Aminoacyl-tRNA Synthetase Inhibitors as Potent and Synergistic Immunosuppressants

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Received January 28, 2008

The aminoacyl-tRNA synthetase family of enzymes is the target of many antibacterials and inhibitors of eukaryotic hyperproliferation. In screening analogues of 5′-*O*-(*N*-L-aminoacyl)-sulfamoyladenosine containing all 20 proteinogenic amino acids, we found these compounds to have potent immunosuppressive activity. Also, we found that combinations of these compounds inhibited the immune response synergistically. Based on these data, analogues with modifications at the aminoacyl and ribose moieties were designed and evaluated, and several of these showed high immunosuppressive potency, with one compound having an IC_{50} of 80 nM, when tested in a cellular mixed lymphocyte reaction assay. Apart from showing the potential of aminoacyl-tRNA synthetase inhibitors as immunosuppressants, the current study also provides arguments for careful evaluation of the immunosuppressive activity of developmental antibacterials that target these enzymes.

Introduction

Aminoacyl-tRNA synthetases (aaRSs[a]) are the enzymes that catalyze the transfer of amino acids to their cognate tRNA. They play a pivotal role in protein biosynthesis and are necessary for growth and survival of all cells. Consequently, inhibition of these enzymes is an attractive target for cell-growth inhibition, both as antibacterials and as inhibitors of cell (hyper)proliferation in humans.[1–7] Indeed, an inhibitor of IleRS, mupirocin (**1**), is in clinical use for the topical treatment of infections caused by Gram-positive bacteria and several more are in clinical development.[1]

The largest group of aaRS inhibitors (aaRSi) are the reaction-intermediate mimics. These compounds are analogues of aa-AMP (**2**), which is the natural reaction intermediate of these enzymes. Mupirocin belongs to this group, as do the 20 5′-*O*-(*N*-L-aminoacyl)-sulfamoyladenosines (aaSA, **3**, Figure 1). The latter are commercially available and show K_i values in the low nanomolar range against the corresponding isolated aaRSs.[8–12] Because the aaSA compounds differ only little from the natural reaction intermediates, they do not show noticeable selectivity toward either bacterial or eukaryotic aaRSs. This is a consequence of the use of the same general reaction intermediate by aaRSs of all kingdoms of life, despite extensive divergence in the structure and amino acid sequence of these enzymes. Nevertheless, researchers at Cubist Pharmaceuticals have been able to develop analogues of IleSA with extensive modifications at the nucleobase moiety that inhibit bacterial IleRS at nanomolar concentrations and that show good selectivity with respect to the human homologue. Also, they showed that one of the analogues, CB-432 (**4**) was active in a systemic mouse protection test with *Streptococcus pyogenes*, thereby validating the systemic potential of this class of compounds. Unfortunately, clinical development of this compound was halted as a consequence of low bioavailability, caused by high serum albumin binding (Figure 2).[4,13]

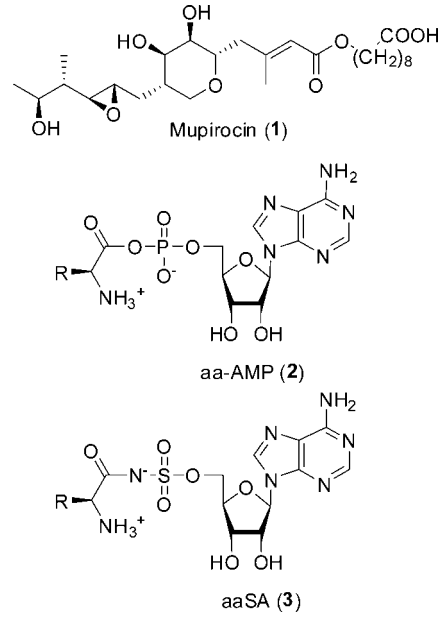

Figure 1. Structures of the IleRS inhibitor mupirocin (**1**), the universal aaRS reaction intermediate aa-Amp (**2**), and the universal aaRS inhibitor aaSA (**3**).

Resistance and resistance development are essential aspects of current antimicrobial development, and it was recently found that mupirocin and REP8839 (**5**), a selective inhibitor of bacterial MetRS, act synergistically in preventing resistance

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[a] Abbreviations: aa, amino acid; aaRS, aminoacyl-tRNA synthetases; aaRSi, aminoacyl-tRNA synthetase inhibitors; aaSA (where aa can be replaced by any amino acid 3-letter code), 5′-*O*-(*N*-aminoacyl)-sulfamoyladenosine; Boc, *tert*-butoxycarbonyl; DBU, 1,8-diazabicyclo [5.4.0]undec-7-ene; DMAc, *N*,*N*-dimethylacetamide; -OSu, *N*-hydroxysuccinimide ester; MLR, mixed lymphocytes reaction; rt, room temperature; TBDMS, *tert*-butyldimethylsilyl; TFA, trifluoroacetic acid; TIPDS, 1,1,3,3-tetraisopropyldisiloxane-1,3-diyl.

10.1021/jm8000746 CCC: $40.75
Published on Web 04/26/2008

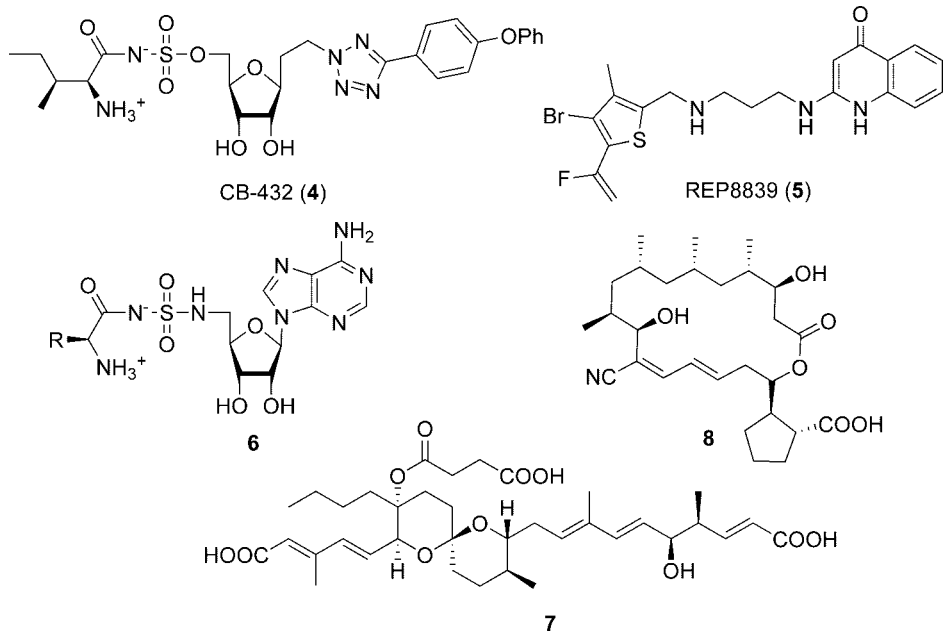


Figure 2. Structures of relevant aminoacyl-tRNA synthase inhibitors. Antibacterial IleRS inhibitor CB-432 (**4**), antibacterial MetRS inhibitor REP8839 (**5**), antipsoriatic 5′-deoxy-adenosine 5′-*N*-(*N*-L-aminoacyl)-sulfamide (**6**), antitumoral IleRS inhibitor reveromycin A (**7**), and borrelidin (**8**), which is an inhibitor of ThrRS and has antiangiogenetic properties.

