16α** (112 mg, 86.0 μmol) in pyridine (0.86 mL), HF·pyridine (86 μL) was added at 0 °C. The reaction was monitored by TLC ($CHCl_3$/MeOH = 10:1). After stirring for 6 h at room temperature, sat. aq. $NaHCO_3$ was carefully added

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to the reaction mixture at 0 °C. The mixture was diluted with $CHCl_3$ and washed with sat. aq. $NaHCO_3$ and brine. The organic layer was dried over Na_2SO_4, filtered off, and concentrated in vacuo. The resulting residue was purified using flash silica gel column chromatography ($CHCl_3$/MeOH = 99:1) to yield **S3** (80.7 mg, 89%) as a yellow syrup: $[\alpha]_D^{25}$ + 42.1 (c 0.5, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.67, 7.90 (2 s, 2 H, H-2, H-8), 7.82−7.14 (m, 27 H, Ar), 5.80 (d, 1 H, $J_{1',2'}$ = 7.0 Hz, H-1′), 5.14 (m, 2 H, H-1″, H-1‴), 5.01 (d, 1 H, J_{gem} = 12.1 Hz, CH_2Ar), 5.00−4.89 (m, 3 H, H-2′, H-4‴, CH_2Ar), 4.77 (d, 1 H, J_{gem} = 11.1 Hz, CH_2Ar), 4.65 (d, 2 H, CH_2Ar), 4.56−4.40 (m, 6 H, CH_2Ar, 5′-OH), 4.37−4.28 (m, 4 H, H-3′, H-4′, H-4″, H-2‴), 4.23 (brs, 1 H, 3′-OH), 4.05 (d, 1 H, $J_{2''',3'''}$ = 5.1 Hz, H-3‴), 3.98 (m, 2 H, H-2″, H-3″), 3.86 (d, 1 H, J_{gem} = 12.7 Hz, H-5‴a), 3.68 (m, 1 H, 5‴b), 3.52−3.44 (m, 2 H, H-5′a, H-5′b), 3.40−3.33 (m, 2 H, H-5″a, H-5″b); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 152.0, 151.5, 150.6, 145.5, 138.2, 138.0, 137.9, 137.8, 135.3, 133.3, 133.3, 133.2, 128.5, 128.5, 128.4, 128.3, 128.1, 128.1, 127.8, 127.8, 127.8, 127.7, 127.6, 127.5, 127.1, 126.4, 126.4, 126.2, 101.1, 100.1, 89.5, 87.7, 83.7, 82.4, 78.5, 78.4, 78.3, 77.4, 77.0, 76.4, 73.6, 73.5, 72.9, 72.7, 72.3, 70.2, 70.0, 63.0; HRMS (ESI) *m/z* $[M + Na]^+$ calcd for $C_{59}H_{59}ClN_4O_{12}$, 1073.3710; found $[M + Na]^+$, 1073.3710.

*3‴,5‴-Di-O-benzyl-2‴-O-(2-naphthylmethyl)-α-D-ribofuranosyl-(1‴ → 2″)-3″,5″-di-O-benzyl-α-D-ribofuranosyl-(1″ → 2′)-adenosine (**17**).* To a solution of **S3** (56.3 mg, 54.0 μmol) in THF (1.08 mL), NH_3 (2 M solution in MeOH: 2.14 mL) was added at room temperature. The reaction was monitored by TLC ($CHCl_3$/MeOH = 10:1). After stirring for 1 h at 160 °C under microwave irradiation, the reaction mixture was diluted with $CHCl_3$ and washed with water and brine. The organic layer was dried over Na_2SO_4, filtered off, and concentrated in vacuo. The resulting residue was purified using flash silica gel column chromatography ($CHCl_3$/MeOH = 19:1) to yield **17** (54.3 mg, 98%) as a colorless syrup: $[\alpha]_D^{25}$ + 21.7 (c 1.0, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.25, 7.87 (2 s, 2 H, H-2, H-8), 7.79−7.10 (m, 27 H, Ar), 6.36 (d, 1 H, $J_{5'\text{-OH},5'a}$ = $J_{5'\text{-OH},5'b}$ = 10.7 Hz, 5′-OH), 6.15 (brs, 2 H, NH_2), 5.78 (d, 1 H, $J_{1',2'}$ = 7.3 Hz, H-1′), 5.13 (m, 2 H, H-1″, H-1‴), 5.02−4.92 (m, 3 H, H-2′, CH_2Ar), 4.76−4.58 (m, 4 H, CH_2Ar), 4.52−4.39 (m, 5 H, CH_2Ar, 3′-OH), 4.35−4.27 (m, 5 H, H-3′, H-3″, H-4″, H-4‴, H-2‴), 4.05 (d, 1 H, $J_{2''',3'''}$ = 5.4 Hz H-3‴), 3.97 (m, 2 H, H-2″, H-4′), 3.88 (d, 1 H, J_{gem} = 12.4 Hz, H-5′a), 3.71 (m, 1 H, H-5′b), 3.51−3.43 (m, 2 H, H-5″a, H-5″b), 3.42−3.35 (m, 2 H, H-5‴a, H-5‴b); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 156.1, 152.5, 148.6, 140.7, 138.3, 138.1, 137.9, 137.8, 135.4, 133.3, 133.2, 128.8, 128.4, 128.3, 128.2, 128.1, 127.8, 127.7, 127.7, 127.5, 127.1, 126.4, 126.3, 126.1, 121.1, 100.9, 100.2, 89.3, 88.0, 83.7, 82.2, 78.7, 78.4, 78.2, 77.4, 76.8, 76.3, 73.5, 73.5, 73.5, 72.9, 72.8, 72.7, 70.2, 69.9, 63.3 HRMS (ESI) *m/z* $[M + Na]^+$ calcd for $C_{59}H_{61}N_5O_{12}$, 1054.4209; found $[M + Na]^+$, 1054.4208.

