Inhibition of Siderophore Biosynthesis in *Mycobacterium tuberculosis* with Nucleoside Bisubstrate Analogues: Structure−Activity Relationships of the Nucleobase Domain of 5′-*O*-[*N*-(Salicyl)sulfamoyl]adenosine

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5′-*O*-[*N*-(salicyl)sulfamoyl]adenosine (Sal-AMS) is a prototype for a new class of antitubercular agents that inhibit the aryl acid adenylating enzyme (AAAE) known as MbtA involved in biosynthesis of the mycobactins. Herein, we report the structure-based design, synthesis, biochemical, and biological evaluation of a comprehensive and systematic series of analogues, exploring the structure−activity relationship of the purine nucleobase domain of Sal-AMS. Significantly, 2-phenyl-Sal-AMS derivative **26** exhibited exceptionally potent antitubercular activity with an MIC_{99} under iron-deficient conditions of 0.049 μM while the *N*-6-cyclopropyl-Sal-AMS **16** led to improved potency and to a 64-enhancement in activity under iron-deficient conditions relative to iron-replete conditions, a phenotype concordant with the designed mechanism of action. The most potent MbtA inhibitors disclosed here display in vitro antitubercular activity superior to most current first line TB drugs, and these compounds are also expected to be useful against a wide range of pathogens that require aryl-capped siderphores for virulence.

Introduction

Tuberculosis (TB[a]) is the leading cause of infectious disease mortality by a bacterial pathogen responsible for over 1.5 million deaths each year.[1] While the course of treatment may vary, the shortest recommended therapy for active infection involves no less than 182 doses of a multifrontline antibiotic cocktail over a 6 month period.[2] Increasing reports of drug resistant *Mycobacterium tuberculosis* strains, namely, the virtually incurable extensively drug-resistant TB (XDR-TB) and the serious multidrug resistant TB (MDR-TB), are cause of great concern, as they are resistant to both first-line and second-line anti-TB drugs.[3] Additionally, co-infection with HIV/AIDS is now responsible for the majority of HIV-related deaths. New TB drugs with shortened periods of treatment that are effective against latent TB infections are urgently needed.

Free iron is an essential nutrient for almost all known organisms, but its concentration in serum and human body fluids is approximately 10^{-24} M, far too low to support bacterial colonization and growth. Many bacterial pathogens secrete and reimport small molecule iron-chelators termed siderophores for iron acquisition, making siderophore biosynthesis a logical target for antibiotic development.[4] This approach is particularly attractive with the discovery that many antibiotics ultimately disrupt iron homeostasis.[5] Iron has also recently been shown to play a key role in biofilm formation in *Mycobacterium smegmatis*.[6] Mutational analysis demonstrated that siderophore synthesis was required for biofilm formation.

In response to iron starvation, *M. tuberculosis* produces two series of structurally related siderophores that vary in the appended lipid side chain collectively referred to as mycobactins, critical for virulence and growth (**4** and **5**, Figure 1).[7,8] Mycobactins are required for virulence in THP-1 macrophages and in a murine model of TB infection and may also serve as a short-term iron reservoir potentially mediating reactive oxygen species (ROS) generation.[7–10] Biosynthesis of these aryl-capped siderophores is initiated by the aryl acid adenylation enzyme MbtA (Figure 1), which activates salicylic acid (**1**) forming an acyladenylate intermediate (Sal-AMP, **2**). MbtA is also responsible for loading the acyladenylate intermediate onto the thiolation domain of MbtB.[11] The remaining steps of mycobactins biosynthesis are catalyzed by a mixed nonribosomal peptide synthetase polyketide synthase (NRPS-PKS) assembly line.[11]

Recently, 5′-*O*-[*N*-(salicyl)sulfamoyl]adenosine (Sal-AMS, **6**, Figure 2), a rationally designed bisubstrate inhibitor, was reported as a stable mimic of the intermediate acyladenylate **2**.[12–14] Developing this compound as a useful probe and a potential antibacterial agent requires improving target specificity, potency, and lipophilicity. Herein, we report the structure-based design of a potent picomolar inhibitor with enhanced specificity and in vitro antitubercular activity superior to most current first line TB drugs.

Results

Molecular Modeling. A complete homology model of MbtA was constructed on the basis of the cocrystal structure of DhbE with an adenylated 2,3-dihydroxybenzoic acid (see Experimental

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[a] Abbreviations: AAAE, aryl acid adenylating enzyme; AMP, adenosine monophosphate; ATP, adenosine triphosphate; DMA, dimethylacetamide; DTT, dithiothreitol; HIV/AIDS, human immunodeficiency virus/acquired immune deficiency syndrome; HMWP2, high-molecular-weight protein 2; K_i^{app}, apparent inhibition constant; MDR-TB, multidrug resistant tuberculosis; MIC, minimum inhibitory concentration; MTT, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide; NHS, *N*-hydroxysuccinimide; NRPS, nonribosomal peptide synthetase; PDB, Protein Data Bank; PKS, polyketide synthase; PP_i, pyrophosphate; QM/MM, quantum mechanical/molecular mechanical; rmsd, root-mean-square deviation; ROS, reactive oxygen species; SAR, structure activity relationships; Sal-AMS, 5′-O-[N-(salicyl-)sulfamoyl]adenosine; TB, tuberculosis; TBAF, tetrabutylammonium fluoride; TFA, trifluoroacetic acid; XDR-TB, extensively drug resistant tuberculosis.

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Figure 1. Biosynthesis of the mycobactins and carboxymycobactins.[8] The depicted lipid side chain is representative, as both **4** and **5** are isolated as mixtures with various length lipid residues.

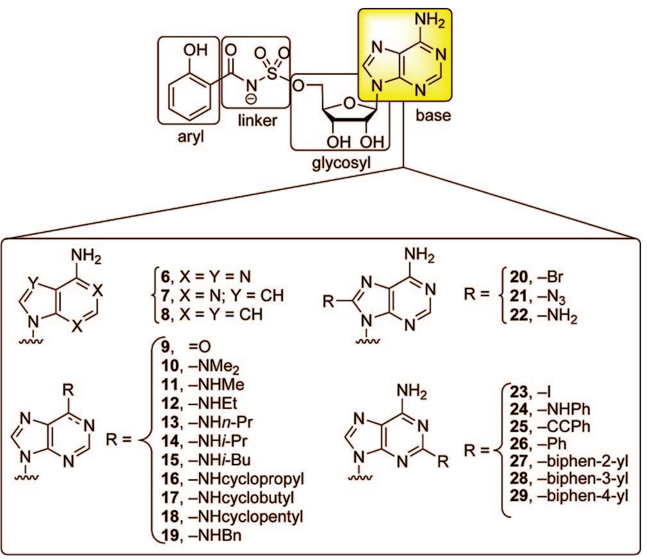


Figure 2. Bisubstrate inhibitors of MbtA. The expanded portion of the figure shows the nucleobase modifications described herein.

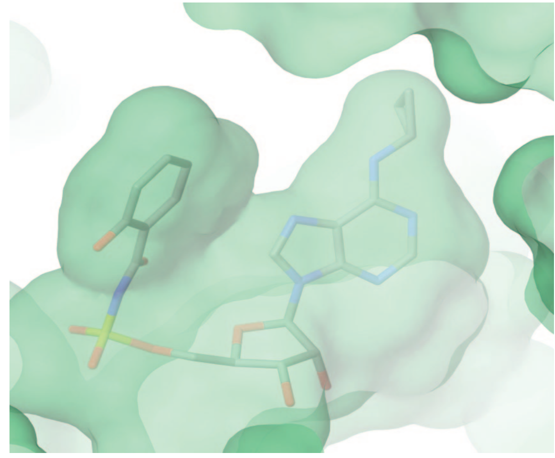


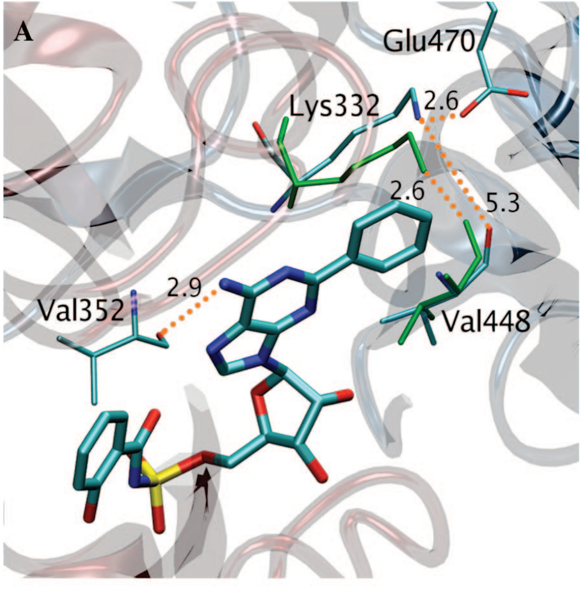
Figure 3. Compound **16** (colored by element, hydrogens not shown) is superimposed on a molecular surface representation of MbtA. The binding pocket and N6 cleft are clearly visible. Substituents larger than three carbons extend beyond the available volume and conflict sterically with the protein.

Section). The active sites are 76% identical, and both show a small unexploited pocket vicinal to N-6 (purine moiety of **6**), as well as a larger cavity near N-3 adjacent to the linker that connects the N- and C-terminal domains.[15] Additionally, a 30 ns molecular dynamics simulation of MbtA showed a hydrogen bond between the Lys332 ζ-amino and Val448 backbone carbonyl that repeatedly breaks and re-forms, increasing the space available near C-2 when broken (see Figures S1–S4, Supporting Information). Finally, the purine/protein interactions are largely hydrophobic, with no major electrostatic interactions present for N-1 and N-3 and only poorly aligned hydrogen bonds between N-7 and the NH of Gly330 and Gly354. On the basis of this analysis, a comprehensive series of 23 compounds (**7**–**29**, Figure 2) were designed to explore the importance of the purine N-1, N-3, and N-7 atoms, the role of the exocyclic N-6 amino group, and the impact of substitution at C-2 and C-8. These structures were docked and analyzed within the MbtA homology model.

In the N-6 alkylamino analogues series, docking of compounds **11**–**14** resulted in purine heavy atom rmsd values of less than 0.8 Å relative to **6**. Compound **15** displaces these atoms over 1.8 Å, indicating that the isobutyl substituent is too large. Similarly, the cyclic analogues **16** and **17** displace the base 0.5 and 0.7 Å, respectively, while the bulkier cyclopentyl and benzyl groups in **18** and **19** result in an rmsd of 1.4 Å. Accordingly, compounds **11**–**14** and **16**–**17** retain the hydrogen bond from the purine N-6 to the carbonyl of Val352 originally observed for **6** (see Figure 4A) while compounds **9** and **10** are unable to perform this hydrogen bond. Figure 3 presents **16** superimposed on a surface representation of MbtA. The cyclopropyl group fills the N-6 cleft, while larger substituents extend beyond the binding pocket into volume occupied by the protein. The N-6 pocket is clearly limited to no more than three carbon atoms for optimal potency in agreement with the small available volume observed in the MbtA homology model.

Compounds **20** and **21** showed unfavorable ligand–ligand van der Waals contacts between the C-8 substituent and the salicyl moiety in the compact receptor-bound conformation demonstrated in Figures 3 and 4A. Tan and co-workers recently reported an innovative approach to improving selectivity for related aminoacyl adenylate inhibitors by introduction of a conformation constraint through modification at C-8.[16] However, our results suggest this strategy will be more challenging to implement for Sal-AMS.

Substitution at C-2 was initially explored with 2-iodo **23**, 2-phenylamino **24**, phenylethynyl **25**, and 2-phenyl **26** in order to occupy the additional space at this binding site predicted by molecular dynamics simulations. QM/MM studies on compound **26** predicted the binding conformation shown in Figure 4A, with the C-2 phenyl group positioned between the side chain of Lys332 and the backbone atoms of Val448. The original conformations of these two residues (in green) blocked access to this pocket, whereas the alternative conformation provides enough room for C-2 substituents such as the phenyl group in **26** or even larger groups. In particular, further space was still available in this region at either the meta or para positions of the phenyl ring in **26**; thus, a series of biphenyl derivatives **27**–**29** were evaluated (see Figure S5, Supporting Information). QM/MM studies on compounds **27**–**29** showed that substitution at the meta and para positions in *m*-biphenyl **28** and *p*-biphenyl **29** successfully access the interdomain pocket (Figure 4B). Substitution at the ortho position in *o*-biphenyl **27** crowds the N-3 pocket rather than exploiting the interdomain region in the



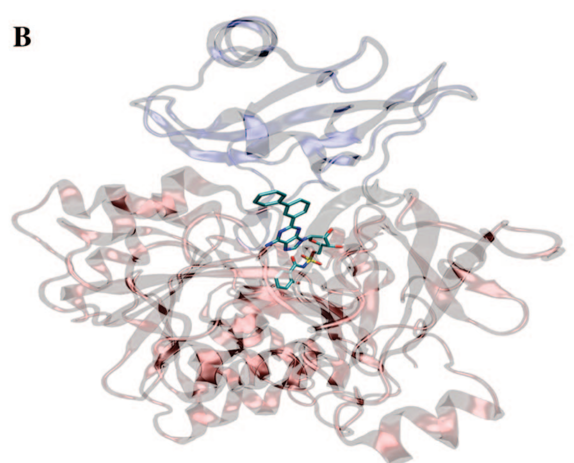


Figure 4. (A) Model of subpicomolar inhibitor **26** bound to MbtA in the predicted binding mode (QM/MM/6-31G(d)). The protein is reduced to a ribbon diagram with the N-terminal in red and C-terminal in blue. Lys332, Val352, and Val448 are shown as sticks (color by atom). The conformations of Lys332 and Val448 in the original homology model with compound **6** are shown in green, indicating the conformational shift necessary to permit binding of ligands with large C2-substituents. The hydrogen bond from N-6 to Val352 explains the ~238-fold activity difference between the N-6-dimethyl **10** and N-6-methyl **11** analogues. Distances are given in angstroms. (B) Model of *m*-biphenyl **28** bound to MbtA in the predicted binding mode (QM/MM/6-31G(d)). The protein is reduced to a ribbon diagram with the N-terminal in red (residues 1−443), C-terminal in blue (residues 455−558), and linker in gray (residues 444−454) to demonstrate the location of C2-substituents with respect to the tertiary structure of the protein.

manner of the other C-2 modifications. Steric clashes with the protein and quite possibly the purine ring indicate that large substituents in this position will result in increasingly strained conformations of the ligand and reduced potency.

Chemistry. The synthesis of 7-deaza analogue **7** commenced from tubercidin **30**, a nucleoside antibiotic produced by *Streptomyces tubercidus*[17] (Scheme 1). Protection of **30** as the acetonide followed by sulfamoylation afforded **32**.[18] Salicylation with NHS ester **33** mediated by Cs_2CO_3 and subsequent deprotection with 80% aqueous TFA furnished **7**.[14,19]

The importance of N-1 and N-3 of the purine moiety was explored with the preparation of indole analogue **8**. The synthesis of indole nucleosides in general is low yielding and

Scheme 1[a]

[a] Reaction conditions: (a) 2,2-dimethoxypropane, CSA, acetone, 88%; (b) NH_2SO_2Cl, NaH, DME, 79%; (c) **33**, Cs_2CO_3, DMF; (d) 80% aqueous TFA, 41% over two steps.

Scheme 2[a]

[a] Reaction conditions: (a) NaH, THF, 30 min, then $PhSO_2Cl$, 1.5 h, 62%; (b) $NaBH_4$, MeOH, 0 °C, 1.5 h, 100%; (c) NaH, THF, 30 min, then pivaloyl chloride, reflux, 4 h, 88%; (d) 4-nitroindole, NaH, $Pd_2(dba)_3 \cdot CHCl_3$ (0.025 equiv), P(O-*i*-Pr)$_3$ (0.20 equiv), 3 h, 55%; (e) OsO_4 (0.05 equiv), NMO (2.0 equiv), THF, 2.5 h; (f) *p*-TsOH, 2,2-dimethoxypropane, 24 h, 50% (~1:1, **39**/**40**) over two steps; (g) NaOH, H_2O/MeOH, 100 °C, 2 h, 89%; (h) NH_2SO_2Cl, DMA, 0 °C, 2.5 h, 86%; (i) **33**, Cs_2CO_3, DMF, 16 h, 77%; (j) *p*-TsOH (1.2 equiv), MeOH, 70 °C, 3 h, 46%; (k) Pd/C, H_2 (1 atm), DMF, 8 h, 44%.

problematic owing to poor anomeric control compounded by difficulty in anomer separation.[20] Therefore, we elected to replace the ribofuranosyl ring oxygen with carbon to circumvent these complications, a modification that we previously showed was well tolerated.[21] Installation of the indole moiety was accomplished via a palladium catalyzed allylic amination of **37** with 4-nitroindole that proceeded with excellent regio- and stereocontrol via a π-allyl intermediate to afford **38** in 55% yield as the only isolated coupled product (Scheme 2).[22] This innovative reaction developed by Jung and Rhee represents a powerful and efficient method for the preparation of carbocyclic nucleoside analogues. Cyclopentylamine substrate **37** was derived from Vince lactam **34**[23] in three steps. Utilization of the phenylsulfonyl group over the reported tosyl group in this

Scheme 3[a]

[a] Reaction conditions: (a) three steps, 80% overall yield;;[28] (b) 2,2′-dimethoxypropane, CSA, acetone, 76%; (c) NH_2SO_2Cl, NaH, DME, 92%; (d) **33**, Cs_2CO_3, DMF; (e) 80% aqueous TFA, 50% over two steps; (f) RNH_2 (1.5 equiv), Et_3N (3.0 equiv), EtOH, 75 °C, 5 h, sealed tube; (g) 2,2-dimethoxypropane (2.0 equiv), *p*-TsOH (1.0 equiv), acetone, 16 h, 49−96%, two steps; (h) NH_2SO_2Cl, NaH, DME, 67−91%; (i) **33**, Cs_2CO_3, DMF, 39−85%; (j) 80% aqueous TFA; (k) Dowex 50WX2-Na^+, 20−54%, two steps.

Scheme 4[a]

[a] Reaction conditions: (a) 2,2′-dimethoxypropane, CSA, acetone, 65%; (b) NaN_3, DMF, 70 °C, 12 h, 83%; (c) NH_2SO_2Cl, NaH, DME, 48−85%; (d) **33**, Cs_2CO_3, DMF; (e) 80% aqueous TFA, 55−57% over two steps; (f) Pd/C, H_2 (1 atm), MeOH, 2 h, 73%.

Scheme 5[a]

[a] Reaction conditions: (a) ref 32; (b) $PdCl_2(PPh_3)_2$ (0.1 equiv), CuI (0.2 equiv), Et_3N (2.1 equiv), DMF, 80 °C, 16 h, 57%; (c) ref 32; (d) 2-, 3-, or 4-biphenylboronic acid (2.0 equiv), $Pd(OAc)_2$ (0.2 equiv), 2-(biphenyl)dicyclohexylphosphine (0.3 equiv), K_3PO_4 (2.5 equiv), 100 °C, 12 h, 58−68%; (e) NH_2SO_2Cl, NaH, DME, 37−90%; (f) NH_2SO_2Cl, DMA, 0 °C, 3 h, 96−97%; (g) **33**, Cs_2CO_3, DMF; (h) 80% aqueous TFA, 45−72% over two steps.

sequence (**34** → **37**) enabled improved overall yields (55% vs 42%).[22,24] Dihydroxylation of **38** followed by acetonide protection provided a separable mixture of diasteromeric carbocyclic nucleosides **39** and **40** in an approximately 1:1 ratio.[25] The relative stereochemistry was confirmed by NOE studies. Sulfamoylation of **39** with sulfamoyl chloride in DMA according to the Okada protocol[26] followed by salicylation with **33** provided **43**. Attempted acetonide deprotection employing 80% aqueous TFA afforded a deep-purple solution and concomitant decomposition likely due to acid catalyzed indole dimerization. Milder deprotection conditions with *p*-TsOH in MeOH successfully provided **44**. The electron withdrawing nitro group served to deactivate the indole moiety during the acid mediated acetonide deprotection and dictated the order of the last two reaction steps. Finally, hydrogenation of the nitro moiety of **44** with Pd/C in EtOAc provided **8**, which was isolated as the triethylammonium salt. Notably, hydrogenation of **44** in MeOH led to major byproducts arising from mono- and dimethylation of the amino group.[27]

Synthesis of N-6 substituted derivatives was achieved via a common reaction sequence (Scheme 3). The key starting material was prepared from inosine in three steps by sequential trifluoroacetylation, chlorination, and methanolysis as described to provide **48**.[28] Amination of **48** was performed with 3.0 equiv of triethylamine in EtOH in a sealed tube at 70 °C.[29] For very low molecular weight amines (methylamine, ethylamine, cyclopropylamine) solvolysis without Et_3N was preferred. The crude nucleosides were directly converted to the corresponding acetonides by treatment with *p*-TsOH and 2,2-dimethoxpropane in acetone to afford nucleosides **49**−**58**. Sulfamoylation provided **59**−**68** and subsequent salicylation with **33** followed by acetonide deprotection with 80% aqueous TFA furnished **10**−**19** isolated as the triethylammonium salts that were converted to the sodium salts by ion exchange with a strong cation exchange resin in the sodium form. Derivative **9** was prepared from inosine in four steps in analogy to the preparation of **7** (Scheme 3).

The impact of substitution at C-8 of the purine moiety was assessed with the preparation of **20**−**22** (Scheme 4). 8-Bromoadenosine **69** was protected as acetonide **70**, which was converted to 8-azido **71** by treatment with NaN_3 in DMF (Scheme 4).[30] Both **70** and **71** were elaborated to the corresponding sulfamates **72** and **73** and then salicylated with **33** and deprotected to provide **20** and **21** in analogy to the preparation of **7**. Hydrogenation of **21** afforded 8-amino derivative **22**.

Modification of C-2 was most efficiently achieved from 2-iodoadenosine, which enabled installation of various substituents via standard palladium catalyzed reactions as described below (Scheme 5). 2-Iodo-2,3-*O*-isopropylieneadenosine **75** was prepared in five steps from guanosine following the Matsuda

Table 1. Inhibition of MbtA and Whole Cell *M. tuberculosis* H37Rv

Z = O, CH_2[a]

Compound, B =	K_I^{app} (nM)[b]	MIC_{99} (μM)[c] (iron-deficient)	MIC_{99} (μM)[d] (iron-replete)	S $\left(\frac{MIC_{99}^{+Fe}}{MIC_{99}^{-Fe}}\right)$
6, X = Y = N	6.6 ± 1.5	0.39	1.56	4
7, X = N; Y = CH	24.3 ± 0.7	6.25	50	8
8, X = Y= CH	20.1 ± 2.3	6.25	50	8
R = **9**, =O[f]	800 ± 50	>100	>100	n.d.[e]
10 –NMe$_2$	380 ± 32	50	n.d.	n.d.
11, –NHMe	1.60 ± 0.20	0.39	1.56	4
12, –NHEt	4.40 ± 0.08	0.39	1.56	4
13, –NH*n*-Pr	3.50 ± 0.20	0.39	1.56	4
14, –NH*i*-Pr	7.00 ± 0.90	25	>100	>4
15, –NH*i*-Bu	1140 ± 230	>100	>100	n.d.
16, –NHcyclopropyl	1.85 ± 0.13	0.098	6.25	64
17, –NHcyclobutyl	124 ± 12	>100	>100	n.d.
18, –NHcyclopentyl	9412 ± 520	>100	>100	n.d.
19, –NHBn	8260 ± 820	>100	>100	n.d.
R = **20**, –Br	(2.05 ± 0.30)·10^3	>50	>50	n.d.
21, –N$_3$	(42.5 ± 6.2)·10^3	>50	>50	n.d.
22, –NH$_2$	(183 ± 19)·10^3	>50	>50	n.d.
R = **23**, –I	3.03 ± 0.34	0.19	3.12	16
24, –NHPh	0.94 ± 0.16	0.049	0.39	8
25, –CCPh	0.40 ± 0.05	0.049	0.39	8
26, –Ph	0.27 ± 0.07	0.049	0.39	8
27, biphen-2-yl	47 ± 2	>50	>50	n.d.
28, biphen-3-yl	1.4 ± 0.2	25–50	>50	n.d.
29, biphen-4-yl	0.99 ± 0.02	25–50	>50	n.d.

[a] Z = O for compounds **6**, **7**, **9**−**29**; Z = CH_2 for compound **8**. [b] Assay performed with 7 nM MbtA, 10 mM ATP, 250 μM salicylic acid, 1 mM PP_i. [c] Grown in normal pH 6.6 glycerol−alanine−salts (GAS) medium without ferric ammonium citrate. [d] Grown in normal pH 6.6 glycerol−alanine−salts (GAS) medium supplemented with 200 μM ferric ammonium citrate. [e] n.d.: not determined. [f] N-1 of the purine is protonated.

protocol.[31] Buchwald−Hartwig coupling of **75** with aniline afforded the phenylamino derivative **76**.[32] Similarly, Sonogashira coupling of **75** with phenylacetylene furnished **77** and Suzuki coupling of **75** with phenylboronic acid as well as 2-, 3-, and 4-biphenylboronic acid provided **78**−**81**, respectively.[32] The palladium-mediated reactions required a 20 mol % catalyst loading in order to obtain complete conversion. We speculate the moderate yields observed in these palladium-catalyzed reactions is likely due to coordination of Pd(II) to N^1 and N^7 of the purine.[33] Such bidentate complexes are expected to have lower catalytic activity and may contribute to deglycosylation of the nucleoside. Elaboration of nucleosides **75**−**81** to sulfamates **82**−**88** and salicylation and subsequent deprotection to **23**−**29** were carried out in analogy to the preparation of **7**.

MbtA Inhibition. Inhibition of MbtA was performed using an ATP−PP_i exchange assay as previously described[21] (see Experimental Section). Assays were performed at 37 °C with recombinant MbtA expressed in *E. coli* in a buffer of 75 mM Tris-HCl, pH 7.5, 10 mM $MgCl_2$, 2 mM DTT, 250 μM salicylic acid, 10 mM ATP, and 1 mM PP_i.[21] The initial rates of pyrophosphate exchange (≤10% reaction) were monitored by measuring the amount of [^{32}P]ATP formed after addition of [^{32}P]PP_i. The apparent inhibition constants (K_i^{app}) were determined by fitting the concentration−response plots either to the Hill equation (eq 1; see Experimental Section) or to the Morrison equation (eq 2; see Experimental Section). The Morrison equation was employed in cases where the inhibitors exhibited tight-binding behavior ($K_i^{app} \leq 100 \times [E]$).