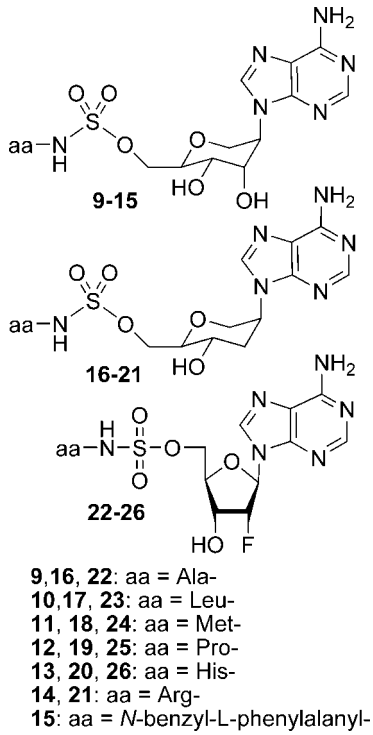


Figure 3. Structures of new analogues modified at the sugar moiety.

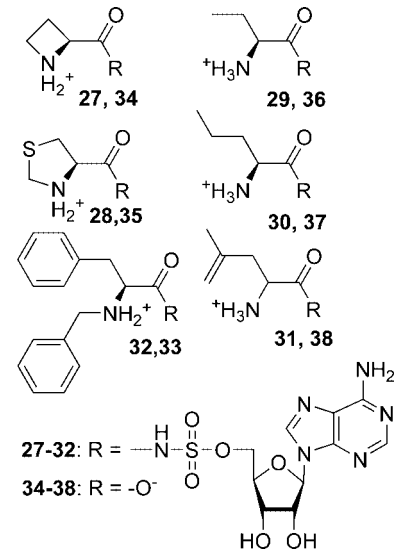
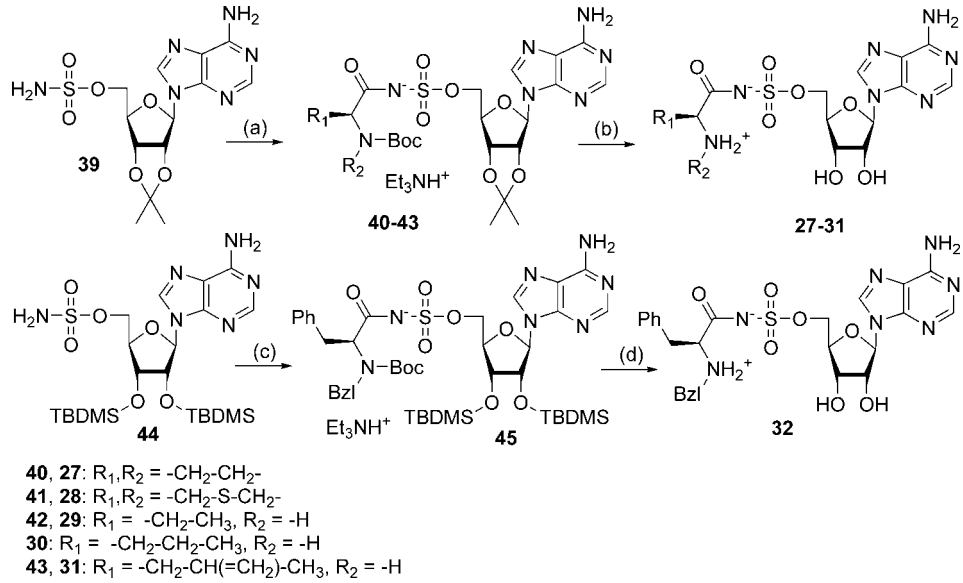


Figure 4. Structures of aminoacyl-modified analogues **27**−**32** and the amino acids these are derived from (**34**−**38**).

development, though neither synergism or subadditivity was found with respect to the MIC values. A fixed combination of mupirocin and REP8839 for the treatment of topical infections is currently in clinical development.[114] It was also found that mutations leading to resistance against REP8839 come with significant fitness costs. The latter makes antibiotic restriction or antibiotic rotation a feasible option to control antibiotic resistance against these agents.

The effects of aaRS inhibitors on human cells have been studied less extensively. Nevertheless, several aaRS inhibitors with potential application in humans have been described in recent years. For instance, Cubist Pharmaceuticals has patented several aaRS inhibitors for the treatment of psoriasis, a common hyperproliferative skin disease.[6] The compounds described in this patent are 19 aminoacyl analogues of 5′-deoxy-adenosine 5′-*N*-(*N*-L-aminoacyl)-sulfamide (**6**), which are structurally very similar to the aaSA compounds. Another well-characterized inhibitor of a human aaRS is reveromycin A (**7**). Reveromycin A, an inhibitor of human IleRS, was discovered in a screening effort to identify compounds that can cause morphology reversion of *src*^{ts}-NRK cells. It was also found to be an inhibitor of the epidermal growth factor-dependent responses of mouse epidermal cells and to have antitumoral effects against human ovarian carcinoma BG-1.[15] Finally, reveromycin A also induces apoptosis in bone osteoclasts at concentrations far lower than those needed for the inhibition of other cell lines. Woo et al. show that this selectivity is a consequence of the acid microenvironment of these cells, which greatly improves the uptake of

Scheme 1. Synthesis of aaSA Analogues of Figure 4[a]



40, **27**: R_1,R_2 = -CH_2-CH_2-
41, **28**: R_1,R_2 = -CH_2-S-CH_2-
42, **29**: R_1 = -CH_2-CH_3, R_2 = -H
30: R_1 = -CH_2-CH_2-CH_3, R_2 = -H
43, **31**: R_1 = -CH_2-CH(=CH_2)-CH_3, R_2 = -H

[a] Reagents and conditions: (a) *N*-Boc amino acid *N*-hydroxysuccinimide ester, DBU, DMF, 77−86%; (b) TFA/H_2O (5:2 v/v), 37−61%; (c) *N*-Boc,*N*-Bzl-L-phenylalanine *N*-hydroxysuccinimide ester, DBU, DMF, 52%; (d) (i) TFA/H_2O (5:2 v/v); (ii) $Et_3N\cdot 3HF$, THF, 43%.

reveromycin A.[16] A third well-characterized inhibitor of both bacterial and eukaryotic cell growth is the macrolide-polyketide Borrelidin (**8**).[17] This compound targets ThrRS and has anti-angiogenetic properties, though recent findings suggest that ThrRS inhibition is not the only mode of action of this compound.[18] In addition and in contrast to the other compounds mentioned here, borrelidin is a noncompetitive inhibitor with respect to both the amino acid and ATP.

Analogue Design and Synthesis

We wanted to investigate the potential use of aaRSi as immunosuppressants. Many antibacterial aaRS inhibitors have been characterized, both of natural and synthetic origin. In contrast, nothing is really known about the immunosuppressive activity of aaRS inhibitors, and it may be reasoned that inhibition of tRNA synthetases might lead to the inhibition of protein synthesis in mammalian cells and, thus, may have an immunosuppressive effect. Therefore, we first subjected all 20 analogues of **3** to an immunosuppressive screen. Based on these initial data, we then designed, synthesized, and evaluated analogues.