*α-D-Ribofuranosyl-(1‴ → 2″)-α-D-ribofuranosyl-(1″ → 2′)-adenosine 5′,5′′,5‴-tris(dibenzyl phosphate) (**18**).* To a solution of **5** (19.7 mg, 37.1 μmol) in DMF (1.5 mL), MS4Å (150 mg) and 1*H*-tetrazole (52.0 mg, 741 μmol) were added at room temperature. After stirring for 4 h, dibenzyl *N,N*-diisopropylphosphoramidite (55.4 μL, 167 mmol) was added at room temperature. The reaction was monitored by TLC ($CHCl_3$/MeOH = 4:1). After stirring for 1.5 h at room temperature, dibenzyl *N,N*-diisopropylphosphoramidite (112 μL, 334 mmol) was re-added to the reaction mixture. After stirring for additional 1 h, dibenzyl *N,N*-diisopropylphosphoramidite (112 μL, 334 mmol) was added again. After stirring for additional 1 h, ^{t}BuOOH (5.5 M solution in decane: 111 μL) was added to the reaction mixture at 0 °C. After stirring for 1 h at room temperature, the reaction was quenched with sat. aq. $Na_2S_2O_3$. The reaction mixture was filtered through a pad of Celite, and the pad was rinsed with $CHCl_3$. The combined filtrate was washed with sat. aq. $NaHCO_3$ and brine. The organic layer was dried over Na_2SO_4, filtered off, and concentrated in vacuo. The resulting residue was purified using flash silica gel column chromatography ($CHCl_3$/MeOH = 5:1) to yield **18** (16.0 mg, 33%) as a colorless syrup: $[\alpha]_D^{25}$ + 53.2 (c 0.5, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): δ 8.27, 8.11 (2 s, 2 H, H-2, H-8), 7.32−7.26 (m, 30 H, Ar), 6.13 (d, 1 H, $J_{1',2'}$ = 5.0 Hz, H-1′), 5.81 (brs, 2 H, NH_2), 5.03−4.96 (m, 13 H, H-1″, CH_2Ar), 4.80 (d, 1 H, $J_{1''',2'''}$ = 5.5 Hz, H-1‴), 4.59 (brs, 1 H, H-2′), 4.35 (brs, 1 H, H-3′), 4.27−4.12 (m, 6 H, H-3″, H-3‴, H-4′, H-4″, H-4‴, H-5′a), 4.00−3.95 (m, 7 H, H-2″, H-2‴, H-5′b, H-5″a, H-5″b, H-5‴a, H-5‴b), 1.88 (brs, 4 H, 3′-OH, 3″-OH, 2‴-OH, 3‴-OH); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 155.7, 153.1, 149.4, 139.4, 135.7, 135.7, 135.7, 135.7, 135.6, 128.8, 128.8, 128.1, 128.1, 120.1, 101.8, 100.8, 86.9, 83.9, 83.6, 79.6, 78.8, 72.5, 70.9, 70.7, 70.5, 69.8, 69.8, 69.8, 69.7, 69.6, 67.2, 67.0; ^{31}P{^{1}H} NMR (200 MHz, $CDCl_3$): δ 1.40, 1.36; HRMS (ESI) m/z $[M + Na]^+$ calcd for $C_{62}H_{68}N_5O_{21}P_3$, 1334.3512; found $[M + Na]^+$, 1334.3510.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.2c04732.