The K_i^{app} of the parent compound **6** has been previously determined to be 6.6 nM (Table 1).[21] The impact of N-7 was evaluated with 7-deaza-analogue **7**, which resulted in a 4-fold loss of potency, consistent with loss of the hydrogen bonds between N-7 and the NH of Gly330 and Gly354 (Table 1). Removal of N-1 and N-3 in indole derivative **8** demonstrated that these nitrogens are dispensable for activity.

Inosine derivative **9** and dimethylamino **10** led to a 121-fold and 58-fold loss of potencies respectively, establishing the requirement for at least one hydrogen bond donor appended at C-6. Therefore, a systematic series of monoalkyl and cycloalkyl N-substituted analogues **11**−**19** were synthesized revealing that small C1−C3 alkyl groups were well tolerated, as predicted by the homology model, which showed a small hydrophobic pocket in the protein structure vicinal to N-6. Cyclopropyl **16** was the most potent analogue in this series with a 3.5-fold enhancement in binding affinity. The potency rapidly diminished as the alkyl chain increased beyond C-3; thus, cyclobutyl **17** and isobutyl **15** exhibited a 124- and 1140-fold loss in potencies, respectively. Further increases in chain length with **18** and **19** led to an approximately 10,000-fold decrease in binding affinity.

Inhibitors **20**−**22** bearing substitution at C-8 resulted in 300- to 30,000-fold loss potency concordant with predictions from molecular modeling. Additionally, the propensity of bulky groups at C-8 of adenine to favor the syn conformation about the glycosidic linkage could account for part of the reduced activity, since the inhibitor must bind in an anti conformation to MbtA.[34]

Substitution at C-2 was initially explored with 2-iodo **23**, which exhibited a 2-fold increase in affinity. Encouraged by this result, a small series of analogues were synthesized by various cross-coupling reactions to provide 2-phenylamino **24**, phenylethynyl **25**, and 2-phenyl **26**. These compounds were 7, 17, and 24 times more potent than **6**. Modeling studies predicted that additional room at this binding site was available at either the meta or para positions of the phenyl ring in **26**. Accordingly biphenyl derivatives **27**−**29** were prepared and found to exhibit the following trend: meta ∼ para > ortho. The 174-fold loss in potency of **27** compared to **26** is consistent with a steric clash of the biphenyl with backbone residues in the N-terminal domain, while the meta and para phenyl groups successfully access the interdomain pocket. The ability to tolerate these bulky nonpolar substituents at C-2 was remarkable but consistent with our molecular dynamics simulations, which showed a high degree of flexibility in this region.

Isothermal Calorimetry. The most potent ligand, 2-phenyl-Sal-AMS **26**, was further characterized and shown to be an enthalpy-driven reversible competitive inhibitor with respect to salicylic acid and ATP. Notably, the reported K_i^{app} values vastly underestimate potency of these bisubstrate inhibitors, since the K_i^{app} values were determined under supersaturating concentrations of both substrates salicylic acid (250 μM or $120K_M^{(Sal)}$) and ATP (10 mM or $55K_M^{(ATP)}$). Attempts to determine its true inhibition constant (K_i) were complicated by the tight-binding behavior and the bisubstrate nature of inhibition, which precluded K_i assessment by traditional steady state kinetic methods.[35] Direct measurement of the dissociation constant (K_D) was obtained by isothermal calorimetry in the presence of saturating salicylic acid at 298 K, which afforded K_D, ΔH, and $-T\Delta S$ values of 0.26 ± 0.23 pM, −15.9 ± 0.6 kcal/mol, and −1.6 ± 1.4 kcal/mol, respectively.

Antitubercular Activity. The antitubercular activity of these compounds was evaluated against whole-cell *M. tuberculosis* H37Rv under iron-deficient and iron-replete conditions. The whole cell assay permits the assessment of *M. tuberculosis* inherent resistance because of factors such as limited membrane permeability. Results are given in Table 1, in the form of MIC_{99}, the minimum concentration of compound that completely inhibited observable growth of *M. tuberculosis* H37Rv.

Overall the MIC_{99} values are well correlated with the K_i^{app} values and analogues **24**−**26** exhibited the most potent whole-cell antitubercular activity yet reported for this new class of antibacterial agents (MIC_{99} of 49 nM in iron-depleted conditions), with good selectivity. This result compares favorably against isoniazid, a first-line antitubercular drug that displayed an MIC_{99} of 0.18 μM under the assay conditions.[14] Even more significantly, cyclopropyl analogue **16** displayed enhanced selectivity with an MIC_{99} of 98 nM under iron-deficient conditions but 6.25 μM under iron-rich conditions, representing a 64-fold difference in activity, a phenotype concordant with the designed mechanism of action. The selectivity factor, *S*, in Table 1 is defined as the ratio of MIC_{99} values in iron-replete and iron-deficient conditions. Thus, cyclopropyl derivative **16** provides a 16-fold enhancement in selectivity relative to Sal-AMS **6**.

Cell Cytotoxicity. To assess potential toxicity, all of the compounds were evaluated against Vero cells using an MTT assay and found not to inhibit cell growth (IC_{50} > 100 μM), providing a therapeutic index (IC_{50}/MIC_{99}) greater than 2000 for **24**−**26**.

Mechanism of Action Studies. In an effort to delineate the putative secondary mechanism of action of these adenylation inhibitors, **6** was evaluated for enzyme inhibition against a panel of adenylating enzymes (FadD17, FadD19, FadD26, and FadD28) involved in fatty acid metabolism in *M. tuberculosis* (see Experimental Section).[36] However, **6** showed no inhibition of any of the four FadDs evaluated at 100 μM. MbtA is functionally related to aminoacyl t-RNA synthetases involved in protein synthesis. Therefore, **6** and 5′-*O*-(sulfamoyl)adenosine,[37] a potent inhibitor of protein synthesis, were evaluated for inhibition of protein synthesis using an in vitro protein translation assay (see Experimental Section).[37] Remarkably, **6** exhibited no protein inhibition (IC_{50} > 100 μM) despite the structural similarity to 5′-*O*-(sulfamoyl)adenosine.

Discussion

Given the importance of mycobacterial iron metabolism and the critical function of the mycobactins, the design of small molecule siderophore biosynthesis inhibitors represents a novel, promising strategy to develop new antitubercular agents.[4,12–14,21] An alternative approach developed by Miller and co-workers involves the synthesis of siderophore analogues as potential antagonist of siderophore function. Indeed a full-length mycobactin analogue was found to have potent activity with an MIC of 0.2 μg/mL.[38] Quadri and co-workers recently identified a series of thiacarbamoylpyrazoline analogues designed as small molecule mimics of the terminal salicyl-oxazoline cap of mycobactin with several compounds displaying low micromolar antitubercular activity and selectivity under iron deficient conditions.[39] Finally, it is noted that a conceptually related and very successful approach also pioneered by Miller and co-workers involves the synthesis of siderophore−antibiotic conjugates that exploit siderophore receptors for uptake and delivery of the antibiotics.[40]

In this report, we have used molecular modeling to guide modifications to the nucleobase domain of Sal-AMS **6**. Deletion of N-1 or N-3 in the purine ring was tolerated, while removal of N-7 led to a slight decrease in inhibitor potency. We found that at least one hydrogen bond donor at N-6 is essential for activity of this class of compounds, as inosine **9** and dimethyl **10** analogues exhibited pronounced reductions in potency. Substitution of N-6 with C1−C3 chains was beneficial leading to incremental increases in potency reaching a maximum with cyclopropyl **16**. The potency followed a logarithmic decline as the chain length increased to C4 and C5, then leveled off at C5. Although compounds **11**−**13** exhibited the same antitubercular activity as the parent compound **6**, the N-6 cyclopropyl analogue **16** showed a significant improvement in biological activity. Additionally, **16** displayed the best selectivity of any Sal-AMS derivative investigated with a 64-fold difference in antitubercular activity between iron-deficient and iron-rich conditions. The C-8 modified analogues **20**−**22** exhibited drastically reduced potency, illustrating the exquisite sensitivity to modification at this position that had been predicted by our homology model. The most potent compounds, both in the in vitro and the whole-cell assays, were in the C-2 modified series, with MbtA showing a remarkable ability to tolerate bulky nonpolar substituents in this position. By contrast, relatively modest changes to the salicyl, sulfamoyl, and ribosyl subunits

of **6** have in the majority of cases resulted in drastic losses in binding affinity.[21,41–44] The high selectivity observed for several potent MbtA inhibitors, including compound **16** but also compounds **10–13** and **24–26**, which displayed potent submicromolar MIC_{99} values, makes these compounds very promising antitubercular agents.

Our SAR results using a bisubstrate inhibitor are consistent with the extensive substrate specificity studies of aminoacyl adenylation domains embedded in multifunctional nonribosomal peptide synthetases (NRPSs) reported by von Döhren and co-workers using a series of ATP analogues.[45] Significantly, ATP analogues modified at the C-2 position of adenine were competent substrates whereas C-8 modified analogues were not accepted by the various NRPSs investigated. These co-workers also demonstrated that deletion of the N-7 nitrogen atom of adenine led to a marginal reduction in activity. Consequently, the SAR observed for MbtA, a stand-alone aryl acid adenylating enzyme (AAAE), is expected to be applicable to the abundant aminoacyl adenylation domains found in multifunctional NRPSs.

With the exception of compounds **28** and **29**, whole-cell activity of the nucleobase-modified analogues correlated nicely with the in vitro enzyme inhibition results. As previously suggested, the difference between MIC_{99} and K_i^{app} likely reflects limited permeability across the mycobacterial cell envelope, which provides intrinsic resistance to many antibiotics.[21] Furthermore, if MbtA does not catalyze the rate-limiting biosynthetic step of the mycobactins, then it may be necessary to fully abrogate MbtA activity to have an appreciable effect on mycobactin production.[21] The low whole-cell activity of compounds **28** and **29**, despite their high potency against MbtA, is most likely due to their inability to use a transporter for uptake in *M. tuberculosis*. We hypothesize that the transporter still recognizes and imports compounds such as **24–26**, with a significant change introduced by the bulky substituents in the C-2 position of the adenine ring. However, the more significant structural change caused by the introduction of a second phenyl ring in compounds **27–29** probably prevents recognition by these putative transporters. *M. tuberculosis* encodes several ABC transporters (ATP-depending binding cassette) that have been unambiguously assigned as nucleoside importers.[46] Other candidate transporters include porins such as MshA and the mycobactin transporters IrtAB.[47,48]

Bisubstrate Inhibition. Sal-AMS and the derivatives described herein are considered bisubstrate inhibitors, since these simultaneously occupy both salicylic acid and nucleoside binding sites.[49] Bisubstrate inhibitors (A–B) can realize substantial enhancement in binding energy compared to the sum of the Gibbs binding energies of the respective fragments (A and B) as first articulated by Jencks because of a smaller entropy barrier to binding of A–B compared to A + B.[50] In most cases, bisubstrate inhibitors fail to even display simple additivity in binding, as linking each fragment typically results in loss of some interaction energy with the corresponding enzyme. In the present case, the acylsulfamate closely mimics the native acylphosphate linkage maintaining all of the key H-bond and electrostatic interactions with the active site. Isothermal calorimetry revealed the true potency of these inhibitors showing that 2-phenyl **26** binds with an astonishing K_D of 0.27 pM or $\Delta G^0 = -17.5$ kcal/mol wherein binding is primarily enthalpy driven with a minor entropic contribution. The Gibbs binding energy of **26** is 4 kcal/mol more than the sum of the free energy of binding of the respective acyladenylate fragments salicylic acid and AMP ($\Delta G^0_{AMP} = -5.0$ kcal/mol, $\Delta G^0_{Sal} = -7.9$ kcal/mol).[51] By contrast, Callahan, Wolfenden and co-workers have shown that a related bisubstrate inhibitor incorporating an acylhydroxamoylsulfamate linkage, which is approximately 2 Å longer than the acylsulfamate linkages employed in Sal-AMS, displayed simple additivity in binding relative to the corresponding fragments AMP and 2,3-dihydroxybenzoic acid toward the adenylation enzyme EntE involved in biosynthesis of the enterobactin siderophore in *E. coli*.[52]

The tight-binding of the intermediate acyladenylate (Sal-AMP, **2**) is necessary to prevent diffusional loss to the bulk solvent and adventitious hydrolysis of the mixed carboxylic–phosphoric acid anhydride, since this intermediate must be channeled to a downstream protein. Unlike the stand-alone aryl adenylating enzyme MbtA, most NRPS adenylation domains are embedded with carrier domains in multifunctional proteins so that transfer to downstream carrier domains is entropically favored.[53–55] Further, many adenylating enzymes such as CoA ligases employ abundant diffusible small molecules. In both cases, tight binding of the intermediate acyladenylate may not be necessary because the effective concentration of the nucleophilic acceptor residue is expected to be significantly higher. Consequently, simple bisubstrate inhibitors toward such enzymes are likely to be intrinsically less potent. Indeed the K_i^{app} value of cysteyl-AMS for the cysteine adenylation domain in the multifunctional NRPS module known as HMWP2 from *Yersinia pestis* is only 0.24 μM.[16]

Mechanism of Action. Siderophore biosynthesis is not required for growth under iron-replete conditions; thus, antitubercular activity under these conditions suggests off-target binding and subsequent inhibition of alternate biochemical pathways.[12] Despite the off-target binding of the nucleoside derivatives described herein, these compounds remain very promising antitubercular agents. Antitubercular agents that disrupt multiple biosynthetic pathways may actually be preferred over single-target therapeutics. Coincidently, many of the currently used antitubercular agents including isoniazid,[56] ethionamide, *p*-aminosalicylic acid, and cycloserine have been implicated to possess multiple mechanisms of action.[57] *M. tuberculosis* encodes for more than 60 adenylation enzymes that represent potential off-target candidates proteins including 20 aminoacyl tRNA synthetases involved in protein synthesis,[58] 34 FadDs involved in lipid metabolism,[36] MshC that catalyzes the third step in mycothiol biosynthesis,[59] PanC that catalyzes the second step in biosynthesis of coenzyme A (CoASH),[60] LigA, a NAD-dependent DNA ligase,[61] and 5 adenylation domains embedded in NRPS modules from the mycobactin-1[11] and nrp loci.[62] Several of the most logical potential candidates were evaluated including the aminoacyl tRNA synthetases involved in protein synthesis as well as a panel of fatty acid adenylating enzymes required for lipid metabolism. Significantly, Sal-AMS did not inhibit the functionally related aminoacyl tRNA synthetases using an in vitro luciferase translation assay. Additionally, we evaluated the parent compound Sal-AMS **6** against a panel of fatty acid adenylating (FadD) enzymes, which carry out the analogous adenylation of fatty acids in *M. tuberculosis*.[36] The FadDs can be grouped into two classes: acyl-CoA synthetases (ACSs) involved in fatty acid catabolism; long chain fatty acyl-AMP ligases involved in fatty acid biosynthesis. The long chain acyl-AMP ligases FadD26 and FadD28 were investigated because these are involved in production of the dimycocerosyl phthiocerols (PDIM), which are important virulence factors.[36,63–65] Similarly, the acyl-CoA synthetases FadD17 and FadD19 were investigated, since these have been implicated in mycobacterial persistence via catabolism of cholesterol.[66] The in vitro findings that **6** was inactive against

all four of the FadDs evaluated clearly demonstrate a promising level of selectivity. Future efforts will explore systematic proteome profiling of adenylating enzymes to help identify potential off-target protein targets and measure selectivity.

Other Pathogens That Require Aryl-Capped Siderophores. Although we have focused on *M. tuberculosis*, many other pathogens utilize related aryl-capped siderophores for iron acquisition and we expect AAAE inhibitors to be effective against a broad array of clinically relevant pathogens.[67] For example, *Yersinia pestis* (plague) and *Klebsiella pneumoniae* (opportunistic infections) synthesize yersiniabactin,[68,69] *Bacillus anthracis* (anthrax) makes petrobactin,[70,71] enteric bacteria such as *Escherischia coli* and *Salmonella enterica* serovar Typhimurium produce enterobactin,[72,73] *Vibrio cholerae* (cholera) synthesizes vibriobactin,[74] *Vibrio vulnificus* (septic infections) synthesizes vulnibactin,[75] *Pseudomonas aeruginosa* (opportunistic infections) makes pyochelin,[76] and *Acinetobacter baumannii* (opportunistic infections) produces acinetobactin.[77] The requirement for siderophore synthesis has been demonstrated to be essential for virulence in in vivo models for invasive pathogens such as *Y. pestis*, *K. pneumoniae*, *V. vulnificus*, *B. anthracis*, and *S. typhimurium*.[68–70,75] Surface pathogens such as *V. cholera* and *P. aeruginosa* employ redundant mechanisms of iron acquisition so that the aryl-capped siderophores are not required for virulence.[78,79] Adenylation inhibitors based on the prototypical Sal-AMS template have been shown to inhibit the adenylating enzymes AsbC,[71] EntE,[52] DhbE,[13] and YbtE[12] from *B. anthracis*, *E. coli*, *B. subtilis*, and *Y. pestis*. Additionally, in vitro data against *Y. pestis* and *Y. pseudotuberculosis* indicated that Sal-AMS and derivatives possesses activity against these Gram-negative organisms.[12,21]

Conclusion

Molecular modeling studies were used to guide modifications to the nucleobase of the parent compound Sal-AMS **6** and clear trends in the SAR data emerged demonstrating the remarkable ability to tolerate bulky substituents at the C-2 position and small alkyl substituents appended at N-6 of the purine but not modification at the C-8 position. The comprehensive series of analogues confirmed that N-1 and N-3 were dispensable for activity, while deletion of N-7 results in modest decreases in potency and biological activity. N-6 cyclopropyl **16** was found to exhibit the highest selectivity against *M. tuberculosis* under iron-rich conditions. Isothermal calorimetry revealed the intrinsic potency of this class of inhibitors with 2-phenyl **26** displaying femtomolar enzyme inhibition and the most potent antitubercular activity yet observed for this class of antitubercular agents. Attempts to delineate the putative secondary mechanism of action of this class of compounds under iron-rich conditions revealed that Sal-AMS does not inhibit the functionally related aminoacyl t-RNA synthetases or several mycobacterial fatty acid adenylating enzymes (FadDs) involved in lipid metabolism. Further, these compounds displayed no overt cytotoxicity providing large therapeutic indexes. Recent studies have shown that Sal-AMS **6** is stable in human plasma and human liver microsomes.[80] Taken together, these results provide a promising profile for the described 5′-*O*-[*N*-(salicyl)sulfamoyl]nucleoside inhibitors and the modifications described herein have enabled significant improvements in potency, selectivity, and lipophilicity of this new class of antitubercular agents. Nucleoside inhibitors based on Sal-AMS have great potential as novel antibacterial agents targeting siderophore biosynthesis. More than 500 structurally different siderophores have been isolated. Thus, the development of a broad-spectrum agent targeting siderophore synthesis is not possible. However, the bisubstrate nucleoside inhibitors described herein hold promise to provide agents with a narrow therapeutic window against pathogens such as *M. tuberculosis* that require aryl-capped siderophores for virulence, which may in fact be advantageous since it is expected that beneficial commensal bacteria will not be disturbed.

Experimental Section

Molecular Modeling. A homology model of MbtA was constructed on the basis of the crystal structure of the 2,3-dihydroxybenzoic acid (DHB) adenylating enzyme DhbE (PDB code 1MDB[15]) in complex with the adenylated reaction product of 2,3-dihydroxy acid (2,3-DHB-AMP), a close analogue of **2**, which serves as the rational basis for 5′-*O*-[*N*-(salicyl)sulfamoyl]adenosine **6**. A primary sequence alignment was generated for DhbE and MbtA with ClustalW,[81] examined, and edited by hand. Modeller 9v1[82] was used to generate 10 conformations in the presence of **2** obtained by deleting the 3-hydroxyl from the crystal structure. After examination, a structure with one of the lowest DOPE and highest objective scores (Modeller structure rankings) was chosen because of the reasonable positioning of active site side chains. Hydrogens were added and minimized with Macromodel 9.1[83] and the MMFFs[84] force field in a GB/SA[85] implicit solvation model. A 130 000 step conformational search including 18 key active site side chains and the ligand was undertaken to explore the translational and torsional energy surface of residues near the binding pocket. In recognition of the 76% identical active sites, select hydrogen bonding interactions were restrained through the addition of harmonic constraints with a large penalty for distances outside a 2.7−3.5 Å range. A structure that differed from the global minimum by 1.5 kcal/mol and a torsion of the Asp436 (DhbE Asp413) side chain was chosen for future modeling studies because it positions the aspartic acid side chain such that it doubly hydrogen-bonds the ribosyl moiety (one side chain oxygen hydrogen-bonded to the 2′-hydroxyl, the other to the 3′-hydroxyl) in agreement with the DhbE crystal structure.

Molecular Dynamics. The MbtA homology model, with the ligand absent, was prepared for molecular dynamics simulation using VMD.[86] Charges were neutralized through addition of Na^+ and Cl^- ions, and the model was soaked in a water box extending at least 15 Å beyond any atom of the structure in each direction. The entire model was first minimized in NAMD,[87] using the CHARMM[88,89] force field, and then warmed from a temperature of 5 to 300 K over 60 ps. Following the model preparation a 32 ns simulation was completed.

QM/MM Studies. QM/MM geometry optimizations were performed in Qsite[90] using the homology model described above. Ligands were treated at the B3LYP[91–93]/6-31G(d)[94,95] level of theory polarized by OPLS 2005[96] charges in the positions of the protein atoms. The lack of covalent bonds between the two regions circumvented the problem of treating link atoms. Both ligand and protein were allowed to minimize freely.

Chemistry General Procedures. All commercial reagents (Sigma-Aldrich, Acros) were used as provided unless otherwise indicated. Tubercidin **30** was kindly provided by Prof. Herbert Nagasawa (VA Medical Center, Minneapolis, MN). 8-Bromoadenosine was obtained from Berry and Associates (Dexter, MI). Sulfamoyl chloride was prepared by the method of Heacock except that this was used directly without recrystallization.[97] *N*-Hydroxysuccinimidyl 2-(methoxymethoxy)benzoate **33** was prepared as described.[21] An anhydrous solvent dispensing system (J. C. Meyer) using two packed columns of neutral alumina was used for drying THF, DMF, and CH_2Cl_2, and the solvents were dispensed under argon. Anhydrous DME was purchased from Aldrich and used as provided. Flash chromatography was performed with silica P grade silica gel 60 (Silicycle) or with an ISCO Combiflash Companion purification system with the prepacked silica gel cartridges with the indicated solvent system. All reactions were performed under an inert atmosphere of dry Ar or N_2 in oven-dried (150 °C) glassware. ^{1}H and ^{13}C NMR spectra were recorded on a Varian

600 MHz spectrometer. Proton chemical shifts are reported in ppm from an internal standard of residual chloroform (7.26 ppm) or methanol (3.31 ppm), and carbon chemical shifts are reported using an internal standard of residual chloroform (77.0 ppm) or methanol (49.1 ppm). Proton chemical data are reported as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, p = pentet, m = multiplet, br = broad), coupling constant, integration. High resolution mass spectra were obtained on Agilent TOF II TOF/MS instrument equipped with either an ESI or APCI interface. Optical rotations were measured on a Rudolph Autopol III polarimeter. Melting points were measured on an electrothermal Mel-Temp manual melting point apparatus and are uncorrected. Preparative HPLC was performed on a Varian Microsorb MV 100-8 C18 column (41.4 mm × 250 mm, 8 μm particle size) operating at 40 mL/min with detection at 254 nm and the following mobile phases (solvent A, 50 mM $Et_3NH \cdot HCO_3$; solvent B1, 9:1 MeCN/H_2O; solvent B2, MeOH) and the indicated HPLC conditions (methods 1 and 2). Method 1 involves elution with solvent A and 20−100% linear gradient of solvent B1 over 30 min. Method 2 involves elution with solvent A and 20−100% linear gradient of solvent B2 over 30 min. HPLC purity of synthesized compounds was determined using a Varian Microsorb MV 100-8 C18 column (4.6 mm × 250 mm, 8 μm particle size) operating at 0.5 mL/min with detection at 254 nm and the conditions described in the Supporting Information.

General Procedure for Acetonide Protection. A mixture of nucleoside (1.0 equiv), 2,2-dimethoxypropane (18 equiv), and camphorsulfonic acid·H_2O (1.0 equiv) in acetone (0.1 M) was stirred at room temperature for 16 h. Solid $NaHCO_3$ (3.0 equiv) was added, and the heterogeneous mixture was stirred for 30 min. The mixture was then partitioned between $CHCl_3$ (25 mL/mmol) and H_2O (10 mL/mmol). The aqueous layer was back-extracted with $CHCl_3$ (3 × 25 mL), and the combined organic extracts were dried (Na_2SO_4) and concentrated. Purification by flash chromatography (EtOAc/hexane) afforded the title compound.

General One-Pot Procedure for N^6-Amination and Isopropylidene Protection. To a suspension of **45** (1.0 equiv) in ethanol (10 mL/mmol) in a glass pressure vessel were added Et_3N (3.0 equiv) and the amine (1.5 equiv). The glass tube was sealed with the Teflon screw cap and heated at 75 °C for 5−6 h. The pressure vessel was cooled to room temperature, and the contents were transferred to a round-bottom flask and concentrated and dried thoroughly under high vacuum to remove any residual amine. The resulting solid was suspended in acetone (5 mL/mmol). Dimethoxypropane (2.0 equiv) and *p*-toluenesulfonic acid (1.0 equiv) were added, and the mixture was stirred 16 h at room temperature. After completion solid $NaHCO_3$ (3.0 equiv) was added and the heterogeneous mixture stirred for 30 min. The mixture was then partitioned between $CHCl_3$ (75 mL/mmol) and H_2O (15 mL/mmol). The aqueous layer was back-extracted with $CHCl_3$ (3 × 25 mL), and the combined organic extracts were dried (Na_2SO_4) and concentrated. Purification by flash chromatography (EtOAc/hexane) afforded the title compound.

General Procedure for Sulfamoylation. To a solution of 2′,3′-*O*-isopropylidene protected nucleoside (1 equiv) in DME/THF (1:1, 50 mL/mmol) at 0 °C was added NaH (1.5 equiv). After 30 min, solid sulfamoyl chloride (1.5 equiv) was added and the mixture stirred for 16 h at room temperature. The reaction was quenched at 0 °C with MeOH (30 mL/mmol) and then concentrated under reduced pressure. Purification by flash chromatography afforded the title compound.