In **3**, four moieties can be distinguished, with respectively an aminoacyl, a linker, a sugar, and a nucleobase part. Analogues with modifications at all of these positions have already been made by other researchers, with most attention concentrated on the nucleobase and linker part. Therefore, we focused on sugar and aminoacyl modifications. For the sugar moiety, two six-membered sugar analogues were used, altritol (**9**−**15**) and anhydrohexitol (**16**−**21**). Both of these lack an anomeric carbon, making the compounds more stable toward enzymatic hydrolysis. We also prepared analogues containing 2′-deoxy-2′-fluoro-D-ribose (**22**−**26**) instead of the normal D-ribose sugar.

For the aminoacyl-modified analogues **27**−**32**, we replaced the amino acid moiety with nonproteinogenic amino acids that are either substrates of aaRS or inhibitors thereof. The phenylalanyl analogue *N*-benzyl-L-phenylalanine (**33**) is a substrate for PheRS.[19] Surprisingly, this is the case for both the L- and the D-enantiomers of this compound, although we only prepared analogues containing the L-enantiomer. Similarly, L-azetidine-2-carboxylic acid (**34**) and thioproline (L-4-thiazolidinecarboxylic acid, **35**) are substrates for ProRS and L-α-aminobutyric acid (**36**) and L-norvaline (**37**) are substrates for ValRS and MetRS.[20−22] L-Norvaline is a known substrate for LeuRS and IleRS as well.[2021] L-γ-Methallylglycine is a substrate of PheRS and LeuRS. Furthermore, L-azetidine-2-carboxylic acid, L-thioproline, and DL-γ-methallylglycine (**38**) are bactericidal against *Escherichia coli*.[23]

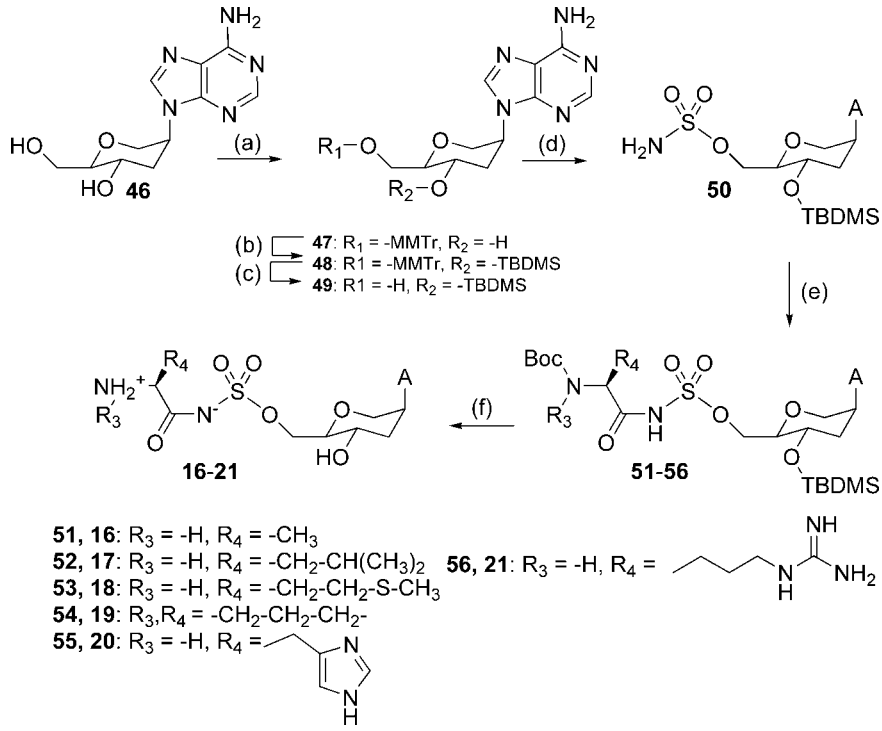
For the synthesis of all compounds, the general synthetic approach devised by Castro-Pichel et al. was used.[24]

In preparing aaSA analogues containing nonproteinogenic amino acids (Scheme 1), appropriately, 2′,3′-*O*-protected adenosine was sulfamoylated using fresh, in situ generated sulfamoylchloride in *N*,*N*-dimethylacetamide (DMAc).[2526] The resulting sulfamate **39/44** was coupled with *N*-Boc protected, *N*-hydroxysuccinimide activated amino acid in the presence of DBU, after which the protecting groups were removed to give the target compounds.[24] Initially, 2′,3′-*O*-isopropylidene was used to protect the secondary hydroxyls of adenosine, as in the original synthesis.[24] We used this procedure for compounds **27**−**31**, but found the starting product **39** and **40**−**43** to be prone to cyclonucleoside formation, resulting in low yields.[27] Therefore, we switched to *O*-*tert*-butyldimethylsilyl (TBDMS) protection for the synthesis of the *N*-benzyl-L-phenylalanyl congener **32**, based on the synthesis of 5′-*O*-(*N*-salicyl)-sulfamoyladenosine by Ferreras et al.[28] This, however, made a second deprotection step necessary, as aqueous TFA alone did not completely remove TBDMS protection. Hence, following acid treatment, the residue was treated with $Et_3N\cdot 3HF$ in THF to give compound **32**.

For the synthesis of the anhydrohexitol analogues **16**−**21** (Scheme 2) and 2′-deoxy-2′-fluoro-β-D-ribose-analogues **22**−**26** (Scheme 3), we started from the unprotected nucleoside analogues, 1,5-anhydro-2-(adenin-9-yl)-2,3-dideoxy-D-arabino-hexitol (**46**). and 2′-deoxy-2′-fluoroadenosine (**57**), respectively.[29,30] Both were first monomethoxytritylated (MMTr) at the primary hydroxyl and subsequently *O*-TBDMS protected at the secondary hydroxyl, after which the MMTr was removed.

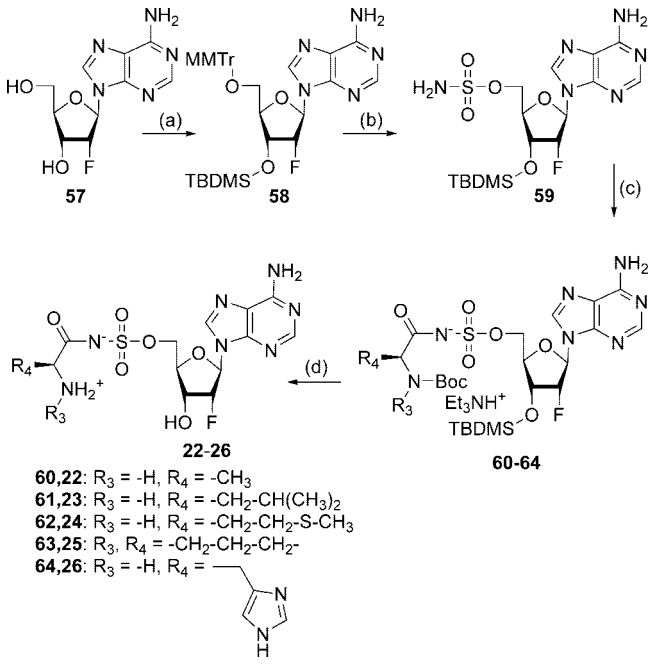
For the synthesis of the altritol compounds (Scheme 4), we started from 1,5-anhydro-2-deoxy-2-(adenin-9-yl)-D-*altro*-hexitol (**65**) and used 1,1,3,3-tetraisopropyldisiloxane-1,3-diyl for pro-