Experimental procedures and NMR spectra for all new compounds (PDF)

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) For selected reviews on ADP-ribosylation, see: (a) Gibson, B. A.; Kraus, W. L. New insights into the molecular and cellular functions of poly(ADP-ribose) and PARPs. *Nat. Rev. Mol. Cell Biol.* **2012**, *13*, 411–424. (b) Ryu, K. W.; Kim, D.-S.; Kraus, W. L. New facets in the regulation of gene expression by ADP-ribosylation and poly(ADP-ribose) polymerases. *Chem. Rev.* **2015**, *115*, 2453–2481. (c) Teloni, F.; Altmeyer, M. Readers of poly(ADP-ribose): designed to be fit for purpose. *Nucleic Acids Res.* **2016**, *44*, 993–1006.

(2) For selected examples of ADP-ribosylation in transcriptional regulation, see: (a) Hassa, P. O.; Haenni, S. S.; Buerki, C.; Meier, N. I.; Lane, W. S.; Owen, H.; Gersbach, M.; Imhof, R.; Hottiger, M. O. Acetylation of poly(ADP-ribose) polymerase-1 by p300/CREB-binding protein regulates coactivation of NF-κB-dependent transcription. *J. Biol. Chem.* **2005**, *280*, 40450–40464. (b) Krishnakumar, R.; Kraus, W. L. PARP-1 regulates chromatin structure and transcription through a KDM5B-dependent pathway. *Mol. Cell* **2010**, *39*, 736–749. (c) Chen, H.; Ruiz, P.; Novikov, L.; Casill, A. D.; Park, J. W.; Gamble, M. J. MacroH2A1.1 and PARP-1 cooperate to regulate transcription by promoting CBP-mediated H2B acetylation. *Nat. Struct. Mol. Biol.* **2014**, *21*, 981–989.

(3) For selected examples of ADP-ribosylation in DNA repair, see: (a) Kang, H. C.; Lee, Y.-I.; Shin, J.-H.; Andrabi, S. A.; Chi, Z.; Gagné, J.-P.; Lee, Y.; Ko, H. S.; Lee, B. D.; Poirier, G. G.; Dawson, V. L.; Dawson, T. M. Iduna is a poly(ADP-ribose) (PAR)-dependent E3 ubiquitin ligase that regulates DNA damage. *Proc. Natl. Acad. Sci. U. S. A.* **2011**, *108*, 14103–14108. (b) Khoury-Haddad, H.; Guttmann-Raviv, N.; Ipenberg, I.; Huggins, D.; Jeyasekharan, A. D.; Ayoub, N. PARP1-dependent recruitment of KDM4D histone demethylase to DNA damage sites promotes double-strand break repair. *Proc. Natl. Acad. Sci. U. S. A.* **2014**, *111*, E728–E737. (c) Singh, H. R.; Nardozza, A. P.; Möller, I. R.; Knobloch, G.; Kistemaker, H. A. V.; Hassler, M.; Harrer, N.; Blessing, C.; Eustermann, S.; Kotthoff, C.; Huet, S.; Mueller-Planitz, F.; Filippov, D. V.; Timinszky, G.; Rand, K. D.; Ladurner, A. G. A poly-ADP-ribose trigger releases the auto-inhibition of a chromatin remodeling oncogene. *Mol. Cell* **2017**, *68*, 860–871.e7. (d) Ruiz, P. D.; Hamilton, G. A.; Park, J. W.; Gamble, M. J. MacroH2A1 regulation of poly(ADP-ribose) synthesis and stability prevents necrosis and promotes DNA repair. *Mol. Cell. Biol.* **2019**, *40*, e00230-19.