General Procedure for Salicylation. To a solution of 2′,3′-*O*-isopropylidene-5′-*O*-sulfamoyl-nucleoside (1 equiv) in DMF (0.1 M) at 0 °C were added **33** (1.5 equiv) and Cs_2CO_3 (3.0 equiv), and the mixture was stirred for 16 h at room temperature. The mixture was filtered and then concentrated by rotary evaporation ($P \approx 5$ mbar, water bath ∼35 °C). Purification by flash chromatography (10−20% MeOH/EtOAc + 1% Et_3N) afforded the title compound.

General Procedure for TFA Deprotection. To a solution of 5′-*O*-{*N*-[2-methoxymethoxy)benzoyl]sulfamoyl}-2′,3′-*O*-isopropylidene-nucleoside (0.2 mmol) was added 80% aqueous TFA (2.5 mL). The resulting solution was stirred for 30 min at 0 °C and then concentrated under reduced pressure. Purification by flash chromatography (EtOAc/MeOH/Et_3N) afforded the title compound.

General Procedure for Ion Exchange. A solution of a nucleoside triethylammonium salt (0.25 mmol, 1 equiv) in H_2O (0.5 mL) was added to a short column (10 mm × 50 mm, Dowex 50WX8-100-Na^+) and incubated for 10 min before eluting with H_2O (20 mL). The fractions containing the product were lyophilized to afford the sodium salt as a flocculent white solid. The Dowex cation exchange resin was converted to the sodium form by sequentially washing the column with MeOH (50 mL), H_2O (50 mL), 1 N aqueous NaOH (25 mL), and H_2O (100 mL).

7-Deaza-2′,3′-*O*-isopropylideneadenosine (31). 7-Deazaadenosine (tubercidin) **30** (300 mg, 1.12 mmol, 1.00 equiv) was converted to the title compound using the general procedure for acetonide protection. Purification by flash chromatography (99:1 EtOAc/MeOH) afforded the title compound (304 mg, 88%) as a colorless oil: R_f = 0.6 (3:17 MeOH/EtOAc); $[\alpha]_D^{20}$ −220 (*c* 0.230, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.33 (s, 3 H), 1.58 (s, 3H), 3.68 (dd, *J* = 12.0, 4.2 Hz, 1H), 3.76 (dd, *J* = 12.0, 3.0 Hz, 1H), 4.25 (app q, *J* = 3.6 Hz, 1H), 4.96 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.12 (dd, *J* = 6.0, 4.2 Hz, 1H), 6.12 (d, *J* = 4.2 Hz, 1H), 6.57 (d, *J* = 3.6 Hz, 1H), 7.26 (d, *J* = 3.6 Hz, 1H), 8.06 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.4, 26.5, 62.4, 81.4, 83.9, 85.6, 91.5, 99.8, 104.0, 114.1, 123.3, 149.3, 151.0, 157.9; MS (APCI+) calcd for $C_{14}H_{19}N_4O_4$ $[M + H]^+$ 307.1, found 307.1.

7-Deaza-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (32). Compound **31** (360 mg, 1.18 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (200:1 EtOAc/MeOH) afforded the title compound (350 mg, 79%): R_f = 0.7 (85:15 EtOAc/MeOH); $[\alpha]_D^{20}$ −8.0 (*c* 1.2, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.40 (s, 3H), 1.63 (s, 3H), 4.28 (dd, *J* = 12.0, 4.8 Hz, 1H), 4.32 (dd, *J* = 10.2, 4.6 Hz, 1H), 4.46 (app q, *J* = 3.6 Hz, 1H), 5.08 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.23 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.32 (d, *J* = 3.0 Hz, 1H), 6.66 (d, *J* = 3.6 Hz, 1H), 7.32 (d, *J* = 3.6 Hz, 1H), 8.13 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.7, 27.6, 70.1, 82.7, 84.3, 85.6, 91.2, 101.7, 104.9, 115.8, 123.8, 151.2, 152.7, 159.1; MS (APCI+) calcd for $C_{14}H_{20}N_5O_6S$ $[M + H]^+$ 386.1, found 386.1

7-Deaza-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (7). Compound **32** (340 mg, 0.88 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded the title compound (168 mg, 41%) as a white solid: R_f = 0.3 (4:1 EtOAc/MeOH); $[\alpha]_D^{20}$ −7.71 (*c* 1.27, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.26 (t, *J* = 7.3 Hz, 9H), 3.15 (q, *J* = 7.3 Hz, 6H), 4.25−4.27 (m, 1H), 4.34−4.38 (m, 3H), 4.52 (t, *J* = 5.7 Hz, 1H), 6.26 (d, *J* = 6.3 Hz, 1H), 6.61 (d, *J* = 3.7 Hz, 1H), 6.78−6.81 (m, 2H), 7.30 (t, *J* = 7.7 Hz, 1H), 7.54 (d, *J* = 3.7 Hz, 1H), 7.95 (d, *J* = 7.7 Hz, 1H), 8.07 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 48.0, 69.9, 72.6, 76.1, 84.0, 88.5, 101.4, 104.8, 118.0, 119.4, 120.8, 123.5, 131.5, 134.5, 151.6, 152.2, 158.9, 162.2, 175.1; HRMS (ESI−) calcd for $C_{18}H_{18}N_5O_8S$ $[M - H]^-$ 464.0882, found 464.0873 (error 1.9 ppm).

(1*R*)-2-Aza-2-(benzenesulfonyl)bicyclo[2.2.1]hept-5-en-3-one (35).[22] To a solution of (1*R*)-(−)-azabicyclo[2.2.1]hept-5-ene-3-one (**34**) (3.00 g, 27.5 mmol) in THF (250 mL) at 0 °C was added NaH (60% dispersion in mineral oil, 1.32 g, 33.0 mmol, 1.2 equiv) and the slurry stirred for 30 min at room temperature. Next, neat benzenesulfonyl chloride (4.21 mL, 33.0 mmol, 1.2 equiv) was added and the mixture stirred an additional 1.5 h at room temperature. The resulting dark-black reaction was quenched with ice (25 g) and then concentrated. The residue was taken up in CH_2Cl_2 (200 mL) and washed with saturated aqueous $NaHCO_3$ (150 mL), dried (Na_2SO_4), and concentrated. Purification by flash chromatography (CH_2Cl_2) afforded the title compound (4.27 g, 62%) as a white solid: mp 89−91 °C; R_f = 0.35 (1:9 CH_2Cl_2); $[\alpha]_D^{20}$ +10.9 (*c* 0.110, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 2.18 (d, *J* = 8.4 Hz, 1H), 2.42 (d, *J* = 8.4 Hz, 1H), 3.38 (s, 1H), 5.05 (s, 1H), 6.37 (s, 1H), 6.62 (d, *J* = 4.8 Hz, 1H), 7.50 (t, *J* = 7.8 Hz,

2H), 7.61 (t, $J = 7.8$ Hz, 1H), 7.91 (d, $J = 7.8$ Hz, 2H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 54.4, 55.5, 65.4, 127.7, 128.9, 133.8, 136.4, 137.7, 139.3, 174.6; HRMS (ESI−) calcd for $C_{26}H_{28}N_3O_{10}S$ [M − H]$^-$ 574.1501, found 564.1498 (error 0.5 ppm).

(1*R*,4*S*)-1-(Benzenesulfonylamino)-4-(hydroxymethyl)cyclopent-2-ene (36).[22] To a solution of lactam **35** (3.99 g, 16.0 mmol, 1.0 equiv) in MeOH (75 mL) at 0 °C was added $NaBH_4$ (1.81 g, 48.0 mmol, 3.0 equiv) in five portions over 5 min. The mixture was stirred for 1.5 h at 0 °C and then quenched by the dropwise addition of saturated aqueous NH_4Cl (40 mL) and partitioned between EtOAc (300 mL) and H_2O (100 mL). The aqueous layer was separated and back-extracted with EtOAc (2 × 100 mL), and the combined organic extracts were dried (Na_2SO_4) and concentrated. Purification by flash chromatography (Et_2O) afforded the title compound (4.05 g, 100%) as a viscous colorless oil: $R_f = 0.27$ (Et_2O); $[\alpha]_D^{20} - 36.9$ (*c* 1.94, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.18 (t, $J = 7.2$ Hz, 1H), 1.30 (d, $J = 14.4$ Hz, 1H), 2.17 (dt, $J = 14.4, 9.0$ Hz, 1H), 2.66−2.76 (m, 1H), 3.44−3.50 (m, 2H), 3.58 (dd, $J = 10.8, 3.0$ Hz, 1H), 4.32 (d, $J = 8.4$ Hz, 1H), 5.55 (d, $J = 5.4$ Hz, 1H), 5.70 (d, $J = 5.4$ Hz, 1H), 7.49 (t, $J = 7.8$ Hz, 2H), 7.55 (t, $J = 7.8$ Hz, 1H), 7.87 (d, $J = 7.8$ Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 34.3, 46.1, 58.6, 63.5, 126.9, 129.1, 132.4, 132.5, 135.1, 141.2; HRMS (ESI−) calcd for $C_{12}H_{15}NNaO_3S$ [M + Na]$^+$ 276.0665, found 276.0677 (error 4.7 ppm).

(1*R*,4*S*)-1-(*N*-Benzenesulfonyl-*N*-pivaloylamino)-4-[(pivaloyloxy)methyl]cyclopent-2-ene (37).[22] To a slurry of NaH (6.40 g, 160 mmol, 10.0 equiv) in THF (100 mL) at 0 °C was cannulated a solution of alcohol **36** (4.05 g, 16.0 mmol, 1.0 equiv) in THF (100 mL) over 5 min, and the mixture was stirred at room temperature for 30 min. Next, neat pivolyl chloride (19.7 mL, 160 mmol, 10.0 equiv) was added and the mixture was refluxed 4 h. After the mixture was cooled to room temperature, the reaction was quenched by the slow addition of saturated aqueous NH_4Cl (100 mL), extracted with Et_2O (2 × 250 mL), dried (Na_2SO_4), and concentrated to a yellow oil. Purification by flash chromatography (90:10 hexane/EtOAc) afforded the title compound (5.74 g, 88%) as a white crystalline solid: mp 66−67 °C; $R_f = 0.24$ (90:10 hexane/EtOAc); $[\alpha]_D^{20}$ −18.1 (*c* 0.93, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.17 (s, 9H), 1.32 (s, 9H), 1.79 (dt, $J = 13.8, 7.2$ Hz, 1H), 2.20 (dt, $J = 13.8, 8.4$ Hz, 1H), 2.80−2.90 (m, 1H), 3.94−4.04 (m, 2H), 4.82 (t, $J = 7.8$ Hz, 1H), 5.63 (d, $J = 5.4$ Hz), 5.79 (d, $J = 5.4$ Hz, 1H), 7.51 (t, $J = 7.8$ Hz, 2H), 7.60 (t, $J = 7.8$ Hz, 1H), 7.89 (d, $J = 7.8$ Hz, 2H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 27.1, 28.2, 32.6, 38.7, 43.8, 43.9, 65.2, 66.8, 128.5, 128.8, 131.1, 133.2, 134.2, 139.8, 178.3, 185.9.

(1*R*,4*S*)-1-{4-[(Pivaloyloxy)methyl]cyclopent-2-ene-1-yl}-4-nitroindole (38).[22] To a solution of 4-nitroindole (1.31 g, 8.06 mmol, 1.1 equiv) in DMSO (100 mL) was added NaH (60% dispersion in mineral oil, 322 mg, 8.06 mmol, 1.1 equiv). The resulting deep-purple solution was stirred for 10 min, and then $Pd_2(dba)_3 \cdot CHCl_3$ (190 mg, 0.183 mmol, 0.025 equiv) and neat P(O-*i*-Pr)$_3$ (362 μL, 1.47 mmol, 0.20 equiv) were added sequentially. Next, a solution of **37** (3.00 g, 7.33 mmol, 1.0 equiv) in THF (100 mL) was cannulated and the mixture stirred for 3 h at room temperature. The reaction mixture was partitioned between EtOAc (500 mL) and H_2O (100 mL). The organic phase was separated and washed successfully with H_2O (2 × 250 mL) and saturated aqueous NaCl (250 mL), dried (Na_2SO_4), and concentrated. Purification by flash chromatography (6:1 hexane/EtOAc) afforded the title compound (1.25 g, 55%) as a light-orange oil: $R_f = 0.13$ (90:10 hexane/EtOAc); $[\alpha]_D^{20} - 20.7$ (*c* 1.60, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.18 (s, 9H), 1.60 (td, $J = 13.8, 7.2$ Hz, 1H), 2.80 (dt, $J = 13.8, 8.4$ Hz, 1H), 3.12−3.22 (m, 1H), 4.04−4.14 (m, 2H), 5.58−5.68 (m, 1H), 5.99−6.02 (m, 1H), 6.15−6.17 (m, 1H), 7.23 (ovlp d, $J = 3.0$ Hz, 1H), 7.25 (ovlp t, $J = 7.8$ Hz, 1H), 7.39 (d, $J = 3.0$ Hz, 1H), 7.76 (d, $J = 7.8$ Hz, 1H), 8.12 (d, $J = 7.8$, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 27.2, 35.4, 38.8, 44.5, 61.9, 66.4, 102.3, 116.3, 117.5, 120.3, 123.1, 129.4, 131.1, 136.8, 138.9, 140.4, 178.4.

(1*S*,2*R*,3*S*,5*S*)-1-{1,2-Dihydroxy-1,2-*O*-isopropylidene-5-[(pivaloyloxy)methyl]cyclopent-3-yl}-4-nitroindole (40) and (1*R*,2*S*,3*S*,5*S*)-1-{1,2-Dihydroxy-1,2-*O*-isopropylidene-5-[(pivaloyloxy)methyl]cyclopent-3-yl}-4-nitroindole (39). To a solution of **38** (1.24 g, 3.97 mmol, 1.0 equiv) in THF (40 mL) OsO_4 (2.50 mL, 2.5 wt % solution in *t*-BuOH, 0.19 mmol, 0.05 equiv) and NMO (931 mg, 7.94 mmol, 2.0 equiv) were added, and the mixture was stirred 2.5 h at room temperature. The reaction was quenched with 10% aqueous $Na_2S_2O_5$ (20 mL) and then partitioned between EtOAc (250 mL) and H_2O (100 mL). The aqueous phase was back-extracted with EtOAc (2 × 125 mL), and the combined organic extracts were dried (Na_2SO_4) and concentrated to an orange foam. The crude diol was dissolved in 2,2-dimethoxypropane (50 mL). Then *p*-TsOH·H_2O (75 mg, 3.97 mmol, 0.10 mmol) was added and the resulting dark-brown solution was stirred at room temperature for 24 h. The reaction was quenched with solid $NaHCO_3$ (1.0 g). The mixture was filtered through Celite washing with acetone (25 mL), and the filtrate was concentrated. Purification by flash chromatography (10−20% EtOAc/hexane) afforded two diastereomers **39** (400 mg, 26%) and **40** (371 mg, 24%). Assignment was based on ^{1}H−^{1}H COSY and NOESY spectra.

Data for **39**: $R_f = 0.50$ (70:30 hexane/EtOAc); $[\alpha]_D^{20} + 49.4$ (*c* 0.77, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.25 (s, 9H, *t*-Bu), 1.30 (s, 3H, acetonide), 1.63 (s, 3H, acetonide), 2.21 (q, $J = 12.6$ Hz, 1H, H-4′β), 2.53 (dt, $J = 12.6, 6.0$ Hz, 1H, H-4′α), 2.62 (dp, $J = 12.6, 6.0$ Hz, 1H, H-5′), 4.24 (dd, $J = 10.8, 6.0$ Hz, 1H, PivO-C$\underline{H_2}$-), 4.30 (dd, $J = 10.8, 6.0$ Hz, 1H, PivO-C$\underline{H_2}$-), 4.55 (t, $J = 6.6$ Hz, 1H, H-1′), 4.61 (dd, $J = 7.2, 5.4$ Hz, 1H, H-2′), 4.84 (dt, $J = 12.0, 6.0$ Hz, 1H, H-3′), 7.28 (ovlp d, $J = 3.0$ Hz, 1H, H-3), 7.30 (ovlp t, $J = 7.8$ Hz, 1H, H-6), 7.40 (d, $J = 3.0$ Hz, 1H, H-2), 7.92 (d, $J = 7.8$ Hz, 1H, H-7), 8.14 (d, $J = 7.8$ Hz, 1H, H-5); NOE ^{1}H NMR (600 MHz, $CDCl_3$) (Irr, H-3′), enhancements at 2.53 (H-4′α), 2.62 (H-5′), 7.92 (H-7); ^{13}C NMR (150 MHz, $CDCl_3$) δ 24.9, 27.2, 27.5, 33.5, 38.9, 43.2, 61.9, 64.4, 81.1, 85.3, 102.7, 114.1, 117.2, 117.8, 120.7, 122.9, 128.2, 138.7, 140.4, 178.3; HRMS (ESI−) calcd for $C_{22}H_{28}N_2NaO_6$ [M + Na]$^+$ 439.1840, found 439.1841 (error 0.2 ppm).

Data for **40**: $R_f = 0.38$ (70:30 hexane/EtOAc); $[\alpha]_D^{20} + 60.5$ (*c* 0.11, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.22 (s, 9H, *t*-Bu), 1.25 (s, 3H, acetonide), 1.50 (s, 3H, acetonide), 2.12−2.15 (m, 1H, H-4′β), 2.24−2.35 (m, 2H, H-5′, H-4′α), 4.28 (dd, $J = 10.8, 7.2$ Hz, 1H, PivO-C$\underline{H_2}$-), 4.38 (dd, $J = 13.8, 6.6$ Hz, 1H, PivO-C$\underline{H_2}$-), 4.62 (dt, $J = 11.4, 5.4$ Hz, 1H, H-3′), 4.73−4.76 (m, 2H, H-1′, H-2′), 7.25−7.28 (m, 2H, H-3, H-6), 7.59 (d, $J = 3.0$ Hz, 1H, H-2), 7.68 (d, $J = 7.8$ Hz, 1H, H-7), 8.13 (d, $J = 7.8$ Hz, 1H, H-5); NOE ^{1}H NMR (600 MHz, $CDCl_3$) 4.62 (Irr, H-3′), enhancement at 2.12−2.15 (H-4′β), 2.24−2.35 (H-5′, H-4′α), 4.73−4.76 (H-1′, H-2′), 7.68 (H-7); ^{13}C NMR (150 MHz, $CDCl_3$) δ 23.7, 25.6, 27.2, 29.9, 38.8, 39.8, 57.6, 63.1, 78.3, 79.1, 101.9, 111.0, 115.5, 117.7, 120.3, 122.8, 131.5, 138.5, 140.5, 178.4; HRMS (ESI−) calcd for $C_{22}H_{29}N_2O_6$ [M + H]$^+$ 417.2020, found 417.2032 (error 2.9 ppm).

(1*R*,2*S*,3*S*,5*S*)-1-{1,2-Dihydroxy-5-[(hydroxy)methyl]-1,2-*O*-isopropylidenecyclopent-3-yl}-4-nitroindole (41). A suspension of **39** (264 mg, 0.68 mmol, 1.0 equiv) in MeOH/0.5 M aqueous NaOH (3:2, 100 mL) was heated at 100 °C for 2 h. The resulting homogeneous solution was cooled to room temperature and concentrated to remove MeOH, and the remaining aqueous layer was partitioned between EtOAc (100 mL) and aqueous 0.5 M NaH_2PO_4 (50 mL). The organic layer was separated, dried (Na_2SO_4), and concentrated. Purification by flash chromatography (70:30 EtOAc/hexane) afforded the title compound (185 mg, 89%) as a yellow foam: $R_f = 0.24$ (70:30 EtOAc/hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.31 (s, 3H), 1.63 (s, 3H), 1.71 (br s, 1H, OH), 2.26−2.36 (m, 1H), 2.46−2.56 (m, 2H), 3.78−3.92 (m, 2H), 4.56−4.68 (m, 2H), 4.80−4.88 (m, 1H), 7.24−7.34 (m, 2H), 7.46 (d, $J = 3.0$ Hz, 1H), 7.94 (d, $J = 7.8$ Hz, 1H), 8.15 (d, $J = 7.8$ Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 24.9, 27.5, 32.8, 45.8, 62.4, 63.3, 81.3, 85.6, 102.6, 113.9, 117.4, 117.8, 120.6, 123.0, 128.5,

138.8, 140.3; HRMS (ESI−) calcd for $C_{17}H_{20}N_2NaO_5$ [M + Na]$^+$ 355.1270, found 355.1278 (error 2.3 ppm).

(1*R*,2*S*,3*S*,5*S*)-1-(1,2-Dihydroxy-1,2-*O*-isopropylidene-5-{[(sulfamoyl)oxy]methyl}cyclopent-3-yl)-4-nitroindole (42). To a stirred solution of alcohol **41** (183 mg, 0.61 mmol, 1.0 equiv) in dimethylacetamide (4.0 mL) at 0 °C was added solid sulfamoyl chloride (280 mg, 2.42 mmol, 4.0 equiv). The mixture was stirred for 2.5 h at 0 °C and then partitioned between H_2O (150 mL) and EtOAc (150 mL). The organic layer was separated and washed with H_2O (2 × 150 mL), saturated aqueous NaCl (100 mL), dried (Na_2SO_4), and concentrated. Purification by flash chromatography (40−70% EtOAc/hexane) afforded the title compound (200 mg, 86%) as a yellow oil: ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.29 (s, 3H), 1.62 (s, 3H), 2.33 (q, *J* = 12.6, 1H), 2.56 (dt, *J* = 12.6, 6.6 Hz, 1H), 2.64 (dp, *J* = 12.6, 6.0 Hz, 1H), 4.36−4.41 (m, 2H), 4.61 (t, *J* = 6.6 Hz, 1H), 4.65 (t, *J* = 6.6 Hz, 1H), 4.82 (dt, *J* = 12.6, 6.6 Hz, 1H), 7.24 (d, *J* = 3.6 Hz, 1H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.45 (d, *J* = 3.6 Hz, 1H), 7.92 (d, *J* = 7.8 Hz, 1H), 8.12 (d, *J* = 7.8 Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 24.9, 27.3, 32.9, 43.1, 61.8, 69.9, 80.2, 85.1, 102.5, 114.4, 117.3, 117.8, 120.6, 122.8, 128.5, 138.6, 140.2; HRMS (ESI+) calcd for $C_{17}H_{21}N_3NaO_7S$ [M + Na]$^+$ 434.0992, found 434.1014 (error 5.1 ppm).

(1*R*,2*S*,3*S*,5*S*)-1-{1,2-Dihydroxy-5-[({*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}oxy)methyl]-1,2-*O*-isopropylidenecyclopent-3-yl}-4-nitroindole Triethylammonium Salt (43). Sulfamate **42** (154 mg, 0.403 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation. Purification by flash chromatography (9:1 EtOAc/hexanes) provided the product (180 mg, 77%) as a viscous oil: R_f = 0.40 (7:3 EtOAc/hexanes); $[\alpha]_D^{20}$ +44.6 (*c* 0.473, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.28 (ovlp t, *J* = 7.2 Hz, 9H), 1.29 (ovlp s, 3H), 1.60 (s, 3H), 2.46 (q, *J* = 12.0 Hz, 1H), 2.54 (dt, *J* = 12.0, 6.0 Hz, 1H), 2.65 (dp, *J* = 12.0, 5.4 Hz, 1H), 3.17 (q, *J* = 7.2 Hz, 6H), 3.45 (s, 3H), 4.34−4.40 (m, 2H), 4.69 (t, *J* = 6.0 Hz, 1H), 4.75 (t, *J* = 6.0 Hz, 1H), 4.95 (dt, *J* = 12.6, 6.6 Hz, 1H), 5.16−5.19 (m, 2H), 6.98 (t, *J* = 7.8 Hz, 1H), 7.12 (d, *J* = 8.4 Hz, 1H), 7.16 (d, *J* = 3.0 Hz, 1H), 7.26−7.34 (m, 2H), 7.46 (d, *J* = 7.2 Hz, 1H), 7.77 (d, *J* = 3.0 Hz, 1H), 8.03 (d, *J* = 8.4 Hz, 1H), 8.08 (d, *J* = 7.8 Hz, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 25.5, 27.9, 34.5, 44.5, 47.9, 56.7, 63.6, 70.7, 82.2, 86.8, 96.6, 103.2, 115.1, 117.2, 118.5, 118.9, 121.5, 122.6, 124.1, 130.0, 130.9, 131.3, 132.3, 140.4, 141.4, 155.8, 176.3; HRMS (ESI−) calcd for $C_{26}H_{28}N_3O_{10}S$ [M − H]$^-$ 574.1501, found 574.1498 (error 0.5 ppm).

(1*R*,2*S*,3*S*,5*S*)-1-[1,2-Dihydroxy-5-({[*N*-(2-hydroxybenzoyl)sulfamoyl]oxy}methyl)cyclopent-3-yl]-4-nitroindole Triethylammonium Salt (44). To a solution of **43** (33 mg, 0.057 mmol, 1.0 equiv) in MeOH (5 mL) was added *p*-TSA·H_2O (12 mg, 0.070 mmol, 1.2 equiv), and the mixture was stirred at 70 °C for 3 h. The mixture was cooled to room temperature, neutralized with triethylamine (100 μL, 0.71 mmol, 10 equiv), and concentrated. Purification by flash chromatography (1:9 MeOH/EtOAc) afforded the title compound (13 mg, 46%) as a viscous oil: R_f = 0.29 (EtOAc); $[\alpha]_D^{20}$ +57.1 (*c* 0.210, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.29 (t, *J* = 7.2 Hz, 9H), 1.96−2.02 (m, 1H), 2.42−2.50 (m, 2H), 3.29 (q, *J* = 7.2 Hz, 6H), 4.08 (dd, *J* = 5.4, 2.4 Hz, 1H), 4.25−4.35 (m, 3H), 4.91−4.96 (m, 1H), 6.77−6.82 (m, 2H), 7.16 (d, *J* = 3.0 Hz, 1H), 7.27−7.31 (m, 2H), 7.83 (d, *J* = 3.0 Hz, 1H), 7.94 (d, *J* = 7.8 Hz, 1H), 7.99 (d, *J* = 8.4 Hz, 1H), 8.07 (d, *J* = 8.4 Hz, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 30.2, 44.2, 48.0, 71.4, 73.9, 77.9, 103.1, 117.9, 118.2, 118.5, 119.4, 120.9, 121.1, 124.0, 131.1, 131.4, 134.4, 140.8, 141.4, 162.2, 174.9 (missing 1 Ar C); HRMS (ESI−) calcd for $C_{21}H_{20}N_3O_9S$ [M − H] 490.0926, found 490.0922 (error 0.8 ppm).