Scheme 2. Synthesis of the Anhydrohexitol Analogues of Figure 3[a]



[a] Reagents and conditions: (a) MMTrCl, pyr, 93%; (b) TBDMSCl, imidazole, DMF, 68%; (c) TsOH in $MeOH/CH_2Cl_2$ (2:8 v/v), 81%; (d) H_2NSO_2Cl, CH_3CN/DMAc, 69%; (e) *N*-Boc L-amino acid *N*-hydroxysuccinimide ester, DBU, DMF, 80−96%; (f) TFA/H_2O (5:2 v/v), 57−87%.

Scheme 3. Synthesis of the 2′-Deoxy-2′-fluoro-β-D-ribose Analogues of Figure 3[a]



[a] Reagents and conditions: (a) (i) MMTrCl, Et_3N, pyr; (ii) TBDMSCl, imidazole, DMF; (iii) TsOH in $MeOH/CH_2Cl_2$ (2:8 v/v), 67%; (b) H_2NSO_2Cl, CH_3CN/DMAc, 88%; (c) *N*-Boc L-amino acid *N*-hydroxysuccinimide ester, DBU, DMF, 61−85%; (d) TFA/H_2O (5:2 v/v), 34−57%.

tection of the secondary hydroxyls.[31] The remainder of the synthesis of these sugar analogues was similar to the synthesis of aaSA analogue **32** (Scheme 1).

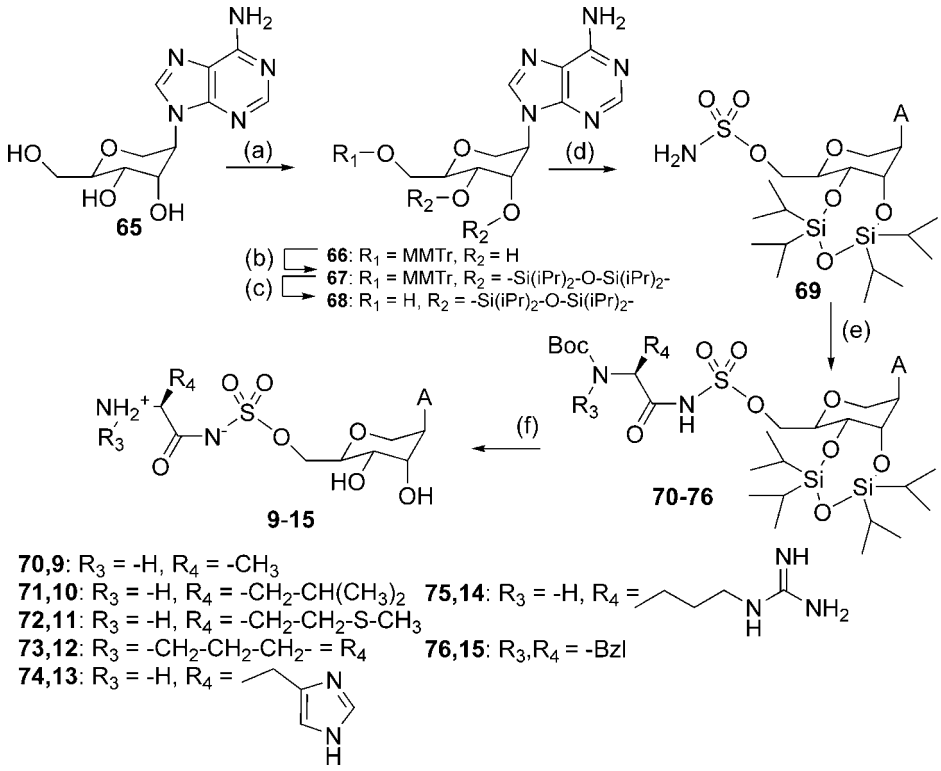
Purification of the final compounds was found to be rather difficult, probably because of their highly polar, zwitterionic nature. Therefore, they were purified by silica gel chromatography and size exclusion chromatography, subsequently. Before use, the compounds were further purified by reverse phase HPLC.

Biological Evaluation

a. Immunosuppressive Properties of the 20 aaSA Compounds. All 20 aaSA analogues were screened for their immunosuppressive potential (Table 1), by determining their ability to inhibit the allogeneic mixed lymphocyte reaction (MLR), a laboratory test which is clinically used as a predictive in vitro test of in vivo transplant rejection.[32] Interestingly, all 20 compounds showed activity in the 0.1−10 μM range. For reference purposes, we also subjected mupirocin to this test and this compound showed no immunosuppressive effect up to the highest concentration used (10 μM).

As we expected that the simultaneous inhibition of multiple aminoacyl-tRNA synthetases would lead to an increase in the biological effect, we then evaluated the effect of combinations of aaSA compounds with other aaSA compounds, or with the clinically used immunosuppressants rapamycin (Rapa) or cyclosporin A (CsA). Rapamycin is an inhibitor of mTOR (mammalian target of rapamycin), while cyclosporine A is a calcineurin inhibitor. In practice, this was done by comparing the MLR inhibiting effect of compounds alone with the effect of compounds together at the same concentration. Testing for statistical significance was done by 2×2 multifactorial analysis of variance (ANOVA).[33] The results of these tests are summarized in Table 2; interestingly, all combinations of aaSA compounds showed synergism. In contrast, no statistically significant interaction was found between aaSA compounds and rapamycin. With cyclosporine A, subadditivity was found in all but one of the combinations tested. A possible explanation for the antagonistic effect seen between aaRS inhibitors and CsA would be that aaRS inhibition limits the synthesis of proteins that are necessary for the action of this immunosup-

Scheme 4. Synthesis of the Altritol Analogues of Figure 3[a]



[a] Reagents and conditions: (a) MMTrCl, pyr; (b) $TIPDSCl_2$, pyr, 80% over steps a and b; (c) TsOH in $MeOH/CH_2Cl_2$ (2:8, v/v), 71%; (d) H_2NSO_2Cl, CH_3CN/DMAc, 71%; (e) *N*-Boc L-aminoacid *N*-hydroxysuccinimide ester, DBU, DMF, 90%; (f) (i) TFA/H_2O (5:2, v/v); (ii) $Et_3N \cdot 3HF$, THF, 40−71%.