(4) For selected examples of ADP-ribosylation in cell differentiation, see: (a) Hemberger, M.; Nozaki, T.; Winterhager, E.; Yamamoto, H.; Nakagama, H.; Kamada, N.; Suzuki, H.; Ohta, T.; Ohki, M.; Masutani, M.; Cross, J. C. *Parp1*-deficiency induces differentiation of ES cells into trophoblast derivatives. *Dev. Biol.* **2003**, *257*, 371–381. (b) Gao, F.; Kwon, S.; Zhao, Y.; Jin, Y. PARP1 poly(ADP-ribosyl)ates Sox2 to control Sox2 protein levels and FGF4 expression during embryonic stem cell differentiation. *J. Biol. Chem.* **2009**, *284*, 22263–22273. (c) Roper, S. J.; Chrysanthou, S.; Senner, C. E.; Sienerth, A.; Gnan, S.; Murray, A.; Masutani, M.; Latos, P.; Hemberger, M. ADP-ribosyltransferases Parp1 and Parp7 safeguard pluripotency of ES cells. *Nucleic Acids Res.* **2014**, *42*, 8914–8927.

(5) For a selected example of ADP-ribosylation in stress response, see: Aberle, L.; Krüger, A.; Reber, J. M.; Lippmann, M.; Hufnagel, M.; Schmalz, M.; Trussina, I. R. E. A.; Schlesiger, S.; Zubel, T.; Schütz, K.; Marx, A.; Hartwig, A.; Ferrando-May, E.; Bürkle, A.; Mangerich, A. PARP1 catalytic variants reveal branching and chain length-specific functions of poly(ADP-ribose) in cellular physiology and stress response. *Nucleic Acids Res.* **2020**, *48*, 10015–10033.

(6) For selected examples of ADP-ribosylation in apoptosis, see: (a) Yu, S.-W.; Andrabi, S. A.; Wang, H.; Kim, N. S.; Poirier, G. G.; Dawson, T. M.; Dawson, V. L. Apoptosis-inducing factor mediates poly(ADP-ribose) (PAR) polymer-induced cell death. *Proc. Natl. Acad. Sci. U. S. A.* **2006**, *103*, 18314–18319. (b) Kumari, A.; Iwasaki, T.; Pyndiah, S.; Cassimere, E. K.; Palani, C. D.; Sakamuro, D. Regulation of E2F1-induced apoptosis by poly(ADP-ribosyl)ation. *Cell Death Differ.* **2015**, *22*, 311–322.

(7) For selected examples of ADP-ribosylation in proliferation of cancer cells, see: (a) Seimiya, H.; Smith, S. The telomeric poly(ADP-ribose) polymerase, tankyrase 1, contains multiple binding sites for telomeric repeat binding factor 1 (TRF1) and a novel acceptor, 182-kDa Tankyrase-binding protein (TAB182). *J. Biol. Chem.* **2002**, *277*, 14116–14126. (b) Seimiya, H.; Muramatsu, Y.; Smith, S.; Tsuruo, T. Functional subdomain in the ankyrin domain of tankyrase 1 required for poly(ADP-ribosyl)ation of TRF1 and telomere elongation. *Mol. Cell. Biol.* **2004**, *24*, 1944–1955. (c) Muramatsu, Y.; Tahara, H.; Ono, T.; Tsuruo, T.; Seimiya, H. Telomere elongation by a mutant tankyrase 1 without TRF1 poly(ADP-ribosyl)ation. *Exp. Cell Res.* **2008**, *314*, 1115–1124. (d) Li, N.; Zhang, Y.; Han, X.; Lian, K.; Wang, J.; Feng, L.; Wang, W.; Songyang, Z.; Lin, C.; Yang, L.; Yu, Y.; Chen, J. Poly-ADP ribosylation of PTEN by tankyrases promotes PTEN degradation and tumor growth. *Genes Dev.* **2015**, *29*, 157–170.

(8) For selected examples of ADP-ribosylation in viral and bacterial infections, see: (a) Fehr, A. R.; Channappanavar, R.; Jankevicius, G.; Fett, C.; Zhao, J.; Athmer, J.; Meyerholz, D. K.; Ahel, I.; Perlman, S. The conserved coronavirus macrodomain promotes virulence and suppresses the innate immune response during severe acute respiratory syndrome coronavirus infection. *MBio* **2016**, *7*, e01721-16. (b) McPherson, R. L.; Abraham, R.; Sreekumar, E.; Ong, S.-E.; Cheng, S.-J.; Baxter, V. K.; Kistemaker, H. A. V.; Filippov, D. V.; Griffin, D. E.; Leung, A. K. L. ADP-ribosylhydrolase activity of Chikungunya virus macrodomain is critical for virus replication and virulence. *Proc. Natl. Acad. Sci. U. S. A.* **2017**, *114*, 1666–1671. (c) Frick, D. N.; Virdi, R. S.; Vuksanovic, N.; Dahal, N.; Silvaggi, N. R. Molecular basis for ADP-ribose binding to the Mac1 domain of SARS-CoV-2 nsp3. *Biochemistry* **2020**, *59*, 2608–2615.