(1*R*,2*S*,3*S*,5*S*)-4-Amino-1-{1,2-dihydroxy-5-({[*N*-(2-hydroxybenzoyl)sulfamoyl]oxy}methyl)cyclopent-3-yl]indole Triethylammonium Salt (8). To a solution of compound **44** (5.0 mg, 8.4 μmol, 1.0 equiv) in DMF (2.0 mL) was added 10% w/v Pd/C (3.0 mg), and the mixture was stirred under H_2 (1 atm) at room temperature for 8 h. The mixture was filtered through Celite and the filtrate concentrated. Purification by flash chromatography (80:20:1 EtOAc/MeOH/Et_3N) afforded the title compound (2.1 mg, 44%) as a light-brown solid: ^{1}H NMR (600 MHz, CD_3OD) δ 1.29 (t, *J* = 7.2 Hz, 9H), 1.84−1.94 (m, 1H), 2.36−2.46 (m, 2H), 3.17 (q, *J* = 7.2 Hz, 6H), 4.08 (t, *J* = 4.8 Hz, 1H), 4.23−4.31 (m, 3H), 4.75 (q, *J* = 9.6 Hz, 1H), 6.37 (dd, *J* = 6.0, 1.2 Hz, 1H), 6.52 (d, *J* = 3.6 Hz, 1H), 6.71−6.81 (m, 2H), 6.90−6.94 (m, 2H), 7.29 (td, *J* = 7.8, 1.8 Hz, 1H), 7.32 (d, *J* = 3.6 Hz, 1H), 7.94 (dd, *J* = 7.8, 1.8 Hz, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.4, 30.4, 44.0, 48.0, 61.5, 71.5, 73.9, 77.6, 99.6, 102.5, 105.6, 117.9, 119.3, 120.1, 120.9, 123.4, 123.8, 131.5, 134.3, 139.5, 140.8, 162.1, 174.9; HRMS (ESI−) calcd for $C_{21}H_{22}N_3O_7S$ [M − H]$^-$ 460.1184, found 460.1168 (error 3.5 ppm).

2′,3′-*O*-Isopropylideneinosine (46). Inosine **45** (1.00 g, 3.73 mmol, 1.00 equiv) was converted to the title compound using the general procedure for acetonide protection. Purification by flash chromatography (95:5 EtOAc/MeOH) provided the product (0.87 g, 76%) as a white solid: R_f = 0.5 (1:9 MeOH/EtOAc); $[\alpha]_D^{20}$ −35 (*c* 0.50, DMSO); ^{1}H NMR (600 MHz, CD_3OD) δ 1.41 (s, 3H), 1.30 (s, 3H), 3.74 (dd, *J* = 12.0, 4.8 Hz, 1H), 3.80 (dd, *J* = 12.0, 3.6 Hz, 1H), 4.37−4.40 (m, 1H), 5.04 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.29 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.22 (d, *J* = 2.4 Hz, 1H), 8.10 (s, 1H), 8.35 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.6, 63.4, 83.0, 86.0, 88.6, 92.5, 115.4, 126.0, 141.1, 146.9, 149.6, 158.9; HRMS (APCI−) calcd for $C_{13}H_{15}N_4O_5$ [M − H]$^-$ 307.1048, found 307.1072 (error 7.8 ppm).

2′,3′-*O*-Isopropylidene-5′-*O*-(sulfamoyl)inosine (47). Compound **46** (250 mg, 0.81 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (95:5 EtOAc/MeOH) provided the product (280 mg, 92%) as a white solid: R_f = 0.4 (80:20 EtOAc/MeOH); $[\alpha]_D^{20}$ −14 (*c* 0.75, DMSO); ^{1}H NMR (600 MHz, CD_3OD) δ 1.35 (s, 3H), 1.56 (s, 3H), 4.23−4.31 (m, 2H), 4.49−4.51 (m, 1H), 5.07−5.09 (m, 1H), 5.36 (d, *J* = 3.6 Hz, 1H), 6.21 (s, 1H), 8.07 (s, 1H), 8.20 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.3, 26.2, 68.6, 81.6, 84.5, 84.7, 90.9, 114.4, 124.6, 139.7, 145.9, 148.3, 157.7; HRMS (APCI−) calcd for $C_{13}H_{16}N_5O_7S$ [M − H]$^-$ 386.0776, found 386.0798 (error 5.7 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]inosine Triethylammonium Salt (9). Compound **47** (40 mg, 0.10 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (17:3 EtOAc/MeOH) afforded the title compound (48 mg, 50%) as a white solid: R_f = 0.2 (3:7 MeOH/EtOAc); $[\alpha]_D^{20}$ −11.7 (*c* 1.40, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.27 (t, *J* = 7.2 Hz, 9H), 3.17 (q, *J* = 7.2 Hz, 6H), 4.31−4.32 (m, 1H), 4.35−4.38 (m, 1H), 4.41−4.42 (m, 2H), 4.76 (t, *J* = 6.0 Hz, 1H), 6.07 (d, *J* = 6.0 Hz, 1H), 6.76−6.79 (m, 2H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.92 (d, *J* = 8.4 Hz, 1H), 8.00 (s, 1H), 8.44 (s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 9.4, 47.9, 69.7, 72.6, 76.1, 84.8, 89.9, 118.0, 119.4, 120.8, 125.7, 131.4, 134.5, 140.9, 146.9, 150.3, 159.1, 162.1, 174.9; MS (APCI−) calcd for $C_{17}H_{16}N_5O_9S$ [M − H]$^-$ 466.1 found 466.1.

2′,3′-*O*-Isopropylidene-*N*^{6},*N*$^{6\prime}$-dimethyladenosine (49). To a stirred solution of **48** (200 mg, 0.61 mmol, 1.00 equiv) in *tert*-BuOH (20 mL) in a sealed tube was added *N*,*N*-dimethylamine (2.0 N in MeOH, 1.0 mL, 2.0 mmol, 3.3 equiv). The mixture was heated at 85 °C for 3 h, then cooled to room temperature and concentrated. Purification by flash chromatography (1:1 EtOAc/hexane) afforded the title compound (198 mg, 96%) as white solid: R_f = 0.4 (EtOAc); $[\alpha]_D^{20}$ −62 (*c* 0.50, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.51 (s, 3H), 1.77 (s, 3H), 3.65 (br s, 6H), 3.89 (dd, *J* = 12.6, 3.6 Hz, 1H), 4.00 (dd, *J* = 12.6, 3.0 Hz, 1H), 4.55−4.57 (m, 1H), 5.18 (dd, *J* = 6.6, 1.8 Hz, 1H), 5.34 (dd, *J* = 6.6, 4.2 Hz, 1H), 6.21 (d, *J* = 4.2 Hz, 1H), 8.27 (s, 1H), 8.34 (s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 25.4, 27.9, 39.1 (br), 63.7, 81.9, 82.8, 86.2, 94.6, 114.0, 121.8, 138.2, 148.9, 151.9, 155.3; MS (APCI+) calcd for $C_{15}H_{22}N_5O_4$ [M + H]$^+$ 336.2, found 336.1.

2′,3′-*O*-Isopropylidene-5′-*O*-sulfamoyl-*N*^{6},*N*$^{6\prime}$-dimethyladenosine (59). Compound **49** (175 mg, 0.52 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (3:2 EtOAc/hexane) provided the product (190 mg, 88%) as a white solid: R_f = 0.5 (EtOAc); $[\alpha]_D^{20}$ −27.6 (*c* 1.30, MeOH); ^{1}H NMR (600 MHz,

CD_3OD) δ 1.42 (s, 3H), 1.65 (s, 3H), 3.51 (br s, 6H), 4.31 (dd, *J* = 10.8. 4.8 Hz, 1H), 4.37 (dd, *J* = 10.8, 4.2 Hz, 1H), 4.51−4.58 (m, 1H), 5.16 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.42 (dd, *J* = 6.0. 2.4 1H), 6.26 (d, *J* = 2.4 Hz, 1H), 8.19 (s, 1H), 8.24 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.5, 39.2, 70.1, 83.0, 85.6, 85.7, 91.7, 115.7, 121.3, 139.5, 151.1, 153.5, 156.2; MS (APCI+) calcd for $C_{15}H_{23}N_6O_6S$ $[M + H]^+$ 415.1, found 415.1.

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-N^6,$N^{6\prime}$-dimethyladenosine Triethylammonium Salt (10). Compound **59** (60 mg, 0.143 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH//Et_3N) provided the product (69 mg, 81%) as a white solid: R_f = 0.4 (3:17 MeOH/EtOAc); $[\alpha]_D^{20}$ −32 (*c* 0.50, MeOH); ^1H NMR (600 MHz, CD_3OD) δ 1.24 (t, *J* = 7.2 Hz, 9H), 3.05 (q, *J* = 7.2 Hz, 6H), 3.47 (br s, 6H), 4.34 (q, *J* = 3.6 Hz, 1H), 4.39−4.48 (m, 3H), 4.68 (t, *J* = 5.4 Hz, 1H), 6.08 (d, *J* = 5.4 Hz, 1H), 6.76 (ovlp t, *J* = 8.4 Hz, H), 6.78 (ovlp d, *J* = 8.4 Hz, 1H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.95 (d, *J* = 8.4, 1H), 8.17 (s, 1H), 8.30 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.9, 39.3, 47.8, 70.0, 72.3, 76.0, 84.2, 89.6, 118.1, 119.4, 120.3, 121.3, 131.7, 134.8, 139.1, 151.5, 153.2, 156.2, 162.2, 175.5; HRMS (ESI−) calcd for $C_{19}H_{21}N_6O_8S$ $[M - H]^-$ 493.1147, found 493.1146 (error 0.2 ppm).

2′,3′-*O*-Isopropylidene-N^6-methyladenosine (50). The title compound was synthesized from **48** (500 mg, 1.74 mmol, 1.0 equiv) and methylamine (2 N in THF, 1.74 mL, 3.48 mmol, 2.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (EtOAc) provided the product (430 mg, 77%) as a viscous oil: R_f = 0.20 (EtOAc); ^1H NMR (600 MHz, $CDCl_3$) δ 1.32 (s, 3H), 1.59 (s, 3H), 3.12 (br s, 3H), 3.74 (d, *J* = 12.6 Hz, 1H), 3.93 (d, *J* = 12.6 Hz, 1H), 4.49 (s, 1H), 5.07 (d, *J* = 5.4 Hz, 1H), 5.17 (t, *J* = 5.4 Hz, 1H), 5.80 (d, *J* = 5.4 Hz, 1H), 7.73 (s, 1H), 8.30 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 25.5, 27.6, 27.9, 63.6, 82.0, 83.2, 86.3, 94.5, 114.2, 121.7, 139.6, 147.3, 153.0, 156.1; HRMS (ESI+) calcd for $C_{14}H_{20}N_5O_4$ $[M + H]^+$ 322.1510, found 322.1520 (error 3.1 ppm).

2′,3′-*O*-Isopropylidene-5′-*O*-sulfamoyl-N^6-methyladenosine (60). Compound **50** (420 mg, 1.3 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (4:1 EtOAc/hexane) afforded the title compound (0.45 mg, 86%) as a viscous oil: R_f = 0.59 (9:1 EtOAc/MeOH); ^1H NMR (600 MHz, CD_3OD) δ 1.37 (s, 3H), 1.60 (s, 3H), 3.08 (br s, 3H), 4.27 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.2 Hz, 1H), 4.51 (app dd, *J* = 7.8, 4.8 Hz, 1H), 5.12 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.40 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.22 (d, *J* = 2.4 Hz, 1H), 8.18 (s, 1H), 8.25 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.5, 27.4, 27.7, 69.9, 82.9, 85.5, 85.6, 91.7, 115.6, 120.9, 140.8, 149.1, 154.1, 156.6; HRMS (ESI−) calcd for $C_{14}H_{19}N_6O_6S$ $[M - H]^-$ 399.1092, found 399.1092 (error 0 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-N^6-methyladenosine Sodium Salt (11). Compound **60** (230 mg, 0.57 mmol, 1.0 equiv) was coupled to compound **33** using the general procedure for salicylation. Purification by flash chromatography (10:90:1 MeOH/EtOAc/Et_3N) afforded 2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}-N^6-methyladenosine triethylammonium salt (320 mg, 84%) as a viscous oil: R_f = 0.50 (4:1 EtOAc/MeOH); ^1H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 1.37 (s, 3H), 1.61 (s, 3H), 3.09, (br s, 3H), 3.12 (q, *J* = 7.2 Hz, 6H), 3.42 (s, 3H), 4.34 (dd, *J* = 10.8, 3.9 Hz, 1H), 4.37 (dd, *J* = 10.8, 3.9 Hz, 1H), 4.59−4.60 (m, 1H), 5.16 (s, 2H), 5.22 (dd, *J* = 6.0, 2.1 Hz, 1H), 5.41 (dd, *J* = 6.0, 3.6 Hz, 1H), 6.25 (d, *J* = 3.6 Hz, 1H), 6.97 (t, *J* = 7.8 Hz, 1H), 7.12 (d, *J* = 7.8 Hz, 1H), 7.27 (t, *J* = 7.8 Hz, 1H), 7.42 (dd, *J* = 7.8, 1H), 8.25 (br s, 1H), 8.44 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 25.6, 27.5, 27.6, 47.8, 56.6, 69.9, 83.4, 85.66, 85.75, 91.8, 96.4, 115.2, 117.0, 120.7, 122.5, 129.7, 131.0, 132.4, 140.8, 149.7, 154.0, 155.6, 156.7, 176.8; HRMS (ESI−) calcd for $C_{23}H_{27}N_6O_9S$ $[M - H]^-$ 563.1566, found 563.1572 (error 1.1 ppm).

2′,3′-*O*-Isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl-]sulfamoyl}-N^6-methyladenosine triethylammonium salt (140 mg, 0.21 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (10:90:1 MeOH/EtOAc/Et_3N) followed by preparative HPLC using method 2 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (33 mg, 31%) as a white solid (reported NMR data are for the TEA salt of the title compound): mp >200 °C (dec); R_f = 0.35 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −22.7 (*c* 0.233, MeOH); ^1H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 3.09 (br s, 3H), 3.15 (q, *J* = 7.2 Hz, 6H), 4.32 (dd, *J* = 6.0, 3.0 Hz, 1H), 4.37 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.41−4.44 (m, 2H), 4.72 (t, *J* = 6.0 Hz, 1H), 6.08 (d, *J* = 6.0 Hz, 1H), 6.75−6.79 (m, 2H), 7.28 (td, *J* = 7.8, 1.8 Hz, 1H), 7.94 (dd, *J* = 7.8, 1.8 Hz, 1H), 8.22 (br s, 1H), 8.44 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 27.7, 47.9, 69.6, 72.4, 76.1, 84.5, 89.2, 117.9, 119.3, 120.6, 120.7, 131.3, 134.4, 140.5, 149.7, 153.9, 156.6, 162.0, 175.0; HRMS (ESI−) calcd for $C_{18}H_{19}N_6O_8S$ $[M - H]^-$ 479.0991, found 479.0996 (1.0 ppm).

N^6-Ethyl-2′,3′-*O*-isopropylideneadenosine (51). The title compound was synthesized from **48** (400 mg, 1.4 mmol, 1.0 equiv) and ethylamine (2 N in THF, 1.1 mL, 2.1 mmol, 1.5 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (4:1 EtOAc/hexanes) provided the product (450 mg, 95%) as a white solid: mp 109−111 °C; R_f = 0.64 (9:1 EtOAc/MeOH); ^1H NMR (600 MHz, $CDCl_3$) δ 1.19 (t, *J* = 7.2 Hz, 3H), 1.27 (s, 3H), 1.53 (s, 3H), 3.54 (br s, 2H), 3.70 (d, *J* = 10.8 Hz, 1H), 3.86 (d, *J* = 10.8 Hz, 1H), 4.43 (s, 1H), 5.01 (d, *J* = 6.0 Hz, 1H), 5.14 (t, *J* = 4.8 Hz, 1H), 5.79 (d, *J* = 4.8 Hz, 1H), 7.70 (s, 1H), 8.22 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 14.8, 25.2, 27.6, 35.4, 63.2, 81.7, 83.0, 86.1, 93.9, 113.8, 121.0, 139.3, 147.2, 152.7, 155.1; HRMS (ESI+) calcd for $C_{15}H_{22}N_5O_4$ $[M + H]^+$ 336.1666, found 336.1653 (error 3.9 ppm).

N^6-Ethyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (61). Compound **51** (260 mg, 0.78 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (9:1 EtOAc/hexanes) provided the product (245 mg, 76%) as a viscous oil: R_f = 0.58 (EtOAc); ^1H NMR (600 MHz, CD_3OD) δ 1.28 (t, *J* = 7.2 Hz, 3H), 1.38 (s, 3H), 1.60 (s, 3H), 3.59 (br s, 2H), 4.27 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.51 (app dd, *J* = 7.8, 4.8 Hz, 1H), 5.12 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.40 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.22 (d, *J* = 2.4 Hz, 1H), 8.20 (s, 1H), 8.24 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 15.0, 25.5, 27.4, 36.5, 69.9, 82.9, 85.5, 85.6, 91.7, 115.6, 120.7, 140.8, 149.3, 154.1, 156.0; HRMS (ESI+) calcd for $C_{15}H_{23}N_6O_6S$ $[M + H]^+$ 415.1394, found 415.1404 (error 2.4 ppm).

N^6-Ethyl-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (12). Compound **61** (112 mg, 0.27 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (10:90:1 MeOH/EtOAc/Et_3N) afforded N^6-ethyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt as a viscous oil (118 mg, 64%): R_f = 0.53 (9:1 EtOAc/MeOH); ^1H NMR (600 MHz, CD_3OD) δ 1.26−1.30 (m, 12H), 1.37 (s, 3H), 1.61 (s, 3H), 3.14 (q, *J* = 7.2 Hz, 6H), 3.42 (s, 3H), 3.60 (br s, 2H), 4.33 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.37 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.59−4.61 (m, 1H), 5.16 (s, 2H), 5.22 (d, *J* = 6.0 Hz, 1H), 5.39−5.41 (m, 1H), 6.25 (d, *J* = 3.0 Hz, 1H), 6.97 (t, *J* = 7.8 Hz, 1H), 7.12 (d, *J* = 8.4 Hz, 1H), 7.27 (t, *J* = 8.4 Hz, 1H), 7.42 (d, *J* = 7.8 Hz, 1H), 8.24 (br s, 1H), 8.45 (s, 1H); HRMS (ESI−) calcd for $C_{24}H_{29}N_6O_9S$ $[M - H]^-$ 577.1722, found 577.1729 (error 1.2 ppm).

N^6-Ethyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)-benzoyl]sulfamoyl}adenosine triethylammonium salt (70 mg, 0.10 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (10:90:1 MeOH/EtOAc/Et_3N) followed by preparative HPLC using method 2 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (25 mg, 47%) as a white solid (reported

NMR data are for the TEA salt of the title compound): mp >200 °C (dec); R_f = 0.44 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −30.4 (*c* 0.184, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.26−1.31 (m, 12H), 3.18 (q, *J* = 7.2 Hz, 6H), 3.60 (br s, 2H), 4.30−4.36 (m, 1H), 4.37 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.40−4.43 (m, 2H), 4.71 (t, *J* = 5.4 Hz, 1H), 6.08 (d, *J* = 6.0 Hz, 1H), 6.76−6.80 (m, 2H), 7.29 (td, *J* = 7.8, 1.2 Hz, 1H), 7.93 (dd, *J* = 7.8, 1.2 Hz, 1H), 8.21 (br s, 1H), 8.46 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 15.0, 36.5, 47.9, 69.6, 72.4, 76.1, 84.6, 89.2, 117.9, 119.3, 120.5, 120.6, 131.4, 134.4, 140.5, 150.1, 153.9, 156.0, 162.1, 175.0; HRMS (ESI−) calcd for $C_{19}H_{21}N_6O_8S$ [M − H]$^-$ 493.1147, found 493.1153 (1.2 ppm).

2′,3′-*O*-Isopropylidene-*N*^{6}-propyladenosine (52). The title compound was synthesized from **48** (420 mg, 1.46 mmol, 1.0 equiv) and *n*-propylamine (179 μL, 2.19 mmol, 1.5 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (3:1 EtOAc/hexanes) provided the product (443 mg, 87%): R_f = 0.65 (EtOAc); ^{1}H NMR (600 MHz, $CDCl_3$) δ 0.93 (t, *J* = 7.2 Hz, 3H), 1.30 (s, 3H), 1.57 (s, 3H), 1.59−1.65 (m, 2H), 3.49 (br s, 2H), 3.71 (d, *J* = 11.4, 1H), 3.90 (d, *J* = 11.4 Hz, 1H), 4.46 (s, 1H), 5.04 (d, *J* = 5.4, 1H), 5.14 (t, *J* = 5.4 Hz, 1H), 5.78 (d, *J* = 5.4 Hz, 1H), 7.71 (s, 1H), 8.25 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 11.5, 22.9, 25.4, 27.8, 42.5, 63.5, 81.9, 83.2, 86.2, 94.4, 114.1. 121.4, 139.5, 147.3, 152.9, 155.5; MS (APCI−) calcd for $C_{16}H_{22}N_5O_4$ [M − H]$^-$ 348.2, found 348.1.

2′,3′-*O*-Isopropylidene-5′-*O*-sulfamoyl-*N*^{6}-propyladenosine (62). Compound **52** (350 mg, 1.0 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (99:1 EtOAc/hexanes) afforded the title compound (321 mg, 75%) as a viscous oil: R_f = 0.7 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.01 (t, *J* = 7.2 Hz, 3H), 1.38 (s, 3H), 1.60 (s, 3H), 1.62−1.70 (m, 2H), 3.52 (br s, 2H), 4.25 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.51 (app dd, *J* = 7.8, 4.8 Hz, 1H), 5.13 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.23 (d, *J* = 2.4 Hz, 1H), 8.21 (s, 1H), 8.24 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 11.8, 23.8, 25.6, 27.5, 43.5, 70.0, 83.0, 85.6, 85.8, 91.8, 115.6, 120.8, 140.8, 149.5, 154.2, 156.3; MS (ESI+) calcd for $C_{16}H_{22}N_5O_3$ [M − SO_2NH_2]$^+$ 332.2, found 332.2.

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-*N*^{6}-propyladenosine Triethylammonium Salt (13). Compound **62** (200 mg, 0.47 mmol, 1.00 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (95:5 EtOAc/MeOH) afforded the title compound (82 mg, 85%) as a viscous oil: R_f = 0.3 (95:5 EtOAc/MeOH); $[\alpha]_D^{20}$ −31.0 (*c* 0.101, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.01 (t, *J* = 7.2 Hz, 3H), 1.25 (t, *J* = 7.2 Hz, 9H), 1.64−1.76 (m, 2H), 3.16 (q, *J* = 7.2 Hz, 6H), 3.52 (br s, 2H), 4.30−4.34 (m, 1H), 4.35−4.44 (m, 3H), 4.70 (t, *J* = 6.0 Hz, 1H), 6.08 (d, *J* = 6.0 Hz, 1H), 6.74−6.82 (m, 2H), 7.28 (t, *J* = 7.2 Hz, 1H), 7.93 (d, *J* = 7.8, 1H), 8.20 (br s, 1H), 8.45 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 11.8, 23.8, 43.6 (br), 48.0, 69.7, 72.5, 76.2, 84.7, 89.3, 118.0, 119.4, 120.6, 120.7, 131.5, 134.5, 140.5, 154.0, 156.3 (br), 162.1, 175.1 (missing 1 Ar C); HRMS (ESI−) calcd for $C_{20}H_{23}N_6O_8S$ [M − H]$^-$ 507.1304, found 507.1315 (error 2.1 ppm).

***N*^{6}-Isopropyl-2′,3′-*O*-isopropylideneadenosine (53).** The title compound was synthesized from **48** (400 mg, 1.4 mmol, 1.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (9:1 EtOAc/hexanes) afforded the title compound (420 mg, 86%) as a white solid: mp 133−135 °C; R_f = 0.44 (EtOAc); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.19 (d, *J* = 6.6 Hz, 6H), 1.28 (s, 3H), 1.54 (s, 3H), 3.70 (d, *J* = 11.4 Hz, 1H), 3.87 (d, *J* = 11.4 Hz, 1H), 4.40 (br s, 1H), 4.43 (s, 1H), 5.02 (d, *J* = 6.0 Hz, 1H), 5.14 (t, *J* = 5.4 Hz, 1H), 5.78 (d, *J* = 5.4 Hz, 1H), 7.71 (s, 1H), 8.22 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 22.8, 25.2, 27.6, 42.4, 63.3, 81.7, 83.0, 86.1, 94.1, 113.8, 121.1, 139.3, 147.3, 152.8, 154.5; HRMS (ESI+) calcd for $C_{16}H_{24}N_5O_4$ [M + H]$^+$ 350.1823, found 350.1820 (error 0.9 ppm).

***N*^{6}-Isopropyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (63).** Compound **53**(250 mg, 0.72 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (9:1 EtOAc/hexanes) afforded the title compound (205 mg, 67%) as a viscous oil: R_f = 0.57 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.30 (d, *J* = 6.6 Hz, 6H), 1.37 (s, 3H), 1.59 (s, 3H), 4.26 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.42 (br s, 1H), 4.49−4.52 (m, 1H), 5.12 (dd, *J* = 6.6, 3.0 Hz, 1H), 5.40 (dd, *J* = 6.6, 2.4 Hz, 1H), 6.22 (d, *J* = 2.4 Hz, 1H), 8.20 (s, 1H), 8.25 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 22.9, 25.5, 27.4, 43.3, 69.9, 82.9, 85.5, 85.7, 91.7, 115.6, 120.5, 140.7, 149.3, 154.2, 155.4; HRMS (ESI−) calcd for $C_{16}H_{23}N_6O_6S$ [M − H]$^-$ 427.1405, found 427.1416 (error 2.6 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-*N*^{6}-isopropyladenosine Sodium Salt (14). Compound **63** (160 mg, 0.37 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded *N*^{6}-isopropyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (205 mg, 80%) as a viscous oil: R_f = 0.45 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.24 (t, *J* = 7.2 Hz, 9H), 1.30 (d, *J* = 6.6 Hz, 6H), 1.37 (s, 3H), 1.61 (s, 3H), 3.13 (q, *J* = 7.2 Hz, 6H), 3.43 (s, 3H), 4.34 (dd, *J* = 11.4, 4.2 Hz, 1H), 4.37 (dd, *J* = 11.4, 4.2 Hz, 1H), 4.42 (br s, 1H), 4.59−4.60 (m, 1H), 5.16 (s, 2H), 5.22 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.41 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.26 (d, *J* = 3.0 Hz, 1H), 6.97 (t, *J* = 7.8 Hz, 1H), 7.12 (d, *J* = 7.8 Hz, 1H), 7.27 (t, *J* = 7.8 Hz, 1H), 7.42 (d, *J* = 7.8 Hz, 1H), 8.24 (br s, 1H), 8.44 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 22.9, 25.6, 27.5, 43.5, 47.7, 56.6, 69.8, 83.4, 85.7, 91.7, 96.4, 115.2, 117.0, 120.2, 122.5, 129.7, 131.0, 132.5, 140.7, 149.7, 154.2, 155.3, 155.6, 176.6 (missing 1 Ar C); HRMS (ESI−) calcd for $C_{25}H_{31}N_6O_9S$ [M − H]$^-$ 591.1879, found 591.1898 (error 3.2 ppm).