Table 1. Immunosuppressive Activity of aaSA Compounds

cmpd	MLR IC_{50}[a] (μM)	cmpd	MLR IC_{50} (μM)
AsnSA	0.3	HisSA	1.0
CysSA	0.4	LeuSA	1.1
MetSA	0.5	AlaSA	1.7
TrpSA	0.5	AspSA	2.4
ProSA	0.7	SerSA	4.8
GlnSA	0.8	GluSA	4.9
GlySA	0.8	ThrSA	5.2
PheSA	1.0	IleSA	5.6
LysSA	1.0	TyrSA	5.6
ArgSA	1.0	ValSA	5.6

[a] MLR IC_{50} values for the analogues of aaSA.

pressant: this may either be the direct molecular target of CsA or a protein downstream of the target.

b. Immunosuppressive Properties of the Modified aaSA Compounds. For each of the modified aaSA compounds, the IC_{50} in the MLR was determined (Table 3). The compounds were active in the same concentration range as the parent compounds. Replacement of the sugar moiety by anhydrohexitol (**16**−**21**) was found to decrease the activity. In contrast, replacement of the sugar by altritol (**9**−**14**) or 2′-deoxy-2′-fluoro-ribose (**22**−**26**) gave similar or slightly better activity. The aminoacyl-modified analogues (**27**−**32**) also showed good immunosuppressive activity. Notable in this series is the *N*-benzyl-L-Phe containing compound **32**, which was found to have an IC_{50} of 200 nM. To evaluate the modularity of our modifications, we prepared a more extensively modified compound combining the altritol sugar replacement with the *N*-benzyl-L-Phe at the aminoacyl position. This compound (**15**) inhibited the MLR with an IC_{50} of 80 nM, which is comparable to the MLR-IC_{50} of cyclosporin A (74 ± 8 nM).[32]

We also evaluated several combinations of modified aaSA analogues with aaSA analogues (Table 4): in all 3 combinations tested, we found synergism. Interestingly, synergism was also found when combining **32** with PheSA. This was surprising, as both compounds were expected to target PheRS, and this result suggests a more complex mode of action for **32** than inhibition of PheRS alone. As we did not have access to enzymatic assays to evaluate our compounds against isolated aaRS, it is difficult to prove the direct relationship between aaRS inhibition and the inhibitory effects observed with these compounds against lymphocytes. Based on the data available in this article, the overall growth-inhibitory activity of single compounds may be the result of synergistic inhibition of multiple aaRS.

Conclusions and Discussion

Though lacking in whole-cell antibacterial activity, several of the compounds reported in this article show a pronounced effect on the growth of stimulated immune cells, at micromolar and submicromolar concentrations. We also show that combining multiple compounds that target aaRS leads to a significant synergistic effect with respect to the immunosuppressive activity. Finally, the potent immunosuppressive effects found with these agents provide sufficient arguments for the screening of developmental antibacterials that target aaRS enzymes for their immunosuppressive effects in humans, because even slight immunosuppressive effects may hamper the use of these compounds as systemic antibiotics in humans.

Experimental Section

Chemistry. General Information. Reagents and solvents were from commercial suppliers (Acros, Sigma-Aldrich, Bachem, Novabiochem) and used as provided, unless indicated otherwise. DMF and THF were analytical grade and were stored over 4 Å molecular sieves. All other solvents used for reactions were analytical grade and used as provided. Reactions were carried out in oven-dried

Table 2. Combination Studies 1[a]

cmpd **1** (μM)	individual (%)	cmpd **2** (μM)	individual (%)	combined (%)	difference (%)	*P* value
PheSA (0.5)	10.8 ± 7.4	TrpSA (0.5)	27.6 ± 8.6	90.9 ± 7.1	52.5[b]	<0.0001
LysSA (1.0)	67.0 ± 6.5	ProSA (0.1)	−2.7 ± 9.8	87.0 ± 7.1	22.7[b]	0.0105
LeuSA (0.5)	28.4 ± 6.3	MetSA (0.5)	16.3 ± 6.9	94.5 ± 7.0	49.8[b]	<0.0001
TrpSA (0.5)	27.6 ± 8.6	MetSA (0.5)	28.4 ± 6.9	96.9 ± 7.1	40.9[b]	0.0001
ProSA (0.5)	18.6 ± 5.0	LeuSA (0.5)	16.3 ± 6.3	92.5 ± 6.8	57.6[b]	<0.0001
TrpSA (0.1)	2.3 ± 22.8	CsA (0.01)	32 ± 6.9	−4.6 ± 28.6	−38.9	0.0611
TrpSA (0.1)	2.3 ± 22.8	Rapa (0.0004)	13.1 ± 13.5	−4.7 ± 11.3	−20.1	0.7412
AlaSA (0.1)	6.5 ± 8.2	CsA (0.06)	65.7 ± 5.9	36.3 ± 9.9	−35.9[b]	0.0018
AlaSA (0.5)	4.9 ± 13.7	Rapa (0.0008)	32.5 ± 6.5	55.8 ± 15.4	18.4	0.2121
HisSA (0.1)	12.4 ± 6.1	CsA (0.06)	65.7 ± 5.9	54.4 ± 7.7	−24.1[b]	0.0043
HisSA (0.1)	11.1 ± 25.9	Rapa (0.0004)	13.1 ± 13.5	6.8 ± 19.2	−17.4	0.3836
PheSA (0.1)	10.7 ± 9.6	CsA (0.08)	75.2 ± 6.0	62.1 ± 3.7	−23.8[b]	0.0057
PheSA (0.5)	53.4 ± 17.8	Rapa (0.0004)	13.1 ± 13.5	93.1 ± 21.2	26.6	0.1703
LeuSA (0.1)	11.4 ± 11.0	CsA (0.06)	65.7 ± 5.9	32.4 ± 10.9	−44.7[b]	0.0014
LeuSA (0.1)	16.0 ± 18.6	Rapa (0.0008)	32.5 ± 6.5	26.5 ± 10.1	−22	0.0562
ProSA (0.1)	−4.1 ± 8.0	CsA (0.06)	65.7 ± 5.9	37.2 ± 8.2	24.4[b]	0.0101

[a] MLR inhibititory activity of combinations of aaSA compounds with aaSA compounds and with CsA/Rapa. For each combination, the meaningful difference with lowest P value is reported. Activities are % inhibition ± SD. Differences are differences between observed effect of combinations and sum of activities of individual compounds. [b] Indicates significant differences ($P < 0.05$).

Table 3. Immunosuppressive Activity of Modified aaSA Compounds

cmpd	MLR IC_{50}[a] (μM)	cmpd	MLR IC_{50} (μM)
9	0.3	**21**	0.6
10	1	**22**	0.4
11	5.2	**23**	0.45
12	0.75	**24**	0.4
13	1.2	**25**	0.3
14	0.6	**26**	0.45
15	0.08	**27**	0.6
16	5.5	**28**	0.55
17	6.8	**29**	5.5
18	5.5	**30**	4.7
19	2.8	**31**	0.55
20	6.7	**32**	0.2

[a] MLR IC_{50} values for the modified analogues of aaSA.

glassware under a nitrogen atmosphere and stirred at room temperature, unless indicated otherwise.