(9) Alvarez-Gonzalez, R.; Jacobson, M. K. Characterization of polymers of adenosine diphosphate ribose generated in vitro and in vivo. *Biochemistry* **1987**, *26*, 3218–3224.

(10) (a) Miwa, M.; Saikawa, N.; Yamaizumi, Z.; Nishimura, S.; Sugimura, T. Structure of poly(adenosine diphosphate ribose): identification of 2′-[1″-ribosyl-2′′-(or 3′′-)(1‴-ribosyl)]adenosine-5′,5′′,5′′′-tris(phosphate) as a branch linkage. *Proc. Natl. Acad. Sci. U. S. A.* **1979**, *76*, 595–599. (b) Miwa, M.; Ishihara, M.; Takishima, S.; Takasuka, N.; Maeda, M.; Yamaizumi, Z.; Sugimura, T.; Yokoyama, S.; Miyazawa, T. The branching and linear portions of poly(adenosine

diphosphate ribose) have the same $\alpha(1 \rightarrow 2)$ ribose-ribose linkage. *J. Biol. Chem.* **1981**, *256*, 2916−2921.

(11) Chen, Q.; Kassab, M. A.; Dantzer, F.; Yu, X. PARP2 mediates branched poly ADP-ribosylation in response to DNA damage. *Nat. Commun.* **2018**, *9*, 3233.

(12) Liu, Q.; Kistemaker, H. A. V.; Overkleeft, H. S.; van der Marel, G. A.; Filippov, D. V. Synthesis of ribosyl-ribosyl-adenosine-5′,5″,5‴(triphosphate)−the naturally occurring branched fragment of poly(ADP ribose). *Chem. Commun.* **2017**, *53*, 10255−10258.

(13) (a) Ishiwata, A.; Munemura, Y.; Ito, Y. NAP ether mediated intramolecular aglycon delivery: A unified strategy for 1,2-*cis*-glycosylation. *Eur. J. Org. Chem.* **2008**, 4250−4263. (b) Ishiwata, A.; Ito, Y. Synthesis of docosasaccharide arabinan motif of mycobacterial cell wall. *J. Am. Chem. Soc.* **2011**, *133*, 2275−2291. (c) Ishiwata, A.; Ito, Y. Synthetic study and structural analysis of the antifreeze agent xylomannan from *Upis ceramboides*. *J. Am. Chem. Soc.* **2011**, *133*, 19524−19535.

(14) Weng, S.-S.; Lin, Y.-D.; Chen, C.-T. Highly diastereoselective thioglycosylation of functionalized peracetylated glycosides catalyzed by MoO_2Cl_2. *Org. Lett.* **2006**, *8*, 5633−5636.

(15) (a) Kikugawa, K.; Sato, F.; Tsuruo, T.; Imura, N.; Ukita, T. On the benzylation of nucleosides. II. A novel synthesis of 2′-O-benzyluridine. *Chem. Pharm. Bull.* **1968**, *16*, 1110−1115. (b) Nakamura, M.; Shimomura, Y.; Ohtoshi, Y.; Sasa, K.; Hayashi, H.; Nakano, H.; Yamana, K. Pyrene aromatic arrays on RNA duplexes as helical templates. *Org. Biomol. Chem.* **2007**, *5*, 1945−1951. (c) Karmakar, S.; Guenther, D. C.; Hrdlicka, P. J. Recognition of mixed-sequence DNA duplexes_design guidelines for invaders based on 2′-*O*-(pyren-1-yl)methyl-RNA monomers. *J. Org. Chem.* **2013**, *78*, 12040−12048.

(16) Kittaka, A.; Yamada, N.; Tanaka, H.; Nakamura, K. T.; Miyasaka, T. Radical-mediated cyclization of a 6-chloro-9-(2-deoxy-D-erythro-pent-1-enofuranosyl)-8-(2,2-dibromovinyl) purine. *Nucleosides Nucleotides* **1996**, *15*, 1447−1457.