N^6-Isopropyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (203 mg, 0.29 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by preparative HPLC using method 2 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (83 mg, 54%) as a white solid (reported NMR data are for the TEA salt of the title compound): mp >200 °C (dec); R_f = 0.53 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −23.9 (*c* 0.180, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 1.29 (d, *J* = 6.6 Hz, 6H), 3.15 (q, *J* = 7.2 Hz, 6H), 4.32−4.34 (m, 1H), 4.38 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.41−4.44 (m, 3H), 4.71 (t, *J* = 5.4 Hz, 1H), 6.09 (d, *J* = 6.0 Hz, 1H), 6.75−6.79 (m, 2H), 7.28 (t, *J* = 7.8 Hz, 1H), 7.94 (d, *J* = 8.4 Hz, 1H), 8.21 (br s, 1H), 8.43 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.1, 22.9, 43.6, 47.8, 69.7, 72.3, 75.9, 84.4, 89.2, 117.9, 119.3, 120.3, 120.6, 131.3, 134.4, 140.4, 150.0, 154.0, 155.3, 162.0, 174.7; HRMS (ESI−) calcd for $C_{20}H_{23}N_6O_8S$ [M − H]$^-$ 507.1304, found 507.1307 (error 0.6 ppm).

***N*^{6}-Isobutyl-2′,3′-*O*-isopropylideneadenosine (54).** The title compound was synthesized from **64** (500 mg, 1.74 mmol, 1.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (90:10 EtOAc/hexane) afforded the title compound (540 mg, 86%) as a viscous oil: R_f = 0.40 (EtOAc); ^{1}H NMR (600 MHz, $CDCl_3$) δ 0.95 (d, *J* = 6.6 Hz, 6H), 1.33 (s, 3H), 1.60 (s, 3H), 1.91 (hept, *J* = 6.6 Hz, 1H), 3.43 (br s, 2H), 3.74 (d, *J* = 12.6 Hz, 1H), 3.93 (d, *J* = 12.6 Hz, 1H), 4.49 (s, 1H), 5.07 (d, *J* = 6.0 Hz, 1H), 5.17 (t, *J* = 6.0 Hz, 1H), 5.80 (d, *J* = 6.0 Hz, 1H), 7.73 (s, 1H), 8.28 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 20.4, 25.5, 27.9, 28.8, 48.2, 63.7, 82.0, 83.2, 86.3, 94.6, 114.1, 121.5, 139.6, 147.4, 153.0, 155.8; HRMS (ESI+) calcd for $C_{17}H_{26}N_5O_4$ [M + H]$^+$ 364.1979, found 364.1993 (error 3.8 ppm).

***N*^{6}-Isobutyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (64).** Compound **54** (520 mg, 1.4 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (8:1 EtOAc/hexanes) afforded

the title compound (510 mg, 82%) as a viscous oil: R_f = 0.55 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.01 (d, *J* = 6.6 Hz, 6H), 1.40 (s, 3H), 1.61 (s, 3H), 1.99 (hept, *J* = 6.6 Hz, 1H), 3.41 (br s, 2H), 4.26 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.51−4.53 (m, 1H), 5.14 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.42 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.24 (d, *J* = 2.4 Hz, 1H), 8.23 (s, 1H), 8.25 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 20.6, 25.5, 27.6, 29.8, 49.7, 70.1, 83.2, 85.7, 85.9, 91.9, 115.8, 120.8, 140.9, 149.6, 154.3, 156.6; HRMS (ESI−) calcd for $C_{17}H_{26}ClN_6O_6S$ [M + Cl]$^-$ 477.1329, found 477.1331 (error 0.4 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-*N*^{6}-isobutyladenosine Sodium Salt (15). Compound **64** (160 mg, 0.36 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH//Et_3N) afforded *N*^{6}-isobutyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (170 mg, 67%) as a viscous oil: R_f = 0.51 (19:1 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.01 (d, *J* = 6.6 Hz, 6H), 1.28 (t, *J* = 7.2 Hz, 9H), 1.38 (s, 3H), 1.62 (s, 3H), 1.98 (hept, *J* = 6.6 Hz, 1H), 3.17 (q, *J* = 7.2 Hz, 6H), 3.40 (br s, 2H), 3.42 (s, 3H), 4.33 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.38 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.59−4.61 (m, 1H), 5.16 (s, 2H), 5.23 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.41 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.26 (d, *J* = 3.0 Hz, 1H), 6.97 (t, *J* = 7.8 Hz, 1H), 7.12 (d, *J* = 7.8 Hz, 1H), 7.27 (t, *J* = 7.8 Hz, 1H), 7.42 (d, *J* = 7.8 Hz, 1H), 8.23 (br s, 1H), 8.47 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 22.5, 25.6, 27.5, 29.7, 47.8, 48.8, 56.5, 69.9, 83.5, 85.7, 85.8, 91.8, 96.4, 115.2, 117.0, 120.4, 122.5, 129.7, 131.0, 132.4, 140.8, 149.6, 154.1, 155.6, 156.3, 176.8; HRMS (ESI−) calcd for $C_{26}H_{33}N_6O_9S$ [M − H]$^-$ 605.2035, found 605.2049 (error 2.3 ppm).

N^6-Isobutyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (150 mg, 0.21 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by preparative HPLC using method 2 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (26 mg, 23%) as a white solid: mp >200 °C (dec); R_f = 0.53 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −25.6 (*c* 0.156, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.01 (d, *J* = 6.6 Hz, 6H), 1.98 (hept, *J* = 6.6 Hz, 1H), 3.40 (br s, 2H), 4.31−4.33 (m, 1H), 4.37 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.40−4.43 (m, 2H), 4.70 (t, *J* = 5.4 Hz, 1H), 6.08 (d, *J* = 6.0 Hz, 1H), 6.75−6.79 (m, 2H), 7.28 (t, *J* = 7.8 Hz, 1H), 7.94 (d, *J* = 7.8 Hz, 1H), 8.20 (br s, 1H), 8.46 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 20.5, 29.7, 48.8, 69.5, 72.4, 76.1, 84.6, 89.2, 117.9, 119.2, 120.4, 120.6, 131.3, 134.4, 140.4, 149.9, 153.9, 156.3, 162.1, 175.2; HRMS (ESI−) calcd for $C_{21}H_{25}N_6O_8S$ [M − H]$^-$ 521.1460, found 521.1463 (error 0.6 ppm).

***N*^{6}-Cyclopropyl-2′,3′-*O*-isopropylideneadenosine (55).** The title compound was synthesized from **48** (176 mg, 0.61 mmol, 1.0 equiv) and cyclopropylamine (85 μL, 1.22 mmol, 2.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (50:50 EtOAc/hexane) afforded the title compound (200 mg, 94%): R_f = 0.4 (EtOAc); $[\alpha]_D^{20}$ −230.3 (*c* 1.0, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 0.67 (br s, 2H), 0.95 (q, *J* = 6.0 Hz, 2H), 1.37 (s, 3H), 1.63 (s, 3H), 3.06 (br s, 1H), 3.78 (d, *J* = 12.6 Hz, 1H), 3.96 (d, *J* = 13.2 Hz, 1H), 4.53 (s, 1H), 5.11 (d, *J* = 5.4 Hz, 1H), 5.19 (t, *J* = 5.4 Hz, 1H), 5.86 (d, *J* = 5.4 Hz, 1H), 7.81 (br s, 1H), 8.40 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 7.6, 23.9, 25.4, 27.8, 63.5, 81.9, 83.2, 86.3, 94.5, 114.2, 121.5, 140.0, 153.1, 156.2, 164.0; HRMS (APCI−) calcd for $C_{16}H_{20}N_5O_4$ [M + H]$^+$ 346.1521, found 346.1551 (error 8.7).

***N*^{6}-Cyclopropyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (65).** Compound **55** (200 mg, 0.58 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (50:50 EtOAc/hexane) provided the product (190 mg, 76%): R_f = 0.5 (EtOAc); $[\alpha]_D^{20}$ −57.9 (*c* 1.80, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 0.66−0.68 (m, 2H), 0.89−0.93 (m, 2H), 1.41 (s, 3H), 1.63 (s, 3H), 2.97 (br s, 1H), 4.30 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.36 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.55 (td, *J* = 5.4, 2.4 Hz, 1H), 5.16 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.45 (dd, *J* = 6.6, 2.4 Hz, 1H), 6.27 (d, *J* = 2.4 Hz, 1H), 8.24 (s, 1H), 8.33 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 7.7, 24.6 (br), 25.6, 27.5, 70.0, 83.0, 85.6, 85.8, 91.9, 115.7, 121.0, 141.3, 149.9 (br), 154.1, 157.2; HRMS (APCI−) calcd for $C_{16}H_{21}N_6O_6S$ [M − H]$^-$ 425.1249, found 425.1281 (error 7.5).

***N*^{6}-Cyclopropyl-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (16).** Compound **65** (150 mg, 0.352 mmol, 1.00 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded the title compound (84 mg, 39%): R_f = 0.3 (90:10 EtOAc/MeOH); $[\alpha]_D^{20}$ −46.4 (*c* 0.501, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 0.65−0.67 (m, 2H), 0.88−0.92 (m, 2H), 1.09 (t, *J* = 1.2 Hz, 9H), 2.65 (q, *J* = 7.2 Hz, 6H), 2.96 (br s, 1H), 4.32−4.35 (m, 1H), 4.39 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.25−4.44 (m, 2H), 4.72 (t, *J* = 5.4 Hz, 1H), 6.11 (d, *J* = 5.4 Hz, 1H), 6.78 (ovlp t, *J* = 7.8 Hz, 1H), 6.80 (ovlp d, *J* = 8.4 Hz, 1H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.95 (d, *J* = 7.8 Hz, 1H), 8.28 (br s, 1H), 8.48 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 7.7, 10.9, 22.2 (br), 47.7, 69.7, 72.5, 76.2, 84.7, 89.4, 118.0, 119.3, 120.7, 120.8, 131.5, 134.4, 140.9, 153.9, 157.2, 162.2, 175.2 (missing 1 aryl carbon); HRMS (ESI−) calcd for $C_{20}H_{21}N_6O_8S$ [M − H]$^-$ 505.1147, found 505.1161 (error 2.8 ppm).

***N*^{6}-Cyclobutyl-2′,3′-*O*-isopropylideneadenosine (56).** The title compound was synthesized from **48** (400 mg, 1.4 mmol, 1.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (3:1 EtOAc/hexane) provided the product (250 mg, 49%) as a white solid: mp 126−127 °C; R_f = 0.39 (EtOAc); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.37 (s, 3H), 1.64 (s, 3H), 1.77−1.82 (m, 2H), 1.96−2.03 (m, 2H), 2.45−2.51 (m, 2H), 3.77 (d, *J* = 12.6 Hz, 1H), 3.96 (d, *J* = 12.6 Hz, 1H), 4.53 (s, 1H), 4.76 (br s, 1H), 5.11 (d, *J* = 5.4 Hz, 1H), 5.19 (t, *J* = 5.4 Hz, 1H), 5 83 (d, *J* = 5.4 Hz, 1H), 7.77 (s, 1H), 8.31 (br s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 15.3, 25.4, 27.8, 31.7, 45.9, 63.6, 81.9, 83.1, 86.2, 94.6, 114.1, 121.3, 139.7, 147.5, 152.9, 154.4; HRMS (ESI+) calcd for $C_{17}H_{24}N_5O_4$ [M + H]$^+$ 362.1823, found 362.1846 (error 6.4 ppm).

***N*^{6}-Cyclobutyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (66).** Compound **56** (200 mg, 0.55 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (9:1 EtOAc/hexanes) afforded the title compound (220 mg, 91%) as a viscous oil: R_f = 0.53 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.38 (s, 3H), 1.60 (s, 3H), 1.77−1.82 (m, 2H), 2.05−2.11 (m, 2H), 2.41−2.46 (m, 2H), 4.26 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.5 Hz, 1H), 4.50−4.52 (m, 1H), 4.67 (br s, 1H), 5.12−5.13 (m, 1H), 5.39−5.41 (m, 1H), 6.23 (s, 1H), 8.21 (s, 1H), 8.23 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 15.9, 25.5, 27.4, 31.9, 47.0, 69.9, 82.9, 85.5, 85.7, 91.7, 115.6, 120.5, 140.9, 149.5, 154.1, 155.0; HRMS (ESI+) calcd for $C_{17}H_{25}N_6O_6S$ [M + H]$^+$ 441.1556, found 441.1558 (error 0.5 ppm).

***N*^{6}-Cyclobutyl-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (17).** Compound **66** (110 mg, 0.25 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded *N*^{6}-cyclobutyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (150 mg, 85%) as a viscous oil: R_f = 0.49 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 1.37 (s, 3H), 1.61 (s, 3H), 1.77−1.84 (m, 2H), 2.05−2.11 (m, 2H), 2.42−2.47 (m, 2H), 3.13 (q, *J* = 7.2 Hz, 6H), 3.42 (s, 3H), 4.31−4.39 (m, 2H), 4.60 (s, 1H), 4.68 (br s, 1H), 5.16 (s, 2H), 5.22 (d, *J* = 2.4 Hz, 1H), 5.39 (s, 1H), 6.25 (s, 1H), 6.97 (t, *J* = 7.2 Hz, 1H), 7.12 (d, *J* = 8.4 Hz, 1H), 7.27 (t, *J* = 7.2 Hz, 1H), 7.41 (d, *J* = 7.2 Hz, 1H), 8.23 (br s, 1H), 8.46 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 15.9, 25.6, 27.5, 31.9, 47.0, 47.8, 56.6, 69.9, 83.4, 85.72, 85.74, 91.8, 96.4, 115.2, 117.0, 120.2, 122.5, 129.7, 131.0, 132.5, 140.9, 149.8, 154.1, 155.0, 155.6, 176.7; HRMS (ESI−) calcd for $C_{26}H_{31}N_6O_9S$ [M − H]$^-$ 603.1879, found 603.1878 (error 0.2 ppm)

N^6-Cyclobutyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (120 mg, 0.17 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by preparative HPLC using method 1 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (18 mg, 20%) as a white solid (reported NMR data are for the TEA salt of the title compound): mp >200 °C (dec); R_f = 0.55 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −32.5 (*c* 0.117, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.14 (t, *J* = 7.2 Hz, 9H), 1.78−1.83 (m, 2H), 2.06−2.12 (m, 2H), 2.42−2.47 (m, 2H), 2.81 (q, *J* = 7.2 Hz, 6H), 4.30−4.32 (m, 1H), 4.37 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.40−4.43 (m, 2H), 4.62−4.71 (m, 2H), 6.08 (d, *J* = 6.0 Hz, 1H), 6.76−6.80 (m, 2H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.94 (d, *J* = 7.8 Hz, 1H), 8.20 (br s, 1H), 8.48 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 10.3, 15.9, 31.9, 47.1, 47.3, 69.6, 72.4, 76.1, 84.6, 89.1, 117.9, 119.2, 120.3, 120.6, 131.4, 134.3, 140.6, 150.2, 153.9, 155.0, 162.1, 175.0; HRMS (ESI−) calcd for $C_{21}H_{23}N_6O_8S$ [M − H]$^-$ 519.1304, found 519.1306 (0.4 ppm).

***N*^{6}-Cyclopentyl-2′,3′-*O*-isopropylideneadenosine (57).** The title compound was synthesized from **48** (400 mg, 1.4 mmol, 1.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (75:25 EtOAc/hexane) afforded the title compound (300 mg, 57%) as a viscous oil: R_f = 0.35 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.33 (s, 3H), 1.54−1.66 (m, 7H), 1.72−1.78 (m, 2H), 2.01−2.08 (m, 2H), 3.71 (dd, *J* = 12.0, 3.6 Hz, 1H), 3.78 (dd, *J* = 12.0, 3.0 Hz, 1H), 4.34−4.36 (m, 1H), 4.47 (br s, 1H), 5.01 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.24 (dd, *J* = 6.0, 3.8 Hz, 1H), 6.09 (d, *J* = 3.6 Hz, 1H), 8.20 (br s, 1H), 8.22 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.7, 25.6, 27.6, 33.9, 53.4, 63.6, 82.9, 85.2, 87.9, 93.0, 115.1, 120.9, 140.9, 148.9, 153.8, 155.6; HRMS (ESI+) calcd for $C_{18}H_{26}N_5O_4$ [M + H]$^+$ 376.1979, found 376.1982 (error 0.8 ppm).

***N*^{6}-Cyclopentyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (67).** Compound **57** (270 mg, 0.72 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (90:10 EtOAc/hexanes) afforded the title compound (170 mg, 78%) as a viscous oil: R_f = 0.49 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.38 (s, 3H), 1.58−1.71 (m, 7H), 1.77−1.83 (m, 2H), 2.05−2.11 (m, 2H), 4.26 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.33 (dd, *J* = 10.8, 4.5 Hz, 1H), 4.50−4.52 (m, 2H), 5.13 (dd, *J* = 6.3, 4.2 Hz, 1H), 5.41 (dd, *J* = 6.3, 3.0 Hz, 1H), 6.23 (d, *J* = 3.0 Hz, 1H), 8.21 (s, 1H), 8.25 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.7, 25.5, 27.4, 33.9, 53.6, 69.9, 82.9, 85.5, 85.7, 91.7, 115.6, 120.6, 140.7, 149.3, 154.2, 155.7; HRMS (ESI−) calcd for $C_{18}H_{25}N_6O_6S$ [M − H]$^-$ 453.1562, found 453.1536 (error 5.7 ppm).

***N*^{6}-Cyclopentyl-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (18).** Compound **67** (92 mg, 0.33 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded *N*^{6}-cyclopentyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (148 mg, 62%) as a viscous oil: R_f = 0.64 (80:20 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.26 (t, *J* = 7.2 Hz, 9H), 1.37 (s, 3H), 1.58−1.72 (m, 7H), 1.78−1.84 (m, 2H), 2.07−2.12 (m, 2H), 3.14 (q, *J* = 7.2 Hz, 6H), 3.42 (s, 3H), 4.33 (dd, *J* = 11.1, 3.9 Hz, 1H), 4.37 (dd, *J* = 11.1, 3.9 Hz, 1H), 4.37 (br s, 1H), 4.59−4.61 (m, 1H), 5.20 (s, 2H), 5.23 (d, *J* = 5.4 Hz, 1H), 5.39−5.41 (m, 1H), 6.23 (d, *J* = 3.6 Hz, 1H), 6.97 (t, *J* = 7.2 Hz, 1H), 7.12 (d, *J* = 7.2 Hz, 1H), 7.27 (t, *J* = 7.2 Hz, 1H), 7.41 (d, *J* = 7.2 Hz, 1H), 8.24 (br s, 1H), 8.46 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 24.7, 25.6, 27.5, 34.0, 47.8, 53.6, 56.6, 69.9, 83.5, 85.7, 85.8, 91.8, 96.4, 115.2, 117.0, 120.3, 122.5, 129.7, 131.0, 132.5, 133.9, 140.8, 149.5, 154.1, 155.6, 176.8; HRMS (ESI−) calcd for $C_{27}H_{33}N_6O_9S$ [M − H]$^-$ 617.2035, found 617.2043 (error 1.3 ppm).

N^6-Cyclopentyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (96 mg, 0.13 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by preparative HPLC using method 1 and conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (17 mg, 28%) as a white solid (reported NMR data are for the TEA salt of the title compound): mp >200 °C (dec); R_f = 0.53 (80:20 EtOAc/MeOH); $[\alpha]_D^{27}$ −28.8 (*c* 0.163, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 1.60−1.72 (m, 4H), 1.80−1.86 (m, 2H), 2.07−2.12 (m, 2H), 3.09 (q, *J* = 7.2 Hz, 6H), 4.30−4.32 (m, 1H), 4.37 (dd, *J* = 10.8, 2.4 Hz, 1H), 4.40−4.43 (m, 2H), 4.52 (br s, 1H), 4.71 (t, *J* = 5.4 Hz, 1H), 6.09 (d, *J* = 5.4 Hz, 1H), 6.77 (ovlp t, *J* = 7.8 Hz, 1H), 6.79 (ovlp d, *J* = 7.8 Hz, 1H), 7.29 (t, *J* = 7.8 Hz, 1H), 7.94 (d, *J* = 7.8 Hz, 1H), 8.22 (br s, 1H), 8.48 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 24.7, 34.0, 47.9, 53.5, 69.6, 72.4, 76.1, 84.6, 89.1, 117.9, 119.3, 120.4, 120.6, 131.4, 134.3, 140.4, 149.9, 154.0, 155.6, 162.1, 175.0; HRMS (ESI−) calcd for $C_{22}H_{25}N_6O_8S$ [M − H]$^-$ 533.1460, found 533.1450 (1.9 ppm).

***N*^{6}-Benzyl-2′,3′-*O*-isopropylideneadenosine (58).** The title compound was synthesized from **33** (400 mg, 1.4 mmol, 1.0 equiv) using the general one-pot procedure for N^6-amination and isopropylidene protection. Purification by flash chromatography (2:1 EtOAc/hexanes) afforded the title compound as a viscous oil (280 mg, 50%): R_f = 0.48 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.36 (s, 3H), 1.60 (s, 3H), 3.71 (dd, *J* = 12.0, 3.9 Hz, 1H), 3.78 (dd, *J* = 12.0, 3.6 Hz, 1H), 4.35−4.38 (m, 1H), 4.78 (br s, 2H), 5.03 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.26 (dd, *J* = 6.0, 3.6 Hz, 1H), 6.13 (d, *J* = 3.6 Hz, 1H), 7.21 (t, *J* = 7.2 Hz, 1H), 7.28 (t, *J* = 7.2 Hz, 2H), 7.35 (d, *J* = 7.2 Hz, 1H), 8.24 (br s, 2H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.6, 45.05, 63.6, 82.9, 85.2, 88.0, 92.9, 115.2, 121.1, 128.2, 128.5, 129.5, 140.2, 141.3, 149.3, 153.8, 156.1; HRMS (ESI+) calcd for $C_{20}H_{24}N_5O_4$ [M + H]$^+$ 398.1823, found 398.1840 (error 4.3 ppm).

***N*^{6}-Benzyl-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (68).** Compound **58** (180 mg, 0.45 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (4:1 EtOAc/hexanes) afforded the title compound (190 mg, 89%) as a viscous oil: R_f = 0.59 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.36 (s, 3H), 1.59 (s, 3H), 4.26 (dd, *J* = 10.5, 4.8 Hz, 1H), 4.34 (dd, *J* = 10.5, 4.8 Hz, 1H), 4.51 (td, *J* = 4.8, 3.0 Hz, 1H), 4.78 (br s, 2H), 5.12 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.39 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.22 (d, *J* = 2.4 Hz, 1H), 7.21 (t, *J* = 7.5 Hz, 1H), 7.28 (t, *J* = 7.5 Hz, 2H), 7.35 (d, *J* = 7.5 Hz, 1H), 8.19 (s, 1H), 8.26 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.5, 27.4, 45.0, 69.9, 82.9, 85.4, 85.6, 91.7, 115.6, 120.7, 128.2, 128.5, 129.5, 140.1, 140.9, 149.5, 154.1, 156.0; HRMS (ESI+) calcd for $C_{20}H_{25}N_6O_6S$ [M + H]$^+$ 477.1551, found 477.1554 (error 0.6 ppm).

***N*^{6}-Benzyl-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (19).** Compound **68** (130 mg, 0.27 mmol, 1.0 equiv) was coupled to **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded *N*^{6}-benzyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (140 mg, 70%) as a viscous oil: R_f = 0.55 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.24 (t, *J* = 7.2 Hz, 1H), 1.37 (s, 3H), 1.62 (s, 3H), 3.12 (q, *J* = 7.2 Hz, 6H), 3.41 (s, 3H), 4.34 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.37 (dd, *J* = 10.8, 3.6 Hz, 1H), 4.59−4.62 (m, 1H), 4.80 (br s, 2H), 5.15 (s, 2H), 5.23 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.42 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.25 (d, *J* = 3.0 Hz, 1H), 6.95 (t, *J* = 7.2 Hz, 1H), 7.11 (d, *J* = 8.4 Hz, 1H), 7.22−7.27 (m, 2H), 7.30 (t, *J* = 7.5 Hz, 2H), 7.37 (d, *J* = 7.8 Hz, 2H), 7.41 (d, *J* = 7.8 Hz, 1H), 8.26 (br s, 1H), 8.47 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 25.6, 27.5, 45.0, 47.8, 56.6, 69.9, 83.5, 85.7, 85.8, 91.9, 96.4, 115.2, 117.0, 120.5, 122.5, 128.2, 128.6, 129.5, 129.7, 131.0, 132.5, 140.3, 141.0, 150.0, 154.1, 155.6, 156.1, 176.7; HRMS (ESI−) calcd for $C_{29}H_{31}N_6O_9S$ [M − H]$^-$ 639.1879, found 639.1885 (error 0.9 ppm).