^{1}H and ^{13}C NMR spectra of the compounds were recorded on a 200 MHz Varian Gemini spectrometer or a Bruker UltraShield Avance 300 MHz spectrometer. Spectra were recorded in DMSO-d_6, D_2O, or $CDCl_3$. The chemical shifts are expressed as δ values in parts per million (ppm), using the residual solvent peaks (chloroform: ^{1}H, 7.26 ppm, ^{13}C, 77.36 ppm; DMSO: ^{1}H, 2.50 ppm, ^{13}C, 39.60 ppm; HOD: ^{1}H, 4.79 ppm) as a reference. For ^{13}C spectra in D_2O, ~0.1% dioxane (67.19 ppm) was added as a reference. Coupling constants are given in Hertz (Hz). The peak patterns are indicated by the following abbreviations: bs = broad singlet, d = doublet, m = multiple, q = quadruplet, s = singlet, and t = triplet. High resolution mass spectra were recorded on a quadrupole time-of-flight mass spectrometer (Q-Tof-2, Micromass, Manchester, U.K.) equipped with a standard ESI interface; samples were infused in 2-propanol/H_2O (1:1, v/v) at 3 μL·min^{-1}.

For TLC, precoated aluminum sheets were used (Merck, Silica gel 60 F_{254}). The spots were visualized by UV light. Column chromatography was performed on ICN silica gel 60A 60−200. For size exclusion chromatography, a 2 × 30 cm column of Sephadex LH-20 was used as the solid phase and MeOH/H_2O (7:3, v/v) as the eluent. Preparative HPLC was done using a Waters Xbridge Prep. C18 (19 × 150 mm) column connected to a Waters 1525 binary HPLC pump and a Waters 2487 Dual Absorbance Preparative Detector. Method HPLC1: solvent A = H_2O, solvent B = CH_3CN. Method HPLC2: solvent A = triethylammonium bicarbonate 50 mM, CH_3CN 1%, H_2O 99%; solvent B = triethylammonium bicarbonate 50 mM, H_2O 1%, CH_3CN 99%. Eluent compositions are expressed as v/v.

Synthesis of Analogues of 5′-*O*-(*N*-Aminoacyl)-sulfamoyladenosine Containing Nonproteinogenic Amino Acids (27−32). Synthesis of 2′,3′-*O*-Isopropylidene-5′-*O*-sulfamoyladenosine (39). To an ice-cooled solution of 2′,3′-*O*-isopropylidene-adenosine (8.00 g, 26.0 mmol, 1.0 equiv) in *N*,*N*-dimethylacetamide (40 mL) was added an ice-cooled, freshly prepared 2.0 M solution of sulfamoyl chloride in CH_3CN (30 mL, 2.3 equiv).[34] After stirring for 1 h at rt, the reaction mixture was cooled to 0 °C and the reaction was quenched by the subsequent additions of Et_3N (15 mL) and MeOH (50 mL). The volatiles were removed in vacuo and the oily residue was taken up into EtOAc (500 mL), extracted with aqueous $NaHCO_3$ 5% (2 × 150 mL), and brine (2 × 100 mL). Next, the organic layer was dried over anhydrous Na_2SO_4, filtered, and evaporated to give 7.77 g of **39** (21.2 mmol, 82%). Spectral data (^{1}H NMR, ^{13}C NMR, HRMS) were as reported by Heacock et al.[11]

2′,3′-*O*-Bis(*tert*-butyldimethylsilyl)-5′-*O*-sulfamoyladenosine (44). Following the sulfamoylation procedure used for the preparation of **39**, 2′,3′-*O*-bis(*tert*-butyldimethylsilyl)adenosine (10.00 g, 20.2 mmol, 1.0 equiv) was converted to 11.40 g of **44** (19.8 mmol, 98%).[35] NMR spectra (^{1}H NMR, ^{13}C NMR) were as reported by Ferreras et al.[28] HRMS for $C_{22}H_{43}N_6O_6SSi_2$ $[M + H]^+$ calcd, 575.2503; found, 575.2498.

L-(*N*-Boc-azetidine)-2-carboxylic Acid *N*-Hydroxysuccinimide Ester (76). The amino acid L-azetidine-2-carboxylic acid (0.25 g, 2.47 mmol, 1.0 equiv) was dissolved in aqueous Na_2CO_3 solution (10% w/w, 10 mL) and a solution of di*tert*-butyl dicarbonate (0.70 g, 3.21 mmol, 1.3 equiv) in dioxane (5 mL) was added. The mixture was stirred overnight, H_2O was added (15 mL), and the mixture was extracted with Et_2O (2 × 30 mL). The aqueous layer was cooled (0 °C), acidified with concd HCl to pH ~ 2, and extracted with EtOAc (3 × 50 mL). The organic layers were dried (anhydrous Na_2SO_4), filtered, and evaporated. The oily residue (0.50 g) obtained this way was used in the second step without further purification. The residue was dissolved in DMF (15 mL), *N*-hydroxysuccinimide was added (345 mg, 3.0 mmol, 1.2 equiv) and the solution cooled to 0 °C. *N*-(3-Dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (671 mg, 3.50 mmol, 1.4 equiv) was added, and the mixture was allowed to come to rt overnight. The volatiles were removed in vacuo, and the residue was taken up into EtOAc (100 mL). The mixture was washed with HCl 2 N (2 × 100 mL), $NaHCO_3$ satd (100 mL), brine (100 mL), and dried over anhydrous Na_2SO_4. After filtering, the volatiles were removed in vacuo, yielding 615 mg of **76** (2.06 mmol, 85%). ^{1}H NMR (DMSO-d_6): δ 5.02 (dd, 1H, 2-H, J = 5.4 Hz, 9.4 Hz), 3.88 (t, 2H, 4-H_2, J = 7.6 Hz), 2.83 (s, 4H, OSu-H_2), 2.78−2.58 (m, 1H, 3-H_A), 2.30−2.10 (m, 1H, 3-H_B), 1.37 (s, 9H, Boc-C$(CH_3)_3$). ^{13}C NMR (DMSO-d_6): δ 170.1, 167.6, 79.8, 58.0, 47.1, 27.8, 25.5, 20.4. HRMS for $C_{13}H_{19}N_2O_6$ $[M + H]^+$ calcd, 299.1243; found, 299.1248.

2′,3′-*O*-Isopropylidene-5′-*O*-[*N*-(L-(*N*-Boc-azetidine)-2-carbonyl)-sulfamoyl]adenosine (Et_3N Salt; 40). To a solution of **39** (350 mg, 0.91 mmol, 1.0 equiv) and **76** (300 mg, 1.01 mmol, 1.1 equiv) in DMF (10 mL) was added DBU (277 mg, 1.82 mmol, 2 equiv). When TLC analysis (Et_3N 1%, MeOH 5%, CH_2Cl_2) showed complete

Table 4. Combination Experiments with Modified Analogues of aaSA[a]

cmpd 1 (μM)	individual (%)	cmpd 2 (μM)	individual (%)	combined (%)	difference (%)	*P* value
9 (0.5)	68.0 ± 8.2	MetSA(0.5)	1.0 ± 9.8	86.2 ± 9.0	17.2	0.0935
32 (0.1)	31.1 ± 10.1	PheSA(0.5)	−1.8 ± 9.5	92.0 ± 12.7	62.7[b]	0.001
32 (0.1)	31.1 ± 10.1	TrpSA(0.5)	−8.6 ± 9.2	91.6 ± 12.4	69.1[b]	0.0009