(17) (a) Goswami, M.; Ellern, A.; Pohl, N. L. B. Bismuth(V)-mediated thioglycoside activation. *Angew. Chem., Int. Ed.* **2013**, *52*, 8441−8445. (b) Goswami, M.; Ashley, D. C.; Baik, M.-H.; Pohl, N. L. B. Mechanistic studies of bismuth(V)-mediated thioglycoside activation reveal differential reactivity of anomers. *J. Org. Chem.* **2016**, *81*, 5949−5962. (c) Kabotso, D. E. K.; Pohl, N. L. B. Pentavalent bismuth as a universal promoter for S-containing glycosyl donors with a thiol additive. *Org. Lett.* **2017**, *19*, 4516−4519.

(18) Kametani, T.; Kawamura, K.; Honda, T. New entry to the C-glycosilation by means of carbenoid displacement reaction. Its application to the synthesis of showdomycin. *J. Am. Chem. Soc.* **1987**, *109*, 3010−3017.

(19) Larsen, C. H.; Ridgway, B. H.; Shaw, J. T.; Smith, D. M.; Woerpel, K. A. Stereoselective *C*-glycosylation reactions of ribose derivatives: Electronic effects of five-membered ring oxocarbenium ions. *J. Am. Chem. Soc.* **2005**, *127*, 10879−10884.

(20) (a) Cyr, N.; Perlin, A. S. The conformations of furanosides. A 13C nuclear magnetic resonance study. *Can. J. Chem.* **1979**, *57*, 2504−2511. (b) Mizutani, K.; Kasai, R.; Nakamura, M.; Tanaka, O.; Matsuura, H. N.m.r. spectral study of α- and β-L-arabinofuranosides. *Carbohydr. Res.* **1989**, *185*, 27−38.

(21) Kistemaker, H. A. V.; Overkleeft, H. S.; van der Marel, G. A.; Filippov, D. V. Branching of poly(ADP-ribose): Synthesis of the core motif. *Org. Lett.* **2015**, *17*, 4328−4331.

(22) (a) Yoshikawa, M.; Kato, T.; Takenishi, T. Studies of phosphorylation. III. Selective phosphorylation of unprotected nucleosides. *Bull. Chem. Soc. Jpn.* **1969**, *42*, 3505−3508. (b) Sowa, T.; Ouchi, S. The facile synthesis of 5′-nucleotides by the selective phosphorylation of a primary hydroxyl group of nucleosides with phosphoryl chloride. *Bull. Chem. Soc. Jpn.* **1975**, *48*, 2084−2090. (c) Sakakura, A.; Katsukawa, M.; Ishihara, K. Selective synthesis of phosphate monoesters by dehydrative condensation of phosphoric acid and alcohols promoted by nucleophilic bases. *Org. Lett.* **2005**, *7*, 1999−2002. (d) Sakakura, A.; Katsukawa, M.; Ishihara, K. The oxorhenium(VII)-catalyzed direct condensation of phosphoric acid with an alcohol. *Angew. Chem., Int. Ed.* **2007**, *46*, 1423−1426. (e) Domon, K.; Puripat, M.; Fujiyoshi, K.; Hatanaka, M.; Kawashima, S. A.; Yamatsugu, K.; Kanai, M. Catalytic chemoselective *O*-phosphorylation of alcohols. *ACS Cent. Sci.* **2020**, *6*, 283−292. (f) Ociepa, M.; Knouse, K. W.; He, D.; Vantourout, J. C.; Flood, D. T.; Padial, N. M.; Chen, J. S.; Sanchez, B. B.; Sturgell, E. J.; Zheng, B.; Qiu, S.; Schmidt, M. A.; Eastgate, M. D.; Baran, P. S. Mild and chemoselective phosphorylation of alcohols using a Ψ-reagent. *Org. Lett.* **2021**, *23*, 9337−9342.

(23) Beaucage, S. L.; Iyer, R. P. Advances in the synthesis of oligonucleotides by the phosphoramidite approach. *Tetrahedron* **1992**, *48*, 2223−2311.

(24) Haché, B.; Brett, L.; Shakya, S. Phosphonate-free phosphorylation of alcohols using bis-(*tert*-butyl) phosphoramidite with imidazole·hydrochloride and imidazole as the activator. *Tetrahedron Lett.* **2011**, *52*, 3625−3629.

(25) Rack, J. G. M.; Liu, Q.; Zorzini, V.; Voorneveld, J.; Ariza, A.; Ebrahimi, K. H.; Reber, J. M.; Krassnig, S. C.; Ahel, D.; van der Marel, G. A.; Mangerich, A.; McCullagh, J. S. O.; Filippov, D. V.; Ahel, I. Mechanistic insights into the three steps of poly(ADP-ribosylation) reversal. *Nat. Commun.* **2021**, *12*, 4581.