N^6-Benzyl-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)-benzoyl]sulfamoyl}adenosine triethylammonium salt (130 mg, 0.18 mmol, 1.0 equiv) prepared above was converted to the title

compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (44 mg, 42%) as a white solid: mp >200 °C (dec); R_f = 0.62 (4:1 EtOAc/MeOH); $[\alpha]_D^{27}$ −28.4 (*c* 0.134, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 4.32 (dd, *J* = 6.0, 3.0 Hz, 1H), 4.37 (dd, *J* = 11.4, 3.6 Hz, 1H), 4.40−4.43 (m, 2H), 4.70, (t, *J* = 5.4 Hz, 1H), 4.80 (br s, 2H), 6.10 (d, *J* = 6.0 Hz, 1H), 6.74−6.79 (m, 2H), 7.23−7.29 (m, 2H), 7.31 (t, *J* = 7.2 Hz, 2H), 7.38 (d, *J* = 7.2 Hz, 2H), 7.93 (dd, *J* = 7.8, 1.8 Hz, 1H), 8.23 (s, 1H), 8.47 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 45.1, 69.6, 72.3, 76.0, 84.5, 89.3, 117.9, 119.3, 120.5, 120.6, 128.2, 128.5, 129.5, 131.4, 134.4, 140.2, 140.6, 150.1, 153.9, 156.0, 162.0, 174.8; HRMS (ESI−) calcd for $C_{24}H_{23}N_6O_8S$ [M − H]$^-$ 555.1304, found 533.1304 (error 0 ppm).

8-Bromo-2′,3′-*O*-isopropylideneadenosine (70). 8-Bromoadenosine **69** (1.50 g, 4.33 mmol, 1.0 equiv) was converted to the title compound using the general procedure for acetonide protection. Purification by flash chromatography (4:1 EtOAc/hexanes) provided the product (1.09 g, 65%) as a white solid: R_f = 0.3 (EtOAc); $[\alpha]_D^{20}$ −16.2 (*c* 1.25, DMSO); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.36 (s, 3H), 1.66 (s, 3H), 3.78 (d, *J* = 12.6 Hz, 1H), 3.95 (d, *J* = 12.6 Hz, 1H), 4.51 (s, 1H), 5.06 (d, *J* = 5.4 Hz, 1H), 5.26 (t, *J* = 5.4 Hz, 1H), 5.94 (s, 2H, NH_2), 6.09 (d, *J* = 5.4 Hz, 1H), 8.25 (s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 25.7, 28.0, 63.5, 81.8, 82.7, 86.0, 94.0, 114.3, 120.9, 127.4, 150.1, 152.7, 154.8; MS (APCI+) calcd for $C_{13}H_{17}BrN_5O_4$ [M + H]$^+$ 386.0, found 386.1.

8-Bromo-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (72). Compound **70** (500 mg, 1.29 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (7:3 EtOAc/hexane) provided the product (510 mg, 85%) as a colorless oil: R_f = 0.6 (EtOAc); $[\alpha]_D^{20}$ +1.0 (*c* 2.5, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.37 (s, 3H), 1.57 (s, 3H), 4.14 (dd, *J* = 10.2, 7.2 Hz, 1H), 4.28 (dd, *J* = 10.2, 6.0 Hz, 1H), 4.41−4.43 (m, 1H), 5.24 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.73 (d, *J* = 6.0 Hz, 1H), 6.21 (s, 1H), 8.17 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.5, 27.5, 69.7, 83.5, 84.7, 86.9, 93.0, 115.6, 121.0, 128.6, 151.4, 154.2, 156.4; MS (APCI+) calcd for $C_{13}H_{18}BrN_6O_6S$ [M + H]$^+$ 465.0, found 465.2.

8-Bromo-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (20). Compound **72** (460 mg, 0.98 mmol, 1.00 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (9:1 EtOAc/MeOH) provided the product (365 mg, 57%) as a white solid: R_f = 0.2 (1:4 MeOH/EtOAc); $[\alpha]_D^{20}$ −5.7 (*c* 1.0, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.28 (t, *J* = 7.2 Hz, 9H), 3.17 (q, *J* = 7.2 Hz, 6H), 4.30 (td, *J* = 6.0, 4.8 Hz, 1H), 4.35 (dd, *J* = 10.2, 6.0 Hz, 1H), 4.53 (dd, *J* = 10.2, 6.0 Hz, 1H), 4.63 (t, *J* = 4.8 Hz, 1H), 5.39 (t, *J* = 6.0 Hz, 1H), 6.03 (d, *J* = 6.0 Hz, 1H), 6.76−6.81 (m, 2H), 7.30 (t, *J* = 7.8 Hz, 1H), 7.90 (d, *J* = 7.8 Hz, 1H), 8.03 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 47.9, 69.6, 72.3, 72.4, 84.2, 92.2, 117.8, 119.2, 120.7, 121.1, 129.1, 131.2, 134.2, 152.0, 153.8, 156.1, 161.9, 174.7; HRMS (ESI−) calcd for $C_{17}H_{16}BrN_6O_8S$ [M − H]$^-$ 542.9939, found 542.9937 (error 0.4 ppm).

8-Azido-2′,3′-*O*-isopropylideneadenosine (71). Compound **70** (550 mg, 1.42 mmol, 1.0 equiv) and NaN_3 (370 mg, 5.69 mmol, 4 equiv) were heated in DMF (10 mL) at 70 °C for 12 h. The reaction mixture was concentrated in vacuo and the residue was purified by flash chromatography (1:1 EtOAc/hexane) to afford the title compound (410 mg, 83%) as a white solid: R_f = 0.4 (EtOAc); $[\alpha]_D^{20}$ −28 (*c* 1.2, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.35 (s, 3H), 1.63 (s, 3H), 3.75 (d, *J* = 12.6 Hz, 1H), 3.92 (d, *J* = 12.6 Hz, 1H), 4.63 (s, 1H), 5.05 (d, *J* = 5.4 Hz, 1H), 5.19 (t, *J* = 5.4 Hz, 1H), 5.82 (d, *J* = 5.4 Hz, 1H), 8.19 (s, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 25.5, 27.8, 63.5, 81.7, 82.7, 85.9, 91.4, 114.1, 118.5, 145.4, 149.2, 151.7, 154.0; MS (APCI+) calcd for $C_{13}H_{17}N_8O_4$ [M + H]$^+$ calcd 349.1, found 349.1.

8-Azido-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (73). Compound **71** (390 mg, 1.12 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (4:1 EtOAc/hexane) afforded the title compound (230 mg, 48%) as a white solid: R_f = 0.5 (EtOAc); $[\alpha]_D^{20}$ −0.29 (*c* 2.0, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.36 (s, 3H), 1.56 (s, 3H), 4.17 (dd, *J* = 10.2, 5.4 Hz, 1H), 4.30 (dd, *J* = 10.2, 5.4 Hz, 1H), 4.38 (td, *J* = 5.4, 3.0, Hz, 1H), 5.16 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.54 (d, *J* = 6.0 Hz, 1H), 6.03 (s, 1H), 8.10 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.5, 69.7, 83.1, 84.7, 86.4, 90.0, 115.6, 118.7, 146.6, 150.8, 153.2, 155.7; MS (APCI+) calcd for $C_{13}H_{18}N_9O_6S$ [M + H]$^+$ 428.1, found 428.1.

8-Azido-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (21). Compound **73** (427 mg, 1.05 mmol, 1.00 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (95:5:1 EtOAc/MeOH/Et_3N) afforded the title compound (350 mg, 55%): R_f = 0.2 (1:9 MeOH/EtOAc); $[\alpha]_D^{20}$ −21.6 (*c* 1.0, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.28 (t, *J* = 2.4 Hz, 9H), 3.18 (q, *J* = 7.2 Hz, 6H), 4.27 (q, *J* = 5.4 Hz, 1H), 4.38 (dd, *J* = 10.2, 6.6 Hz, 1H), 4.50 (dd, *J* = 12.0, 5.4 Hz, 1H), 4.58 (t, *J* = 5.4 Hz, 1H), 5.10 (t, *J* = 5.4 Hz, 1H), 5.90 (d, *J* = 5.4 Hz, 1H), 6.77−6.81 (m, 2H), 7.30 (t, *J* = 7.8 Hz, 1H), 7.92 (d, *J* = 7.8 Hz, 1H), 8.04 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 48.0, 70.0, 72.3, 72.9, 83.9, 89.3, 118.0, 118.6, 119.3, 120.7, 131.3, 134.4, 147.0, 151.4, 152.9, 155.5, 162.0, 174.8. HRMS (ESI−) calcd for $C_{17}H_{16}N_9O_8S$ [M − H]$^-$ 506.0848, found 506.0849 (error 0.2 ppm).

8-Amino-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (22). Compound **21** (100 mg, 0.164 mmol, 1.0 equiv) was treated with Pd/C (20.0 mg) in MeOH (10 mL) under a H_2 (1 atm) for 2 h at room temperature. The mixture was filtered through Celite and concentrated. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded the title compound (69 mg, 73%) as a white solid: R_f = 0.3 (80:20 EtOAc/MeOH); $[\alpha]_D^{20}$ −19 (*c* 1.0, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.28 (t, *J* = 7.2 Hz, 9H), 3.18 (q, *J* = 7.2 Hz, 6H), 4.28−4.32 (m, 1H), 4.38−4.46 (m, 3H), 4.87−4.90 (ovlp m, 1H), 6.04 (d, *J* = 7.2 Hz, 1H), 6.79−6.83 (m, 2H), 7.32 (t, *J* = 7.8 Hz, 1H), 7.96 (d, *J* = 7.8 Hz, 1H), 7.98 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.35, 48.0, 69.7, 71.9, 72.0, 84.6, 88.6, 117.8, 118.0, 119.4, 120.6, 131.5, 134.6, 150.4, 151.1, 153.5, 153.8, 162.1, 175.2; HRMS (ESI−) calcd for $C_{17}H_{18}N_7O_8S$ [M − H]$^-$ 480.0943, found 480.0960 (error 3.5 ppm).

2-Iodo-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (82). Compound **75**[31,98,99] (150 mg, 0.35 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash column chromatography (80:20 EtOAc/hexane) provided the product (160 mg, 90%) as a viscous oil: R_f = 0.55 (3:1 EtOAc/hexanes); $[\alpha]_D^{20}$ −0.92 (*c* 1.3, CH_3OH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.37 (s, 3H), 1.58 (s, 3H), 4.27 (dd, *J* = 11.1, 5.4 Hz, 1H), 4.32 (dd, *J* = 11.1, 5.4 Hz, 1H), 4.4−4.52 (m, 1H), 5.06−5.09 (m, 1H), 5.32 (d, *J* = 6.0 Hz, 1H), 6.18 (s, 1H), 8.12 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.3, 26.2, 68.8, 81.7, 84.4, 84.9, 90.3, 114.4, 119.0, 119.6, 139.9, 149.4, 155.9; HRMS (ESI+) calcd for $C_{13}H_{18}IN_6O_6S$ [M + H]$^+$ 513.0048, found 513.0056 (error 1.6 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-2-iodoadenosine Triethylammonium Salt (23). Compound **82** (112 mg, 0.21 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded the title compound (100 mg, 69%) as a white solid: R_f = 0.4 (1:9 MeOH/EtOAc); $[\alpha]_D^{20}$ −4.5 (*c* 0.32, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.27 (t, *J* = 7.2 Hz, 9H), 3.18 (q, *J* = 7.2 Hz, 6H), 4.31−4.33 (m, 1H), 4.38 (dd, *J* = 11.4, 3.6 Hz, 1H), 4.41−4.43 (m, 2H), 4.65 (t, *J* = 4.8 Hz, 1H), 6.01 (d, *J* = 5.4 Hz, 1H), 6.74−6.82 (m, 2H), 7.28 (dt, *J* = 8.4, 1.8 Hz, 1H), 7.93 (dd, *J* = 7.8, 1.8 Hz, 1H), 8.36 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 47.9, 69.8, 72.4, 76.1, 84.5, 89.5, 118.0, 119.4, 120.1, 120.7, 120.9, 131.4, 134.5, 140.8, 151.3, 157.1, 162.1, 174.9. HRMS (ESI−) calcd for $C_{17}H_{16}IN_6O_8S$ [M − H]$^-$ 590.9800, found 590.9784 (error 2.7 ppm).

2-Anilino-2′,3′-*O*-isopropylideneadenosine (76). Compound **76** was prepared as described.[32] Results from ^{1}H NMR and HRMS agreed with reported analytical data.[32] Additional data for compound **76**: R_f = 0.3 (1:19 MeOH/EtOAc); $[\alpha]_D^{20}$ −27.6 (*c* 0.31, CH_3OH); ^{13}C NMR (150 MHz, CD_3OD) δ 25.7, 27.6, 63.5, 8.1, 85.4, 88.5, 92.3, 115.2, 115.4, 120.8, 122.8, 129.6, 139.3, 142.3, 151.9, 157.6, 158.5.

2-Anilino-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (83). Compound **76** (90 mg, 0.23 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash column chromatography (1:19 MeOH/EtOAc) provided the product (40 mg, 37%) as a viscous oil: R_f = 0.55 (1:19 MeOH/EtOAc); $[\alpha]_D^{20}$ +35.9 (*c* 0.472, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.39 (s, 3H), 1.59 (s, 3H), 4.17 (dd, *J* = 10.2, 5.4 Hz, 1H), 4.26 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.44−4.50 (m, 1H), 5.02 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.47 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.12 (d, *J* = 2.4 Hz 1H), 6.96 (t, *J* = 7.8 Hz), 7.27 (t, *J* = 7.8 Hz, 2H), 7.64 (d, *J* = 7.8 Hz, 2H), 7.93 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.5, 70.1, 83.1, 85.4, 85.9, 92.1, 115.4, 115.5, 121.2, 123.0, 129.7, 139.1, 142.2, 151.9, 157.5, 158.6; HRMS (ESI+) calcd for $C_{19}H_{24}N_7O_6S$ $[M + H]^+$ 478.1503, found 478.1497 (error 1.3 ppm).

2-Anilino-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Triethylammonium Salt (24). Compound **83** (40 mg, 0.083 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (1:4 MeOH/EtOAc) afforded the title compound (24.4 mg, 45%) as a viscous oil: R_f = 0.7 (75:25:1 EtOAc/MeOH/Et_3N); $[\alpha]_D^{20}$ +6.5 (*c* 0.20, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 3.14 (q, *J* = 7.2 Hz, 6H), 4.28−4.32 (m, 1H), 4.35 (dd, *J* = 11.4, 3.6 Hz, 1H), 4.41 (dd, *J* = 4.8, 3.0 Hz, 1H), 4.43 (dd, *J* = 11.4, 3.6 Hz, 1H), 4.78 (t, *J* = 5.4 Hz, 1H), 6.10 (d, *J* = 6.0 Hz, 1H), 6.74−6.84 (m, 2H), 6.92 (t, *J* = 7.2 Hz, 1H), 7.24−7.32 (m, 3H), 7.68 (d, *J* = 7.8 Hz, 2H), 7.94 (dd, *J* = 7.8, 1.8 Hz, 1H), 8.18 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 48.0, 68.9, 72.6, 75.6, 84.4, 89.5, 115.2, 117.9, 119.4, 120.3, 120.8, 122.5, 129.7, 131.5, 134.4, 138.9, 142.4, 152.9, 157.4, 158.5, 162.2, 175.5; HRMS (ESI−) calcd for $C_{23}H_{22}N_7O_8S$ $[M - H]^-$ 556.1256, found 556.1252 (0.7 ppm error).

2′,3′-*O*-Isopropylidene-2-(phenylethynyl)adenosine (77). A solution of **75** (100 mg, 0.23 mmol, 1.0 equiv), $PdCl_2(PPh_3)_2$ (16.2 mg, 23.1 μmol, 0.1 equiv), CuI (8.8 mg, 46.2 μmmol, 0.2 equiv), Et_3N (68 μL, 0.48 mmol, 2.1 equiv) in DMF (0.4 mL) was heated at 80 °C for 16 h. The mixture was cooled to room temperature, diluted with $CHCl_3$ (10 mL), filtered through Celite, and the filtrate was concentrated. Purification by flash chromatography (95:5 EtOAc/MeOH) afforded the title compound (53 mg, 57%) as a viscous oil: R_f = 0.3 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.40 (s, 3H), 1.66 (s, 3H), 3.83 (d, *J* = 12.6 Hz, 1H), 3.98 (d, *J* = 12.6 Hz, 1H), 4.51 (s, 1H), 5.12 (d, *J* = 5.4 Hz, 1H), 5.21 (t, *J* = 5.4 Hz, 1H), 5.99 (d, *J* = 5.4 Hz, 1H), 7.35−7.45 (m, 3H), 7.66 (d, *J* = 7.8 Hz, 2H), 8.11 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 29.2, 31.4, 67.1, 85.6, 87.5, 90.2, 90.7, 91.6, 97.3, 118.4, 123.7, 125.5, 132.5, 133.6, 136.5, 145.2, 150.6, 159.7 (missing 1 Ar C); HRMS (ESI+) calcd for $C_{21}H_{22}N_5O_4S$ $[M + H]^+$ 408.1666, found 408.1635 (error 7.6 ppm).

2′,3′-*O*-Isopropylidene-2-phenylethynyl-5′-*O*-(sulfamoyl)adenosine (84). Compound **77** (50 mg, 0.12 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash chromatography (1:49 MeOH/EtOAc) provided the product (39 mg, 66%) as a viscous oil: R_f = 0.5 (1:19 MeOH/EtOAc); $[\alpha]_D^{20}$ −15.0 (*c* 0.162, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.41 (s, 3H), 1.63 (s, 3H), 4.31 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.36 (dd, *J* = 11.4, 4.8 Hz, 1H), 4.52−4.58 (m, 1H), 5.14 (dd, *J* = 6.0, 3.0 Hz, 1H), 5.40 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.26 (d, *J* = 3.0 Hz, 1H), 7.38−7.48 (m, 3H), 7.61 (d, *J* = 7.8 Hz, 2H), 8.33 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.6, 27.6, 70.1, 83.0, 85.6, 85.7, 86.3, 89.3, 91.7, 115.9, 123.1, 129.8, 130.0, 130.7, 133.4, 142.3, 147.9, 157.3 (missing 1 Ar C); HRMS (ESI+) calcd for $C_{21}H_{23}N_6O_6S$ $[M + H]^+$ 487.1394, found 487.1395 (error 0.2 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-2-(phenylethynyl)adenosine Triethylammonium Salt (25). Compound **84** (35 mg, 0.072 mmol, 1.0 equiv) was converted to the title compound using the general procedure for salicylation and TFA deprotection. Purification by flash chromatography (95:5:1 EtOAc/MeOH/Et_3N) afforded the product (25 mg, 52%) as a viscous oil: R_f = 0.35 (1:9 MeOH/EtOAc); $[\alpha]_D^{20}$ +10.9 (*c* 0.110, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 3.14 (q, *J* = 7.2 Hz, 6H), 4.32−4.36 (m, 1H), 4.38−4.49 (m, 3 H), 4.66 (t, *J* = 5.4 Hz, 1H), 6.12 (d, *J* = 5.4 Hz, 1H), 6.70−6.82 (m, 2H), 7.28 (t, *J* = 7.2 Hz, 1H), 7.38−7.46 (m, 3H), 7.62 (d, *J* = 8.4 Hz, 2H), 7.94 (d, *J* = 7.2 Hz, 1H), 7.59 (br s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 48.0, 61.6, 69.6, 72.3, 76.5, 84.7, 86.2, 89.4, 117.9, 119.3, 120.8, 123.2, 129.8, 130.7, 131.5, 133.3, 134.4, 142.0, 147.8, 151.2, 157.2, 162.2, 175.2 (missing 1 Ar C); HRMS (ESI−) calcd for $C_{25}H_{21}N_6O_8S$ $[M - H]^-$ 565.1147, found 565.1140 (error 1.2 ppm).

2′,3′-*O*-Isopropylidene-2-phenyladenosine (78). Compound **78** was prepared as described.[32]Results from ^{1}H NMR and HRMS agreed with reported analytical data.[32] Additional data for compound **78**: R_f = 0.5 (EtOAc); $[\alpha]_D^{20}$ −2.6 (*c* 0.41, MeOH); ^{13}C NMR (150 MHz, CD_3OD) δ 25.7, 27.7, 63.5, 83.2, 85.5, 88.7, 92.3, 115.4, 119.5, 129.3, 129.4, 131.0, 139.8, 142.1, 151.6, 157.4, 161.2.

2′,3′-*O*-Isopropylidene-2-phenyl-5′-*O*-(sulfamoyl)adenosine (85). Compound **78** (40 mg, 0.10 mmol, 1.0 equiv) was converted to the title compound using the general procedure for sulfamoylation. Purification by flash column chromatography (9:1 EtOAc/hexane) provided the product as a viscous oil (35 mg, 66%): R_f = 0.4 (EtOAc); $[\alpha]_D^{20}$ +40.2 (*c* 0.781, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.40 (s, 3H), 1.63 (s, 3H), 4.27 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.34 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.50−4.60 (ovlp m, 1H), 5.18 (dd, *J* = 5.4, 2.4 Hz, 1H), 5.50 (dd, *J* = 6.0, 1.8 Hz, 1H), 6.27 (d, *J* = 2.4 Hz, 1H), 7.40−7.50 (m, 3H), 8.12 (s, 1H), 8.20−8.35 (m, 2H); ^{13}C NMR (150 MHz, CD_3OD) δ 24.7, 26.6, 68.5, 81.3, 84.0, 84.2, 90.2, 114.4, 117.8, 127.8, 128.1, 129.8, 137.7, 139.9, 149.9, 155.4, 159.7; HRMS ESI (+) calcd for $C_{19}H_{23}N_6O_6S$ $[M + H]^+$ 463.1394, found 463.1397 (error 0.6 ppm).

5′-*O*-[*N*-(2-Hydroxybenzoyl)sulfamoyl]-2-phenyladenosine Triethylammonium Salt (26). Compound **85** (32 mg, 0.068 mmol, 1.0 equiv) was coupled to compound **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded 2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}-2-phenyladenosine triethylammonium salt (39.3 mg, 87%): R_f = 0.38 (EtOAc); ^{1}H NMR (600 MHz, CD_3OD) δ 1.23 (t, *J* = 7.2 Hz, 9H), 1.40 (s, 3H), 1.65 (s, 3H), 3.12 (q, *J* = 7.2 Hz, 1H), 3.40 (s, 3H), 4.33−4.44 (m, 2H), 4.61−4.67 (m, 1H), 5.13 (s, 2H), 5.28 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.53 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.37 (d, *J* = 3.0 Hz, 1H), 6.94 (t, *J* = 7.2 Hz, 1H), 7.10 (d, *J* = 9.0 Hz, 1H), 7.25 (t, *J* = 7.8 Hz, 1H), 7.37 (d, *J* = 7.8 Hz, 1H), 7.40−7.46 (m, 3H), 8.37 (d, *J* = 7.8 Hz, 2H), 8.44 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.2, 25.7, 27.7, 47.9, 56.6, 70.1, 83.5, 85.8, 85.9, 91.8, 96.4, 106.4, 115.4, 117.0, 119.1, 122.6, 129.3, 129.8, 130.9, 131.1, 132.5, 139.7, 141.7, 151.8, 155.7, 157.3, 161.1, 176.7; HRMS (ESI+) calcd for $C_{28}H_{31}N_6O_9S$ $[M - H]^-$ 625.1722, found 627.1720 (error 0.3 ppm).

2′,3′-*O*-Isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl-]sulfamoyl}-2-phenyladenosine triethylammonium salt (23 mg, 0.032 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded the title compound (16.6 mg, 83%): R_f = 0.2 (3:7 MeOH/EtOAc); $[\alpha]_D^{20}$ +7.4 (*c* 0.56, MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.25 (t, *J* = 7.2 Hz, 9H), 3.14 (q, *J* = 7.2 Hz, 6H), 4.32−4.37 (m, 1H), 4.39 (dd, *J* = 11.4, 3.6 Hz, 1H), 4.45 (dd, *J* = 11.4, 3.6 Hz, 1H,), 4.49 (t, *J* = 4.2 Hz, 1H), 4.82−4.86 (m, 1H), 6.22 (d, *J* = 5.4 Hz, 1H), 6.74−6.84 (m, 2H), 7.28 (dt, *J* = 8.4, 1.8 Hz, 1H), 7.38−7.46 (m, 3H), 7.94 (dd, *J* = 7.8, 1.8 Hz, 1H), 8.36 (dd, *J* = 8.4, 1.8 Hz, 2H), 8.47 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.3, 48.0, 69.8, 72.6, 75.9, 84.5, 89.3, 117.9, 119.1, 119.4, 120.7, 129.3, 129.4, 130.9, 131.4, 134.4, 139.8, 141.5, 152.3, 157.2, 161.1, 162.1, 175.1; HRMS (ESI+) calcd for $C_{23}H_{23}N_6O_8S$ $[M + H]^+$ 543.1293, found 543.1291 (error 0.4 ppm).

2-(Biphen-2-yl)-2′,3′-*O*-isopropylideneadenosine (79). To a solution of compound **75** (150 mg, 0.346 mmol, 1.0 equiv) in 1,4-dioxane (5.0 mL) were added 2-biphenylboronic acid (137 mg, 0.69 mmol, 2.0 equiv), $Pd(OAc)_2$ (15.5 mg, 0.0692 mmol, 0.2 equiv), 2-(biphenyl)dicyclohexylphosphine (36.4 mg, 0.104 mmol, 0.30 equiv), K_3PO_4 (183 mg, 0.87 mmol, 2.5 equiv), and the mixture was heated at 100 °C for 12 h. After cooling to room temperature, the mixture was diluted with MeOH (20 mL), filtered, and concentrated. Purification by flash chromatography (gradient from 80−100% EtOAc/hexane) afforded the title compound (92 mg, 58%) as a white solid: R_f = 0.30 (80:20 EtOAc/hexane); ^{1}H NMR (600 MHz, CD_3OD) δ 1.40 (s, 3H), 1.58 (s, 3H), 3.52 (dd, *J* = 12.0, 4.2 Hz, 1H), 3.56 (dd, *J* = 12.0, 4.2 Hz, 1H), 4.24−4.26 (m, 1H), 4.45 (dd, *J* = 6.0, 4.2 Hz, 1H), 4.71 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.89 (d, *J* = 4.2 Hz, 1H), 7.18−7.26 (m, 5H), 7.45−7.49 (m, 2H), 7.53 (t, *J* = 7.8 Hz, 1H), 7.71 (d, *J* = 7.8 Hz, 1H), 8.17 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 26.1, 27.8, 63.6, 83.1, 84.2, 87.7, 93.4, 115.0, 119.3, 127.7, 128.1, 128.9, 130.2, 130.3, 131.4, 131.5, 139.8, 142.5, 142.6, 143.1, 150.1, 157.3, 163.7; HRMS (ESI+) calcd for $C_{25}H_{26}N_5O_4$ $[M + H]^+$ 460.1979, found 460.2005 (error 5.7 ppm).