[a] MLR inhibititory activity of combinations of modified analogues of aaSA with aaSA. For each combination, the meaningful difference with lowest P value is reported. Activities are % inhibition ± SD. Differences are differences between observed effect of combinations and sum of activities of individual compounds. [b] Indicates significant differences ($P < 0.05$).

conversion of **39** (6 h), the reaction mixture was concentrated in vacuo, and the residue was purified by silica gel LC (Et_3N 1%, MeOH 5% in CH_2Cl_2), giving 473 mg (0.71 mmol, 77%) of **40**. ^{1}H NMR (DMSO-d_6): δ 8.39 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 6.15 (d, 1H, 1′-H, $J = 3.0$ Hz), 5.36 (m, 1H, 2′-H), 5.03 (d, 1H, 3′-H, $J = 5.2$ Hz), 4.38 (m, 1H, 4′-H), 4.18 (dd, 1H, azetidine-2-H, $J = 4.4$ Hz, 8.8 Hz), 4.00 (d, 2H, 5′-H_2, $J = 4.4$ Hz), 3.88−3.44 (m, 2H, azetidine-4-H_2), 2.98 (q, 6H, Et_3NH^+-H_2, $J = 7.2$ Hz), 2.30 (m, 1H, azetidine-3-H_A), 1.79 (m, 1H, azetidine-3-H_B), 1.54 (s, 3H, isopropylidene-H_3), 1.42−0.90 (m, 21H, isopropylidene-H_3, Boc-C(CH_3)$_3$, Et_3NH^+-H_3). ^{13}C NMR (DMSO-d_6): δ 175.6, 156.3, 155.4, 153.0, 149.3, 139.6, 119.0, 113.2, 89.3, 83.9, 83.6, 81.8, 78.0, 67.0, 64.5, 46.9, 45.7, 28.3, 27.1, 25.2, 20.7, 9.0. HRMS for $C_{22}H_{32}N_7O_9S$ $[M + H]^+$ calcd, 570.1982; found, 570.1983.

5′-*O*-[*N*-(L-Azetidine-2-carbonyl)-sulfamoyl]adenosine (27). Compound **40** (440 mg, 0.66 mmol, 1.0 equiv) was dissolved in TFA/H_2O (4 mL, 5:2, v/v) and the reaction mixture was stirred for 2 h, after which the volatiles were evaporated, coevaporated twice with EtOH, and once with EtOH + 1 mL Et_3N, to neutralize any remaining acid. The residue was purified by silica gel LC (Et_3N 1%, MeOH 20 → 50% in CH_2Cl_2) and by size exclusion chromatography, giving 172 mg (0.40 mmol, 61%) of **27** as a white solid after lyophilization from H_2O.

^{1}H NMR (D_2O) δ 8.35 (s, 1H, 8-H), 8.17 (s, 1H, 2-H), 6.08 (d, 1H, 1′-H, $J = 4.4$ Hz), 4.89 (t, 1H, azetidine-2-H, $J = 8.7$ Hz), 4.73 (m, 1H, 2′-H), 4.56−4.34 (m, 4H, 3′-H, 4′-H, 5′-H_2), 4.18−4.03 (m, 1H, azetidine-4-H_A), 3.97−3.82 (m, 1H, azetidine-4-H_B), 2.88−2.71 (m, 1H, azetidine-3-H_A), 2.61−2.44 (m, 1H, azetidine-3-H_A). ^{13}C NMR (D_2O + dioxane) δ 175.0, 156.1, 153.4, 149.5, 140.3, 119.2, 88.0, 82.8, 74.6, 70.7, 69.0, 61.0, 43.9, 24.1. HRMS for $C_{14}H_{20}N_7O_7S$ $[M + H]^+$ calcd, 430.1145; found, 430.1136.

The synthesis and spectroscopic data of other analogues of 5′-*O*-(*N*-aminoacyl)-sulfamoyladenosine containing nonproteinogenic amino acids (**28**−**32**) can be found in the Supporting Information.

Synthesis of the Anhydrohexitol Derivatives (16−21). 2-(Adenin-9-yl)-1,5-anhydro-6-*O*-monomethoxytrityl-2,3-dideoxy-D-arabino-hexitol (47). A suspension of 2-(adenin-9-yl)-1,5-anhydro-2,3-dideoxy-D-arabino-hexitol (2.64 g, 9.95 mmol, 1.0 equiv; **46**) and 4-monomethoxytrityl chloride (3.25 g, 10.5 mmol, 1.05 equiv) in dry pyridine (220 mL) was allowed to react for 2 days at rt.[29] Next, the resulting solution was concentrated under reduced pressure and coevaporated three times with toluene. The residue was purified by column chromatography using 0.5% Et_3N and MeOH 1 → 3% in CH_2Cl_2 as the eluent to give **47** (4.99 g, 93%) as a white foam. ^{1}H NMR (DMSO-d_6/$CDCl_3$ 1:1): δ 8.33 (s, 1H, 8-H), 8.14 (s, 1H, 2-H), 7.50−7.00 (m, 14H, aryl-H, Ade-NH_2), 6.83 (d, 2H, aryl-H, $J = 8.8$ Hz), 4.86 (d, 1H, 4′-OH, $J = 4.0$ Hz), 4.79 (bs, 1H, 2′-H), 4.32 (d, 1H, 1′-H_A, $J = 12.0$ Hz), 3.94 (d, 1H, 1′-H_B, $J = 10.6$ Hz), 3.78−3.50 (m, MMTr-OCH_3, 4′-H), 3.50−3.07 (m, partly obscured by residual H_2O peak, 5′-H, 6′-H_2), 2.41 (m, 1H, 3′-H_A), 1.88 (m, 1H, 3′-H_B). ^{13}C NMR (DMSO-d_6): δ 158.3, 156.3, 152.7, 149.6, 144.8, 142.1, 139.6, 135.4, 130.3, 128.3, 128.0, 126.9, 118.7, 113.3, 85.5, 81.4, 68.3, 63.0, 61.1, 55.1, 50.4, 36.3. HRMS for $C_{31}H_{32}N_5O_4$ $[M + H]^+$ calcd, 538.2454; found, 538.2432.

2-(Adenin-9-yl)-1,5-anhydro-6-*O*-monomethoxytrityl-4-*O*-tert-butyldimethylsilyl-2,3-dideoxy-D-arabino-hexitol (48). A solution of **47** (4.99 g, 9.28 mmol, 1.0 equiv) in DMF (∼100 mL) was reacted with TBDMSCl (3.00 g, 19.9 mmol, 2.1 equiv) in the presence of imidazole (1.40 g, 20.6 mmol, 2.2 equiv) at rt over weekend. The resulting yellow solution was concentrated under reduced pressure and coevaporated (3×) with toluene. The residual oil was purified by silica column chromatography (0.5% Et_3N, MeOH 1→3% in CH_2Cl_2) to give 4.13 g of **48** (6.34 mmol, 68%). ^{1}H NMR (DMSO-d_6/$CDCl_3$ 1:1): δ 8.30 (s, 1H, 8-H), 8.13 (s, 1H, 2-H), 7.50−7.06 (m, 14 H, aryl-H, Ade-NH_2), 6.81 (d, 2H, aryl-H, $J = 9.2$ Hz), 4.81 (bs, 1H, 2′-H), 4.38 (d, 1H, 1′-H_A, $J = 14.2$ Hz), 4.02 (dd, 1H, 1′-H_B, $J = 2.0$ Hz, 11.7 Hz), 3.73 (s, 3H, MMTr-OCH_3), 3.78−3.44 (m, 2H, 4′-H, 5′-H), 3.22 (dd, 1H, 6′-H_A, $J = 5.8$ Hz, 12.4 Hz), 3.07 (dd, 1H, 6′-H_B, $J = 5.1$ Hz, 9.9 Hz), 2.48 (obscured by solvent peak, 3′-H_A), 1.93 (app. dt, 1H, 3′-H_B, $J = 4.0$ Hz, 11.4 Hz), 0.56 (s, 9H, Si-$(CH_3)_3$), −0.26 (s, 3H, Si-CH_3), −0.43 (s, 3H, Si-CH_3).