2-(Biphen-2-yl)-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (86). Sulfamoyl chloride (78 mg, 0.68 mmol, 4.0 equiv) was added to a solution of **79** (78 mg, 0.17 mmol, 1.0 equiv) in DMA (2.5 mL) at 0 °C. The solution was stirred for 4 h at 0 °C and then partitioned between EtOAc (40 mL) and H_2O (40 mL). The organic layer was separated, washed with H_2O (3 × 40 mL), saturated aqueous NaCl (40 mL), dried (Na_2SO_4), and concentrated to afford the title compound (87 mg, 96%) as a white solid: R_f = 0.29 (80:20 EtOAc/benzene); ^{1}H NMR (600 MHz, CD_3OD) δ 1.37 (s, 3H), 1.53 (s, 3H), 4.06 (dd, *J* = 10.8, 6.6 Hz, 1H), 4.12 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.36−4.40 (m, 1H), 4.61 (dd, *J* = 5.4, 2.4 Hz, 1H), 4.69 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.97 (d, *J* = 2.4 Hz, 1H), 7.17−7.24 (m, 5H), 7.44−7.46 (m, 2H), 7.50 (t, *J* = 7.8 Hz, 1H), 7.72 (d, *J* = 7.2 Hz, 1H), 8.13 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 26.3, 27.6, 69.7, 83.3, 84.7, 85.5, 92.5, 115.3, 118.9, 127.7, 128.3, 129.1, 130.06, 130.14, 131.6, 131.7, 140.0, 142.3, 142.4, 143.3, 150.3, 157.2, 163.7; HRMS (ESI+) calcd for $C_{25}H_{27}N_6O_6S$ $[M + H]^+$ 539.1707, found 539.1716 (error 1.7 ppm).

2-(Biphen-2-yl)-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (27). Compound **86** (74 mg, 0.14 mmol, 1.0 equiv) was coupled to compound **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded 2-(biphen-2-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (87 mg, 79%): R_f = 0.56 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.19 (t, *J* = 7.2 Hz, 9H), 1.36 (s, 3H), 1.54 (s, 3H), 3.07 (q, *J* = 7.2 Hz, 6H), 3.39 (s, 3H), 4.20 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.23 (dd, *J* = 10.8, 4.8 Hz, 1H), 4.49−4.53 (m, 1H), 4.84−4.87 (m, 1H), 4.88−4.94 (ovlp m, 1H), 5.11−5.13 (m, 2H), 6.00 (d, *J* = 3.0 Hz, 1H), 6.96 (t, *J* = 7.8 Hz, 1H), 7.10 (d, *J* = 8.4 Hz, 1H), 7.16−7.23 (m, 5H), 7.27 (t, *J* = 7.8 Hz, 1H), 7.37 (d, *J* = 7.8 Hz, 1H), 7.41−7.44 (m, 2H), 7.48 (t, *J* = 7.8 Hz, 1H), 7.72 (d, *J* = 7.8 Hz, 1H), 8.34 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.1, 26.2, 27.7, 47.6, 56.6, 69.8, 83.4, 85.0, 85.5, 92.0, 96.2, 115.1, 116.9, 118.6, 122.5, 127.6, 128.2, 129.0, 129.7, 130.0, 131.0, 131.49, 131.52, 132.3, 140.1, 142.1, 142.6, 143.2, 150.7, 155.6, 157.0, 163.7, 176.6 (missing 1 Ar C); HRMS (ESI−) calcd for $C_{34}H_{33}N_6O_9S$ $[M - H]^-$ 701.2035, found 701.2015 (error 2.9 ppm).

2-(Biphen-2-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (87 mg, 0.11 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N), followed by conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (56 mg, 80%): R_f = 0.35 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 4.22−4.29 (m, 4H), 4.36 (dd, *J* = 10.8, 3.0 Hz, 1H), 5.82 (d, *J* = 4.2 Hz, 1H), 6.76−6.81 (m, 2H), 7.15−7.21 (m, 5H), 7.30 (t, *J* = 7.2 Hz, 1H), 7.41−7.45 (m, 2H), 7.48 (t, *J* = 7.2 Hz, 1H), 7.67 (d, *J* = 7.8 Hz, 1H), 7.93 (d, *J* = 7.2 Hz, 1H), 8.37 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 69.4, 71.7, 75.5, 84.0, 90.2, 117.9, 118.6, 119.3, 120.6, 127.7, 128.1, 129.0, 130.1, 131.2, 131.3, 131.4, 140.2, 141.5, 142.7, 143.0, 150.8, 156.8, 162.1, 163.7, 175.2 (missing 2 Ar C); HRMS (ESI−) calcd for $C_{29}H_{25}N_6O_8S$ $[M - H]^-$ 617.1460, found 617.1463 (error 0.5 ppm).

2-(Biphen-3-yl)-2′,3′-*O*-isopropylideneadenosine (80). To a solution of compound **75** (300 mg, 0.69 mmol, 1.0 equiv) in 1,4-dioxane (10 mL) were added 3-biphenylboronic acid (206 mg, 1.04 mmol, 1.5 equiv), $Pd(OAc)_2$ (15.5 mg, 0.0692 mmol, 0.2 equiv), 2-(biphenyl)dicyclohexylphosphine (36.4 mg, 0.104 mmol, 0.30 equiv), K_3PO_4 (294 mg, 1.38 mmol, 2.0 equiv), and the reaction was heated at 100 °C for 18 h. After cooling to room temperature, the mixture was diluted with EtOAc (50 mL), filtered, and concentrated. Purification by flash chromatography (gradient from 80−100% EtOAc/hexane) afforded the title compound (215 mg, 68%) as a white solid contaminated with approximately 10 mol% of **75**. The solid was recrystallized from EtOAc (15 mL) by refluxing for 15 min. The solution was filtered while hot and the filtrate cooled to room temperature, then placed at −20 °C for 8 h. The title compound was isolated as a white crystalline solid (116 mg, 36%) from the first crop: R_f = 0.20 (80:20 EtOAc/hexane); ^{1}H NMR (600 MHz, DMSO-d_6) δ 1.36 (s, 3H), 1.58 (s, 3H), 3.51−3.59 (m, 2H), 4.22 (td, *J* = 5.4, 2.4 Hz, 1H), 5.02 (t, *J* = 6.0 Hz, 1H), 5.10 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.56 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.27 (d, *J* = 2.4 Hz, 1H), 7.38−7.44 (m, 3H), 7.51 (t, *J* = 7.2 Hz, 2H), 7.58 (t, *J* = 7.2 Hz, 1H), 7.73−7.76 (m, 3H), 8.34−8.35 (m, 2H), 8.66 (s, 1H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 25.9, 27.7, 62.3, 82.3, 84.1, 87.6, 90.0, 113.7, 119.0, 126.8, 126.9, 127.39, 127.41, 128.3, 128.7, 129.7, 139.6, 140.89, 140.90, 141.1, 150.5, 156.6, 158; HRMS (APCI+) calcd for $C_{25}H_{26}N_5O_4$ $[M + H]^+$ 460.1979, found 460.2025 (error 10.0 ppm).

2-(Biphen-3-yl)-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (87). Sulfamoyl chloride (78 mg, 0.68 mmol, 4.0 equiv) was added to a solution of **80** (78 mg, 0.17 mmol, 1.0 equiv) in DMA (2.5 mL) at 0 °C. The solution was stirred 4 h at 0 °C and then partitioned between EtOAc (40 mL) and H_2O (40 mL). The organic layer was separated, washed with H_2O (3 × 40 mL), saturated aqueous NaCl (40 mL), dried (Na_2SO_4), and concentrated to afford the title compound (87 mg, 96%) as a white solid: R_f = 0.40 (80:20 EtOAc/benzene); ^{1}H NMR (600 MHz, CD_3OD) δ 1.38 (s, 3H), 1.60 (s, 3H), 4.14 (dd, *J* = 10.8, 6.6 Hz, 1H), 4.24 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.45 (td, *J* = 6.0, 3.0 Hz, 1H), 5.19 (dd, *J* = 6.0, 3.6 Hz, 1H), 5.61 (dd, *J* = 6.0, 1.8 Hz, 1H), 6.36 (d, *J* = 1.8 Hz, 1H), 7.41 (t, *J* = 7.8 Hz, 1H), 7.47 (br s, 2H), 7.51 (t, *J* = 7.8 Hz, 2H), 7.56 (s, 2H), 7.59 (t, *J* = 7.8 Hz, 1H), 7.73 (ovlp d, *J* = 7.8 Hz, 2H), 7.75 (ovlp d, *J* = 7.8 Hz, 1H), 8.33 (ovlp s, 1H), 8.34 (ovlp d, *J* = 7.8 Hz, 1H), 8.65 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 25.1, 26.9, 68.1, 81.2, 83.4, 83.8, 88.9, 113.5, 118.3, 126.1, 126.71, 126.74, 127.6, 128.1, 129.0, 129.1, 140.23, 140.24, 140.3, 149.7, 156.0, 157.9 (missing 1 Ar C); HRMS (APCI+) calcd for $C_{25}H_{27}N_6O_6S$ $[M + H]^+$ 539.1707, found 539.1706 (error 0.2 ppm).

2-(Biphen-3-yl)-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (28). Compound **87** (87.4 mg, 0.162 mmol, 1.0 equiv) was coupled to compound **33** using the general procedure for salicylation. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) afforded 2-(biphen-3-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (102 mg, 78%): R_f = 0.46 (95:5 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 1.18 (t, *J* = 7.2 Hz, 9H), 1.39 (s, 3H), 1.63 (s, 3H), 3.05 (q, *J* = 7.2 Hz, 6H), 3.37 (s, 3H), 4.32−4.37 (m, 2H), 4.62−4.65 (m, 1H), 5.10 (s, 2H), 5.26 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.55 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.36 (d, *J* = 2.4 Hz, 1H), 6.91 (t, *J* = 7.8 Hz, 1H), 7.07 (d, *J* = 7.8 Hz, 1H), 7.23 (t, *J* = 7.8 Hz, 1H), 7.32 (ovlp d, *J* = 7.8 Hz, 1H), 7.34 (ovlp t, *J* = 7.8 Hz, 1H), 7.44 (t, *J* = 7.8 Hz, 2H), 7.51 (t, *J* = 7.8 Hz, 1H), 7.66 (ovlp d, *J* = 7.8 Hz, 1H), 7.69 (ovlp d, *J* = 7.8 Hz, 2H), 8.35 (d, *J* = 7.8 Hz, 1H), 8.41 (s, 1H), 8.67 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 9.1, 25.7, 27.6, 47.8, 56.5, 70.0, 83.5, 85.8, 86.0, 91.9, 96.3, 115.2, 116.9, 119.1, 122.5, 127.9, 128.0, 128.1, 128.5, 129.4, 129.6, 129.90, 129.92, 131.0, 132.4, 140.2, 141.7, 142.3, 142.4, 151.7, 155.6, 157.2, 160.7, 176.6; HRMS

(ESI−) calcd for $C_{34}H_{33}N_6O_9S$ [M − H]$^-$ 701.2035, found 701.2029 (error 0.9 ppm).

2-(Biphen-3-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine triethylammonium salt (86 mg, 0.107 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by conversion to the sodium salt using the general procedure for ion-exchange afforded the title compound (58 mg, 75%): R_f = 0.40 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 4.36 (d, *J* = 3.0 Hz, 1H), 4.40 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.45−4.50 (m, 2H), 4.81 (t, *J* = 5.4 Hz, 1H), 6.25 (d, *J* = 5.4 Hz, 1H), 6.75−6.79 (m, 2H), 7.28 (t, *J* = 7.8 Hz, 1H), 7.35 (t, *J* = 7.8 Hz, 1H), 7.47 (t, *J* = 7.8 Hz, 2H), 7.50−7.53 (m, 1H), 7.67 (d, *J* = 7.8 Hz, 1H), 7.70 (d, *J* = 7.8 Hz, 2H), 7.94 (d, *J* = 7.8 Hz, 1H), 8.38 (t, *J* = 7.8 Hz, 1H), 8.49−8.51 (m, 1H), 8.63 (s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 70.0, 72.6, 76.1, 84.5, 89.5, 118.0, 119.2, 449.4, 120.7, 128.0, 128.3, 128.5, 128.6, 129.6, 129.96, 130.03, 131.6, 134.5, 140.4, 141.5, 142.59, 142.61, 152.4, 157.3, 161.1, 162.2, 175.5; HRMS (ESI−) calcd for $C_{29}H_{25}N_6O_8S$ [M − H]$^-$ 617.1460, found 617.1456 (error 0.6 ppm).

2-(Biphen-4-yl)-2′,3′-*O*-isopropylideneadenosine (81). To a solution of compound **75** (274 mg, 0.63 mmol, 1.0 equiv) in 1,4-dioxane (10 mL) were added 4-biphenylboronic acid (251 mg, 1.27 mmol, 2.0 equiv), $Pd(OAc)_2$ (28 mg, 0.126 mmol, 0.2 equiv), 2-(biphenyl)dicyclohexylphosphine (66 mg, 0.19 mmol, 0.30 equiv), K_3PO_4 (334 mg, 1.58 mmol, 2.5 equiv), and the mixture was heated at 110 °C for 12 h. After cooling to room temperature, the mixture was diluted with EtOAc (50 mL), filtered, and concentrated. Purification by flash chromatography (gradient from 80−100% EtOAc/hexane) afforded the title compound (192 mg, 67%) as a white solid: R_f = 0.20 (80:20 EtOAc/hexane); ^{1}H NMR (600 MHz, DMSO-d_6) δ 1.37 (s, 3H), 1.59 (s, 3H), 3.53−3.57 (m, 1H), 3.59−3.62 (m, 1H), 4.21−4.24 (m, 1H), 5.03 (t, *J* = 5.4 Hz, 1H), 5.11 (dd, *J* = 6.0, 2.4 Hz, 1H), 5.52 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.26 (d, *J* = 2.4 Hz, 1H), 7.37−7.42 (m, 3H), 7.50 (t, *J* = 7.8 Hz, 2H), 7.75 (d, *J* = 7.8 Hz, 2H), 7.79 (d, *J* = 8.4 Hz, 2H), 8.37 (s, 1H), 8.44 (d, *J* = 8.4 Hz, 2H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 25.3, 27.1, 61.6, 81.5, 83.4, 86.8, 89.1, 113.1, 118.2, 126.5, 126.7, 127.7, 128.3, 129.0, 137.3, 139.6, 140.4, 141.2, 149.9, 155.9, 157.7; HRMS (APCI+) calcd for $C_{25}H_{26}N_5O_4$ [M + H]$^+$ 460.1979, found 460.2078 (error 21.5 ppm).

2-(Biphen-4-yl)-2′,3′-*O*-isopropylidene-5′-*O*-(sulfamoyl)adenosine (88). Sulfamoyl chloride (66 mg, 0.57 mmol, 4.0 equiv) was added to a solution of **81** (66 mg, 0.14 mmol, 1.0 equiv) in DMA (1.0 mL) at 0 °C. The solution was stirred for 4 h at 0 °C and then partitioned between EtOAc (25 mL) and H_2O (25 mL). The organic layer was separated, washed with H_2O (3 × 25 mL), saturated aqueous NaCl (25 mL), dried (Na_2SO_4), and concentrated to afford the title compound (75 mg, 97%) as a white solid: R_f = 0.30 (80:20 EtOAc/benzene); ^{1}H NMR (600 MHz, DMSO-d_6) δ 1.38 (s, 3H), 1.60 (s, 3H), 4.16 (dd, *J* = 10.8, 6.0 Hz, 1H), 4.28 (dd, *J* = 10.8, 5.4 Hz, 1H), 4.45 (td, *J* = 5.7, 3.6 Hz, 1H), 5.20 (dd, *J* = 6.0, 3.6 Hz, 1H), 5.56 (dd, *J* = 6.0, 2.4 Hz, 1H), 6.34 (d, *J* = 2.4 Hz, 1H), 7.40 (t, *J* = 7.8 Hz, 1H), 7.44 (br s, 2H), 7.50 (t, *J* = 7.8 Hz, 2H), 7.58 (s, 2H), 7.75 (d, *J* = 7.8 Hz, 2H), 7.80 (d, *J* = 7.8 Hz, 2H), 8.34 (s, 1H), 8.43 (d, *J* = 7.8 Hz, 2H); ^{13}C NMR (150 MHz, DMSO-d$_6$) δ 25.2, 27.0, 68.1, 81.1, 83.3, 83.6, 88.8, 113.7, 118.2, 126.56, 126.64, 127.7, 128.3, 129.0, 137.2, 139.6, 140.3, 141.2, 149.8, 155.9, 157.9; HRMS (APCI+) calcd for $C_{25}H_{27}N_6O_6S$ [M + H]$^+$ 539.1707, found 539.1711 (error 0.7 ppm).

2-(Biphen-4-yl)-5′-*O*-[*N*-(2-hydroxybenzoyl)sulfamoyl]adenosine Sodium Salt (29). Compound **88** (70 mg, 0.13 mmol, 1.0 equiv) was coupled to compound **33** using the general procedure for salicylation. The mixture was filtered, washing with DMF (10 mL), and the filtrate was concentrated to provide the crude product as a white solid. The crude product was dissolved in MeOH (10 mL) and then slowly concentrated 3-fold by blowing down with a stream of nitrogen gas to provide a white precipitate, which was collected to yield 2-(biphen-4-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine cesium salt (67 mg, 73%): R_f = 0.50 (95:5 EtOAc/MeOH); ^{1}H NMR (600 MHz, DMSO-d_6) δ 1.33 (s, 3H), 1.60 (s, 3H), 3.33 (s, 3H), 4.15 (d, *J* = 4.2 Hz, 2H), 4.49 (td, *J* = 4.2, 1.8 Hz, 1H), 5.07 (s, 2H), 5.20 (dd, *J* = 6.0, 1.8 Hz, 1H), 5.48 (dd, *J* = 6.0, 3.0 Hz, 1H), 6.29 (d, *J* = 3.0 Hz, 1H), 6.89 (t, *J* = 7.8 Hz, 1H), 6.99 (d, *J* = 7.8 Hz, 1H), 7.18 (t, *J* = 7.8 Hz, 1H), 7.32 (d, *J* = 7.8 Hz, 1H), 7.37−7.42 (m, 3H), 7.49 (t, *J* = 7.8 Hz, 2H), 7.73 (d, *J* = 7.8 Hz, 2H), 7.78 (d, *J* = 8.4 Hz, 2H), 8.45 (ovlp d, *J* = 8.4 Hz, 2H), 8.46 (ovlp s, 1H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 25.2, 27.1, 55.5, 67.2, 81.7, 83.4, 83.8, 89.0, 95.0, 113.2, 116.8, 118.0, 121.2, 126.5, 126.7, 127.7, 128.3, 128.7, 128.8, 129.0, 133.1, 137.3, 139.7, 140.1, 141.1, 150.1, 153.8, 156.0, 157.8, 171.6; HRMS (ESI−) calcd for $C_{34}H_{33}N_6O_9S$ [M − H]$^-$ 701.2035, found 701.1984 (error 7.3 ppm).

2-(Biphen-4-yl)-2′,3′-*O*-isopropylidene-5′-*O*-{*N*-[(2-methoxymethoxy)benzoyl]sulfamoyl}adenosine cesium salt (42 mg, 0.059 mmol, 1.0 equiv) prepared above was converted to the title compound using the general procedure for TFA deprotection. Purification by flash chromatography (90:10:1 EtOAc/MeOH/Et_3N) followed by conversion to the sodium salt using the general procedure for ion exchange afforded the title compound (36 mg, 80%): R_f = 0.40 (90:10 EtOAc/MeOH); ^{1}H NMR (600 MHz, CD_3OD) δ 4.35−4.37 (m, 1H), 4.41 (dd, *J* = 10.8, 3.0 Hz, 1H), 4.47−4.52 (m, 2H), 4.88 (ovlp t, *J* = 5.4 Hz, 1H), 6.22 (d, *J* = 5.4 Hz, 1H), 6.75 (t, *J* = 7.8 Hz, 1H), 6.78 (d, *J* = 7.8 Hz, 1H), 7.27 (t, *J* = 7.8 Hz, 1H), 7.36 (t, *J* = 7.8 Hz, 1H), 7.46 (t, *J* = 7.8 Hz, 2H), 7.68 (d, *J* = 7.8 Hz, 2H), 7.70 (d, *J* = 7.8 Hz, 2H), 7.94 (d, *J* = 7.8 Hz, 1H), 8.46 (ovlp d, *J* = 7.8 Hz, 2H), 8.47 (ovlp s, 1H); ^{13}C NMR (150 MHz, CD_3OD) δ 69.9, 72.7, 75.8, 84.5, 89.6, 118.0, 119.3, 119.4, 120.8, 127.9, 128.2, 128.7, 130.00, 130.03, 131.6, 134.5, 138.9, 141.7, 142.1, 143.9, 152.4, 157.3, 160.9, 162.2, 175.4; HRMS (ESI−) calcd for $C_{29}H_{25}N_6O_8S$ [M − H]$^-$ 617.1460, found 617.14541 (error 1.0 ppm).

Enzyme Kinetic Studies. ATP−PP_i Exchange Assay for MbtA, FadD17, FadD19, FadD26, and FadD28. MbtA was expressed in *E. coli* and purified as described.[21] FadD17, FadD19, FadD26, and FadD28 were cloned and expressed in *E. coli* and purified as described.[36] MbtA concentration was determined by active site titration with **6** as described.[21] The inhibition assays were performed in duplicate as described.[21] In brief, the reaction was initiated by adding 10 μL of [^{32}P]PP_i with 7 nM MbtA in 90 μL of reaction buffer (278 μM salicylic acid, 11.1 mM ATP, 1.11 mM PP_i, 83.3 mM Tris-HCl, pH 7.5, 11.1 mM $MgCl_2$, 2.22 mM DTT) at 37 °C in the presence of eight different concentrations of the inhibitor. The reaction was terminated by the addition of 200 μL of quenching buffer (350 mM $HClO_4$, 100 mM PP_i, 1.8% w/v activated charcoal). The charcoal was pelleted by centrifugation and washed once with 500 μL of H_2O and analyzed by liquid scintillation counting as described. The K_I^{app} values were calculated using either the Hill−Morrison (eq 1) or Morrison (eq 2) equations as described.[21]

$$v_i/v_0 = \frac{1}{1 + ([I]/K_I^{app})^h} \quad (1)$$

$$v_i/v_0 = \frac{([E] - [I] - K_I^{app}) + \sqrt{([E] - [I] - K_I^{app})^2 + 4[E][K_I^{app}]}}{2[E]} \quad (2)$$

For assays involving FadD enzymes, the assay buffer and incubation times were identical, but 0.30−3.0 μM corresponding FadD protein was employed.

Isothermal Calorimetry. ITC displacement titration experiments were performed with a Microcal VP-ITC titration microcalorimeter (Microcal, Inc.) using salicylic acid as the weak ligand (K_D = 1.63 ± 0.52 μM [51]).[100] MbtA was dialyzed (2 × 1 L) against 30 mM Tris-HCl, pH 8.0, 1 mM $MgCl_2$, and 1 mM TCEP for 24 h prior to titrations. MbtA concentration was determined by active site titration as described[21] and diluted to 4.0 μM with dialysate buffer and 50−200 mM salicylic acid immediately prior to the experiment. A solution of the ligand **26** was prepared in dialysate buffer containing 50−200 mM salicylic acid. Protein and ligand solutions were degassed by vacuum aspiration (5−10 min) at room temperature

prior to loading the samples in the ITC cell and syringe. All titrations were carried out at 25 °C with a stirring speed of 264 rpm and a 900 s duration between 10 μL injections. The initial injection was not used for data fitting. Titrations were run past the point of enzyme saturation to determine the heat of dilution. The heats of dilution were negligible in all cases and were subtracted from the respective titrations prior to data analysis. Thermodynamic parameters N (stoichiometry), apparent K_A^{app} (association constant), and apparent ΔH^{app} (enthalpy change) were obtained by nonlinear least-squares fitting of the experimental data using a single-site-binding model of the Origin software package (version 5.0) provided with the instrument. The true K_A and ΔH values were obtained from the apparent values using eqs 3 and 4, where [Sal] represents the concentration of salicylic acid and $\Delta H_{(Sal)}$ and $K_{A(Sal)}$ correspond to the enthalpy and association constant of salicylic acid with MbtA ($K_{A(Sal)} = 6.64 \times 10^5$ M^{-1}, $\Delta H_{(Sal)} = -6.77$ kcal/mol).[51]

$$K_A^{app} = \frac{K_A}{1 + [\mathrm{Sal}]K_{A(\mathrm{Sal})}} \tag{3}$$

$$\Delta H = \frac{\Delta H^{app} + \Delta H_{(\mathrm{Sal})}[\mathrm{Sal}]K_{A(\mathrm{Sal})}}{[1 + [\mathrm{Sal}]K_{A(\mathrm{Sal})}]} \tag{4}$$

The free energy of binding (ΔG) and entropy change were obtained from the Gibbs free energy equation (eq 5).

$$\Delta G = -RT\ln K_A = \Delta H - T\Delta S \tag{5}$$

The affinity of the ligand to protein is given as the dissociation constant ($K_D = 1/K_A$). Three independent experiments were performed and analyzed independently, and the thermodynamic values obtained were averaged.

***M. tuberculosis* H37Rv MIC Assay.** Minimum inhibitory concentrations (MICs) were experimentally determined as previously described.[14] MICs were determined in quadruplicate in iron-deficient GAST and GAST supplemented with 200 μM $FeCl_3$ according to the broth microdilution method using compounds from DMSO stock solutions or with control wells treated with an equivalent amount of DMSO.[10] Isoniazid was used as a positive control, while DMSO was employed as a negative control. All measurements reported herein used an initial cell density of 10^4-10^5 cells/assay, and growth was monitored at 10−14 days, with the untreated and DMSO-treated control cultures reaching an OD_{620} of 0.2−0.3. Plates were incubated at 37 °C (100 μL/well), and growth was recorded by measurement of optical density at 620 nm.

Cell Cytotoxicity-MTT Assay. African green monkey *Cercopithecus aethiops* kidney cells (Vero, ATCC) cells were plated in 96-well plates at (2.5−5.0) $\times 10^4$ cells per well (200 μL). Vero cells were maintained in minimum essential medium (MEM) supplemented with 5% fetal bovine serum (FBS), 100 U/mL penicillin, and 100 μg/mL streptomycin. Compounds were prepared as 20 mM stock solutions in DMSO, and 1 μL of the compound stock solution was added to each well in 200 μL of MEM, yielding a final compound concentration of 100 μM. Control wells contained either 1% DMSO (negative control) or 100 μM 5′-*O*-(sulfamoyl)adenosine (positive control), and all reactions were done in triplicate. The plate was incubated for 48 h at 37 °C in a 5% CO_2/95% air humidified atmosphere. Measurement of cell viability was carried out using a modified method of Mosmann based on 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT).[101] MTT was prepared fresh at 1 mg/mL in serum-free, phenol red-free RPMI 1640 media. MTT solution (200 μL) was added to each well, and the plate was incubated as described above for 3 h. The MTT solution was removed, and the formazan crystals were solubilized with 200 μL of isopropanol. The plate was read on a M5e spectrophotometer (Molecular Devices) at 570 nm for formazan and 650 nm for background subtraction. Cell viability was estimated as the percentage absorbance of sample relative to the DMSO control.

In Vitro Translation Assay. In vitro translation of luciferase was performed using a T7 coupled rabbit reticulocyte system (Promega catalog no. L4610), according to the manufacturers instructions. 5′-*O*-(Sulfamoyl)adenosine (AMS, a known potent inhibitor of protein synthesis[37]) was used as a positive control.

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Supporting Information Available: Table of HPLC purities of **7**−**29**, ^{1}H and ^{13}C NMR data of all final targets **7**−**29** and intermediates **35**−**44**, molecular dynamics simulations, QM/MM studies, and complete author list for refs 58 and 88. This material is available free of charge via the Internet at http://pubs.acs.org.

References

(1) World Health Organization. Fact sheet on tuberculosis. http://www.who.int/mediacentre/factsheets/fs104/en/. **2005**
(2) CDC treatment of tuberculosis. *MMWR* **2003**, *52*, 1−77.
(3) CDC Emergence of *Mycobacterium tuberculosis* with extensive resistance to second-line drugs worldwide, 2000−2004. *MMWR* **2006**, *55*, 301−305.
(4) Miethke, M.; Marahiel, M. A. Siderophore-based iron acquisition and pathogen control. *Microbiol. Mol. Biol. Rev.* **2007**, *71*, 413–451.
(5) Kohanski, M. A.; Dwyer, D. J.; Hayete, B.; Lawrence, C. A.; Collins, J. J. A common mechanism of cellular death induced by bactericidal antibiotics. *Cell* **2007**, *130*, 797–810.
(6) Ojha, A.; Hatfull, G. F. The role of iron in *Mycobacterium smegmatis* biofilm formation: the exochelin siderophore is essential in limiting iron conditions for biofilm formation but not for planktonic growth. *Mol. Microbiol.* **2007**, *66*, 468–483.
(7) Snow, G. A. Mycobactins−iron-chelating growth factors from mycobacteria. *Bacteriol. Rev.* **1970**, *34*, 99–125.
(8) Vergne, A. F.; Walz, A. J.; Miller, M. J. Iron chelators from mycobacteria (1954−1999) and potential therapeutic applications. *Nat. Prod. Rep.* **2000**, *17*, 99–116.
(9) Barry, C. E., 3rd. Unpublished results.
(10) De Voss, J. J.; Rutter, K.; Schroeder, B. G.; Su, H.; Zhu, Y. Q.; Barry, C. E. The salicylate-derived mycobactin siderophores of *Mycobacterium tuberculosis* are essential for growth in macrophages. *Proc. Natl. Acad. Sci. U.S.A.* **2000**, *97*, 1252–1257.
(11) Quadri, L. E. N.; Sello, J.; Keating, T. A.; Weinreb, P. H.; Walsh, C. T. Identification of a *Mycobacterium tuberculosis* gene cluster encoding the biosynthetic enzymes for assembly of the virulence-conferring siderophore mycobactin. *Chem. Biol.* **1998**, *5*, 631–645.
(12) Ferreras, J. A.; Ryu, J. S.; Di.Lello, F.; Tan, D. S.; Quadri, L. E. N. Small-molecule inhibition of siderophore biosynthesis in *Mycobacterium tuberculosis* and *Yersinia pestis*. *Nat. Chem. Biol.* **2005**, *1*, 29–32.
(13) Miethke, M.; Bisseret, P.; Beckering, C. L.; Vignard, D.; Eustache, J.; Marahiel, M. A. Inhibition of aryl acid adenylation domains involved in bacterial siderophore synthesis. *FEBS J.* **2006**, *273*, 409–419.
(14) Somu, R. V.; Boshoff, H.; Qiao, C. H.; Bennett, E. M.; Barry, C. E.; Aldrich, C. C. Rationally designed nucleoside antibiotics that inhibit siderophore biosynthesis of *Mycobacterium tuberculosis*. *J. Med. Chem.* **2006**, *49*, 31–34.
(15) May, J. J.; Kessler, N.; Marahiel, M. A.; Stubbs, M. T. Crystal structure of DhbE, an archetype for aryl acid activating domains of modular nonribosomal peptide synthetases. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 12120–12125.
(16) Cisar, J. S.; Ferreras, J. A.; Soni, R. K.; Quadri, L. E. N.; Tan, D. S. Exploiting ligand conformation in selective inhibition of non-ribosomal peptide synthetase amino acid adenylation with designed macrocyclic small molecules. *J. Am. Chem. Soc.* **2007**, *129*, 7752–7753.
(17) Anzai, K.; Nakamura, G.; Suzuki, S. A new antibiotic, tubercidin. *J. Antibiot. (Tokyo)* **1957**, *10*, 201–204.
(18) Shuman, D. A.; Robins, M. J.; Robins, R. K. The synthesis of nucleoside sulfamates related to nucleocidin. *J. Am. Chem. Soc.* **1970**, *92*, 3434–3440.
(19) Catro-Pichel, J.; Garcia-Lopez, M. T.; De las Heras, F. G. A facile synthesis of ascamycin and related analogs. *Tetrahedron* **1987**, *43*, 383–389.
(20) Preobrazhenskaya, M. N.; Korbukh, I. A. The Synthesis and Reactions of Pyrrole, Pyrazole, Triazole, Indole, Indazole, and Benzotriazole

Nucleosides and Nucleotides. In *Chemistry of Nucleosides and Nucleotides*; Plenum Press: New York, 1994; Vol. 3; p 1−47.
(21) Somu, R. V.; Wilson, D. J.; Bennett, E. M.; Boshoff, H. I.; Celia, L.; Beck, B. J.; Barry, C. E.; Aldrich, C. C. Antitubercular nucleosides that inhibit siderophore biosynthesis: SAR of the glycosyl domain. *J. Med. Chem.* **2006**, *49*, 7623–7635.
(22) Jung, M. E.; Rhee, H. π-Allylpalladium formation from allylic amines via *N*,*N*-ditosylimides and *N*-tosylamides. Efficient synthesis of the antiviral agent carbovir. *J. Org. Chem.* **1994**, *59*, 4719–4720.
(23) Rouhi, A. M. Simplifying syntheses is always a key goal. *Chem. Eng. News* **2003**, *81*, 40.
(24) Daluge, S.; Vince, R. A synthetic route to carbocyclic aminonucleosides. *Tetrahedron Lett.* **1976**, *17*, 3005–3008.
(25) Vince, R.; Daluge, S. Facile synthesis of carbocyclic lyxo- and ribonucleosides. *J. Org. Chem.* **1980**, *45*, 531–533.
(26) Okada, M.; Iwashita, S.; Koizumi, N. Efficient general method for sulfamoylation of a hydroxyl group. *Tetrahedron Lett.* **2000**, *41*, 7047–7051.
(27) Mazaleyrat, J.-P.; Xie, J.; Wakselman, M. Selective hydrogenolysis of the benzyloxycarbonyl protecting group of N^{ϵ}-lysine in cyclopeptides containing benzylic phenyl ether function. Evidence for N^{ϵ}-methylated lysine side products. *Tetrahedron* **1992**, *33*, 4301–4302.
(28) Zhao, H.; Pagano, A. R.; Wang, W. M.; Shallop, A.; Gaffney, B. L.; Jones, R. A. Use of a ^{13}C atom to differentiate two ^{15}N-labeled nucleosides. Syntheses of [$^{15}NH_2$]-adenosine, [1, NH_2-$^{15}N_2$]- and [2-^{13}C-1, NH_2-$^{15}N_2$]-guanosine, and [1,7, NH_2-$^{15}N_3$]- and [2-^{13}C-1,7, NH_2-$^{15}N_3$]-2′-deoxyguanosine. *J. Org. Chem.* **1997**, *62*, 7832–7835.
(29) Fleysher, M. H. N^6-Substituted adenosines: Synthesis, biological activity, and some structure−activity relationships. *J. Med. Chem.* **1972**, *15*, 187–191.
(30) Holmes, R. E.; Robins, R. K. Purine nucleosides. IX. The synthesis of 9-β-D-ribofuranosyl uric acid and other related 8-substituted purine ribonucleosides. *J. Am. Chem. Soc.* **1965**, *87*, 1772–1776.
(31) Matsuda, A.; Shinozaki, M.; Yamaguchi, T.; Homma, H.; Nomoto, R.; Miyasaka, T.; Watanabe, Y.; Abiru, T. Nucleosides and nucleotides. 103. 2-Alkynyladenosines. A novel class of selective adenosine-A2 receptor agonists with potent antihypertensive effects. *J. Med. Chem.* **1992**, *35*, 241–252.
(32) Chen, L. Q.; Gao, G. Y.; Felczak, K.; Bonnac, L.; Patterson, S. E.; Wilson, D.; Bennett, E. M.; Jayaram, H. N.; Hedstrom, L.; Pankiewicz, K. W. Probing binding requirements of type I and type II isoforms of inosine monophosphate dehydrogenase with adenine-modified nicotinamide adenine dinucleotide analogues. *J. Med. Chem.* **2007**, *50*, 5743–5751.
(33) Lakshman, M. K.; Hilmer, J. H.; Martin, J. Q.; Keeler, J. C.; Dinh, Y. Q.; Ngassa, F. N.; Russon, L. M. Palladium catalysis for the synthesis of hydrophobic C-6 and C-2 aryl 2′-deoxynucleosides. Comparison of C−C versus C−N bond formation as well as C-6 versus C-2 reactivity. *J. Am. Chem. Soc.* **2001**, *123*, 7779–7787.
(34) Stolarski, R.; Dudycz, L.; Shugar, D. NMR studies in the syn−anti dynamic equilibrium in purine nucleosides and nucleotides. *Eur. J. Biochem.* **1980**, *108*, 111–121.
(35) Yu, M.; Magalhaes, M. L. B.; Cook, P. F.; Blanchard, J. S. Bisubstrate inhibition: theory and application to *N*-acetyltransferases. *Biochemistry* **2006**, *45*, 14788–14794.
(36) Trivedi, O. A.; Arora, P.; Sridharan, V.; Tickoo, R.; Mohanty, D.; Gokhale, R. S. Enzymatic activation and transfer of fatty acids as acyl-adenylates in mycobacteria. *Nature* **2004**, *428*, 441–445.
(37) Bloch, A.; Coutsogeorgopoulos, C. Inhibition of protein synthesis by 5′-sulfamoyladenosine. *Biochemistry* **1971**, *10*, 4395–4398.
(38) Xu, Y.; Miller, M. J. Total synthesis of mycobactin analogues as potent antimycobacterial agents using a minimal protecting group strategy. *J. Org. Chem.* **1998**, *63*, 4314–4322.
(39) Stirrett, K. L.; Ferreras, J. A.; Sinha, B. N.; Venkatesan, J.; Ren, T.; Quadri, L. E. N. Small-molecules with structural similarities to siderophores as novel antimicrobials against *Mycobacterium tuberculosis* and *Yersinia pestis*. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 2662–2668.
(40) Miller, M. J.; Malouin, F. Microbial iron chelators as drug delivery agents: the rational design and synthesis of siderophore−drug conjugates. *Acc. Chem. Res.* **1993**, *26*, 241–249.
(41) Bisseret, P.; Thielges, S.; Stephane, B.; Miethke, M.; Marahiel, M. A.; Eustache, J. Synthesis of a 2-indolylphosphonamide derivative with inhibitory activity against yersiniabactin biosynthesis. *Tetrahedron Lett.* **2007**, *48*, 6080–6083.
(42) Qiao, C. H.; Gupte, A.; Boshoff, H. I.; Wilson, D. J.; Bennett, E. M.; Somu, R. V.; Barry, C. E.; Aldrich, C. C. 5′-*O*-[(*N*-Acyl)sulfamoyl]adenosines as antitubercular agents that inhibit MbtA: an adenylation enzyme required for siderophore biosynthesis of the mycobactins. *J. Med. Chem.* **2007**, *50*, 6080–6094.
(43) Qiao, C. H.; Wilson, D. J.; Bennett, E. M.; Aldrich, C. C. A mechanism-based aryl carrier protein/thiolation domain affinity probe. *J. Am. Chem. Soc.* **2007**, *129*, 6350–6351.
(44) Vannada, J.; Bennett, E. M.; Wilson, D. J.; Boshoff, H. I.; Barry, C. E.; Aldrich, C. C. Design, synthesis, and biological evaluation of beta-ketosulfonamide adenylation inhibitors as potential antitubercular agents. *Org. Lett.* **2006**, *8*, 4707–4710.
(45) Pavela-Vrancic, M.; Van Liempt, H.; Pfeifer, E.; Freist, W.; von Döhren, H. Nucleotide binding by multienzyme peptide synthetases. *Eur. J. Biochem.* **1994**, *220*, 535–542.
(46) Braibant, M.; Gilot, P.; Content, J. The ATP binding cassette (ABC) transport systems of *Mycobacterium tuberculosis*. *FEMS Microbiol. Rev.* **2000**, *24*, 449–467.
(47) Niederweis, M. Mycobacterial porins, new channel proteins in unique outer membranes. *Mol. Microbiol.* **2003**, *49*, 1167–1177.
(48) Rodriguez, G. M.; Smith, I. Identification of an ABC transporter required for iron acquisition and virulence in *Mycobacterium tuberculosis*. *J. Bacteriol.* **2006**, *188*, 424–430.
(49) Broom, A. D. Rational design of enzyme inhibitors: multisubstrate analogue inhibitors. *J. Med. Chem.* **1989**, *32*, 2–7.
(50) Jencks, W. P. On the attribution and additivity of binding energies. *Proc. Natl. Acad. Sci. U.S.A.* **1981**, *78*, 4046–4050.
(51) Neres, J.; Wilson, D. Unpublished results.
(52) Callahan, B. P.; Lomino, J. V.; Wolfenden, R. Nanomolar inhibition of the enterobactin biosynthesis enzyme, EntE: synthesis, substituent effects, and additivity. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 3802–3805.
(53) Lai, J. R.; Koglin, A.; Walsh, C. T. Carrier protein structure and recognition in polyketide and nonribosomal peptide biosynthesis. *Biochemistry* **2006**, *45*, 14869–14879.
(54) Mercer, A. C.; Burkart, M. D. The ubiquitous carrier protein—a window to metabolite biosynthesis. *Nat. Prod. Rep.* **2007**, *24*, 750–773.
(55) Sieber, S. A.; Marahiel, M. A. Molecular mechanisms underlying nonribosomal peptide synthesis: approaches to new antibiotics. *Chem. Rev.* **2005**, *105*, 715–738.
(56) Argyrou, A.; Jin, L.; Siconilfi-Baez, L.; Angeletti, R. H.; Blanchard, J. S. Proteome-wide profiling of isoniazid targets in *Mycobacterium tuberculosis*. *Biochemistry* **2006**, *45*, 13947–13953.
(57) Janin, Y. L. Antituberculosis drugs: ten years of research. *Bioorg. Med. Chem.* **2007**, *15*, 2479–2513.
(58) Cole, S. T.; Brosch, R.; Parkhill, J.; Garnier, T.; Churcher, C.; Harris, D.; Gordon, S. V.; Eiglmeier, K.; Gas, S.; Barry, C. E., 3rd; et al. Deciphering the biology of *Mycobacterium tuberculosis* from the complete genome sequence. *Nature* **1998**, *393*, 537–544.
(59) Sareen, D.; Steffek, M.; Newton, G. L.; Fahey, R. C. ATP-dependent L-cysteine:1D-myo-inosityl 2-amino-2-deoxy-alpha-D-glucopyranoside ligase, mycothiol biosynthesis enzyme MshC, is related to class I cysteinyl-tRNA synthetases. *Biochemistry* **2002**, *41*, 6885–6890.
(60) Zheng, R.; Blanchard, J. S. Steady-state and pre-steady-state kinetic analysis of *Mycobacterium tuberculosis* pantothenate synthetase. *Biochemistry* **2001**, *40*, 12904–12912.
(61) Srivastava, S. K.; Tripathi, R. P.; Ramachandran, R. NAD+-dependent DNA ligase (Rv3014c) from *Mycobacterium tuberculosis*. Crystal structure of the adenylation domain and identification of novel inhibitors. *J. Biol. Chem.* **2005**, *280*, 30273–30281.
(62) Hotter, G. S.; Wards, B. J.; Mouat, P.; Besra, G. S.; Gomes, J.; Singh, M.; Bassett, S.; Kawakami, P.; Wheeler, P. R.; de Lisle, G. W.; Collins, D. M. Transposon mutagenesis of Mb0100 at the ppe1-nrp locus in *Mycobacterium bovis* disrupts phthiocerol dimycocerosate (PDIM) and glycosylphenol-PDIM biosynthesis, producing an avirulent strain with vaccine properties at least equal to those of *M. bovis* BCG. *J. Bacteriol.* **2005**, *187*, 2267–2277.
(63) Trivedi, O. A.; Arora, P.; Vats, A.; Ansari, M. Z.; Tickoo, R.; Sridharan, V.; Mohanty, D.; Gokhale, R. S. Dissecting the mechanism and assembly of a complex virulence mycobacterial lipid. *Mol. Cell* **2005**, *17*, 631–643.
(64) Gokhale, R. S.; Saxena, P.; Chopra, T.; Mohanty, D. Versatile polyketide enzymatic machinery for the biosynthesis of complex mycobacterial lipids. *Nat. Prod. Rep.* **2007**, *24*, 267–277.
(65) Onwueme, K. C.; Vos, C. J.; Zurita, J.; Ferreras, J. A.; Quadri, L. E. The dimycocerosate ester polyketide virulence factors of mycobacteria. *Prog. Lipid Res.* **2005**, *44*, 259–302.
(66) Pandey, A. K.; Sassetti, C. M. Mycobacterial persistence requires the utilization of host cholesterol. *Proc. Natl. Acad. Sci. U.S.A.* **2008**, *105*, 4376–4380.
(67) Ratledge, C.; Dover, L. G. Iron metabolism in pathogenic bacteria. *Annu. Rev. Microbiol.* **2000**, *54*, 881–941.
(68) Bearden, S. W.; Fetherston, J. D.; Perry, R. D. Genetic organization of the yersiniabactin biosynthetic region and construction of avirulent mutants in *Yersinia pestis*. *Infect. Immun.* **1997**, *65*, 1659–1668.
(69) Lawlor, M. S.; O'Connor, C.; Miller, V. L. Yersiniabactin is a virulence factor for *Klebsiella pneumoniae* during pulmonary infection. *Infect. Immun.* **2007**, *75*, 1463–1472.

(70) Cendrowski, S.; MacArthur, W.; Hanna, P. *Bacillus anthracis* requires siderophore biosynthesis for growth in macrophages and mouse virulence. *Mol. Microbiol.* **2004**, *51*, 407–417.
(71) Pfleger, B. F.; Lee, J. Y.; Somu, R. V.; Aldrich, C. C.; Hanna, P. C.; Sherman, D. H. Characterization and analysis of early enzymes for petrobactin biosynthesis in *Bacillus anthracis*. *Biochemistry* **2007**, *46*, 4147–4157.
(72) Raymond, K. N.; Dertz, E. A.; Kim, S. S. Enterobactin: an archetype for microbial iron transport. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 3584–3588.
(73) Crouch, M. L.; Castor, M.; Karlinsey, J. E.; Kalhorn, T.; Fang, F. C. Biosynthesis and IroC-dependent export of the siderophore salmochelin are essential for virulence of *Salmonella enterica* serovar Typhimurium. *Mol. Microbiol.* **2008**, *67*, 971–983.
(74) Griffiths, G. L.; Sigel, S. P.; Payne, S. M.; Neilands, J. B. Vibriobactin, a siderophore from *Vibrio cholerae*. *J. Biol. Chem.* **1984**, *259*, 383–385.
(75) Litwin, C. M.; Rayback, T. W.; Skinner, J. Role of catechol siderophore synthesis in *Vibrio vulnificus* virulence. *Infect. Immun.* **1996**, *64*, 2834–2838.
(76) Cox, C. D.; Rinehart, K. L., Jr.; Moore, M. L.; Cook, J. C., Jr. Pyochelin: novel structure of an iron-chelating growth promoter for *Pseudomonas aeruginosa*. *Proc. Natl. Acad. Sci. U.S.A.* **1981**, *78*, 4256–4260.
(77) Mihara, K.; Tanabe, T.; Yamakawa, Y.; Funahashi, T.; Nakao, H.; Narimatsu, S.; Yamamoto, S. Identification and transcriptional organization of a gene cluster involved in biosynthesis and transport of acinetobactin, a siderophore produced by *Acinetobacter baumannii* ATCC 19606(T). *Microbiology* **2004**, *150*, 2587–2597.
(78) Wyckoff, E. E.; Mey, A. R.; Payne, S. M. Iron acquisition in *Vibrio cholerae*. *BioMetals* **2007**, *20*, 405–416.
(79) Takase, H.; Nitanai, H.; Hoshino, K.; Otani, T. Impact of siderophore production on *Pseudomonas aeruginosa* infections in immunosuppressed mice. *Infect. Immun.* **2000**, *68*, 1834–1839.
(80) Gupte, A.; Subramanian, M.; Remmel, R. P.; Aldrich, C. C. Synthesis of deuterium-labelled 5′-*O*-[*N*-(salicyl)sulfamoyl]adenosine (Sal-AMS-d_4) as an internal standard for quantitation of Sal-AMS. *J. Labelled Compd. Radiopharm.* **2008**, *51*, 118–122.
(81) Thompson, J. D.; Higgins, D. G.; Gibson, T. J. Clustal-W. Improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* **1994**, *22*, 4673–4680.
(82) Sali, A.; Blundell, T. L. Comparative protein modeling by satisfaction of spatial restraints. *J. Mol. Biol.* **1993**, *234*, 779–815.
(83) *Schrodinger*; Schrodinger LLC: New York, 2005.
(84) Halgren, T. A. Merck molecular force field. II. MMFF94 van der.Waals and electrostatic parameters for intermolecular interactions. *J. Comput. Chem.* **1996**, *17*, 520–552.
(85) Still, W. C.; Tempczyk, A.; Hawley, R. C.; Hendrickson, T. Semianalytical treatment of solvation for molecular mechanics and dynamics. *J. Am. Chem. Soc.* **1990**, *112*, 6127–6129.
(86) Humphrey, W.; Dalke, A.; Schulten, K. VMD: visual molecular dynamics. *J. Mol. Graphics* **1996**, *14*, 33–38.
(87) Phillips, J. C.; Braun, R.; Wang, W.; Gumbart, J.; Tajkhorshid, E.; Villa, E.; Chipot, C.; Skeel, R. D.; Kale, L.; Schulten, K. Scalable molecular dynamics with NAMD. *J. Comput. Chem.* **2005**, *26*, 1781–1802.
(88) MacKerell, A. D.; Bashford, D.; Bellott, M.; Dunbrack, R. L.; Evanseck, J. D.; Field, M. J.; Fischer, S.; Gao, J.; Guo, H.; Ha, S.; et al. All-atom empirical potential for molecular modeling and dynamics studies of proteins. *J. Phys. Chem. B* **1998**, *102*, 3586–3616.
(89) MacKerell, A. D.; Feig, M.; Brooks, C. L. Improved treatment of the protein backbone in empirical force fields. *J. Am. Chem. Soc.* **2004**, *126*, 698–699.
(90) *Schrodinger*; Schrodinger LLC: New York, 2007.
(91) Becke, A. D. Density-functional thermochemistry. III. The role of exact exchange. *J. Chem. Phys.* **1993**, *98*, 5648–5652.
(92) Lee, C. T.; Yang, W. T.; Parr, R. G. Development of the Colle−Salvetti correlation-energy formula into a functional of the electron-density. *Phys. Rev. B* **1988**, *37*, 785–789.
(93) Stephens, P. J.; Devlin, F. J.; Chabalowski, C. F.; Frisch, M. J. Ab-initio calculation of vibrational absorption and circular-dichroism spectra using density-functional force-fields. *J. Phys. Chem.* **1994**, *98*, 11623–11627.
(94) Binkley, J. S.; Pople, J. A. Self-consistent molecular-orbital methods. 19. Split-valence Gaussian-type basis sets for beryllium. *J. Chem. Phys.* **1977**, *66*, 879–880.
(95) Dichfield, R.; Hehre, W. J.; Pople, J. A. Self-consistent molecular-orbital methods. IX. An extended Gaussian-type basis for molecular-orbital studies of organic molecules. *J. Chem. Phys.* **1971**, *54*, 724–728.
(96) Kaminski, G. A.; Friesner, R. A.; Tirado-Rives, J.; Jorgensen, W. L. Evaluation and reparametrization of the OPLS-AA force field for proteins via comparison with accurate quantum chemical calculations on peptides. *J. Phys. Chem. B* **2001**, *105*, 6474–6487.
(97) Heacock, D.; Forsyth, C. J.; Shiba, K.; Musier-Forsyth, K. Synthesis and aminoacyl-tRNA synthetase inhibitory activity of prolyl adenylate analogs. *Bioorg. Chem.* **1996**, *24*, 273–289.
(98) Homma, H.; Watanabe, Y.; Abiru, T.; Murayama, T.; Nomura, Y.; Matsuda, A. Nucleosides and nucleotides. 112. 2-(1-Hexyn-1-yl)adenosine-5′-uronamides. A new entry of selective A2 adenosine receptor agonists with potent antihypertensive activity. *J. Med. Chem.* **1992**, *35*, 2881–2890.
(99) Robins, M. J.; Uznanski, B. Nucleic-acid related compounds. 33. Conversions of adenosine and guanosine to 2,6-dichloro, 2-amino-6-chloro, and derived purine nucleosides. *Can. J. Chem.* **1981**, *59*, 2601–2607.
(100) Velazquez-Campoy, A.; Freire, E. Isothermal titration calorimetry to determine association constants for high-affinity ligands. *Nat. Protoc.* **2006**, *1*, 186–191.
(101) Mosmann, T. Rapid colorimetric assay for cellular growth and survival. Application to proliferation and cytotoxicity assays. *J. Immunol. Methods* **1983**, *65*, 55–63.

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