^{13}C NMR (DMSO-d_6/$CDCl_3$, 1:1 v/v): δ 158.3, 156.1, 152.4, 149.5, 144.4, 139.2, 135.2, 130.2, 128.2, 127.6, 126.7, 118.7, 113.0, 85.6, 81.6, 68.4, 63.1, 54.9, 50.1, 36.4, 25.3, 17.2, −4.6, −5.6. HRMS for $C_{37}H_{46}N_5O_4Si$ $[M + H]^+$ calcd, 652.3319; found, 652.3301.

2-(Adenin-9-yl)-1,5-anhydro-4-*O*-tert-butyldimethylsilyl-2,3-dideoxy-D-arabino-hexitol (49). Compound **48** (4.13 g, 6.34 mmol) was added to an ice-cooled solution of 2% *p*-toluenesulfonic acid monohydrate in MeOH/CH_2Cl_2 (2:8, v/v). After one hour, TLC (CH_2Cl_2/MeOH 9:1 v/v) indicated complete conversion, and the reaction was quenched by the addition of $NaHCO_3$. Silica gel (20 g) was added and the volatiles were removed under reduced pressure. The adsorbed residue was purified by silica gel chromatography (CH_2Cl_2/MeOH 9:1 v/v) giving **49** (1.95 g, 5.14 mmol, 81%).

^{1}H NMR (DMSO-d_6): δ 8.30 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.24 (s, 2H, Ade-NH_2), 4.80 (bs, 1H, 2′-H), 4.72 (t, 1H, 6′-OH, $J = 5.7$ Hz), 4.21 (d, 1H, 1′-H_A, $J = 12.4$ Hz), 3.91 (dd, 1H, 1′-H_B, $J = 2.6$ Hz, 14.1 Hz), 3.85−3.45 (m, 3H, 6′-H_2, 4′-H), 3.26 (m, 1H, 5′-H), 2.39 (d, 1H, 3′-H_A, $J = 13.6$ Hz), 1.96 (m, 1H, 3′-H_B), 0.79 (s, 9H, Si−C$(CH_3)_3$), −0.07 (s, 3H, Si-CH_3), −0.13 (s, 3H, Si-CH_3). ^{13}C NMR (DMSO-d_6): δ 156.2, 152.6, 149.6, 139.8, 118.5, 82.7, 68.1, 62.8, 60.1, 50.1, 36.2, 25.6, 17.6, −4.7, −5.1. HRMS for $C_{17}H_{30}N_5O_3Si$ $[M + H]^+$ calcd, 380.2118; found, 380.2119.

2-(Adenin-9-yl)-1,5-anhydro-6-*O*-sulfamoyl-4-*O*-tert-butyldimethylsilyl-2,3-dideoxy-D-arabino-hexitol (50). Following the procedure used for the synthesis of **39**, compound **49** (1.0 g, 2.63 mmol, 1.0 equiv) was sulfamoylated to give **50** (0.83 g, 1.80 mmol, 69%). ^{1}H NMR (DMSO-d_6): δ 8.20 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.54 (bs, 2H, sulfamoyl-NH_2), 7.24 (bs, 2H, Ade-NH_2), 4.81 (bs, 1H, 2′-H), 4.40−4.10 (m, 3H, 6′-H_2, 1′-H_A), 3.97 (m, 1H, 1′-H_B), 3.75−3.35 (m, 2H, 4′-H, 5′-H), 2.5 (3′-H_A, hidden under DMSO signal), 2.02 (m, 1H, 3′-H_B), 0.80 (s, 9H, Si−C$(CH_3)_3$), −0.04 (s, 3H, Si-CH_3), −0.09 (s, 3H, Si-CH_3). ^{13}C NMR (DMSO-d_6): δ 156.3, 152.7, 149.6, 139.5, 118.6, 79.5, 68.1, 67.9, 63.2, 50.0, 36.0, 25.6, 17.6, −4.5, −5.1. HRMS for $C_{17}H_{29}N_6O_5SSi$ $[M - H]^-$ calcd, 457.1689; found, 457.1690.

2-(Adenin-9-yl)-1,5-anhydro-4-*O*-tert-butyldimethylsilyl-6-*O*-[*N*-(*N*-Boc-L-alanyl)sulfamoyl]-2,3-dideoxy-D-arabino-hexitol (51). To a solution of compound **50** (200 mg, 0.44 mmol, 1.0 equiv) and *N*-Boc-L-alanine *N*-hydroxysuccinimide ester (187 mg, 0.65 mmol, 1.5 equiv) in dry DMF (5 mL) was added DBU (165 mg, 1.09 mmol, 2.5 equiv). The reaction mixture was stirred at rt until complete conversion (2 h) of the starting compound (TLC, HOAc 1%, MeOH 5% in CH_2Cl_2). Next the volatiles were evaporated and the oily residue was purified by silica gel column chromatography (HOAc 1%, MeOH 5% in CH_2Cl_2) giving the title compound **51** as a white solid (252 mg, 0.40 mmol, 91%). ^{1}H NMR (DMSO-d_6): δ 10.6 (bs, 1H, α-NH) 8.22 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.25 (bs, 2H, Ade-NH_2), 6.76 (bs, 1H, sulfamoyl-NH), 4.80 (bs, 2′-H), 4.40−4.10 (m, 3H, 1'H_A, 6′-H_2,), 4.00−3.75 (m, 2 H, 1′-

H_B, α-H), 3.70−3.50 (m, partially obscured by H_2O peak, 5′-H, 4′-H), 2.44 (m, partially obscured by DMSO-d_6 peak, 3′-H_A), 2.10−1.90 (m, 1H, 3′-H_B), 1.36 (s, 9H (Boc-C-$(CH_3)_3$), 1.19 (d, 3H, β-H_3, J = 7.4 Hz), 0.78 (s, 9H, Si−C$(CH_3)_3$), −0.07 (s, 3H, Si-CH_3), −0.12 (s, 3H, Si- CH_3). ^{13}C NMR (DMSO-d_6): δ 173.1, 156.2, 155.2, 152.6, 149.6, 139.7, 118.5, 79.5, 78.2, 69.8, 68.1, 63.3, 50.8, 50.0, 36.1, 28.3, 25.3 and 25.6, 17.6, 8.7, −4.6, −5.1. HRMS for $C_{25}H_{44}N_7O_8SSi$ $[M + H]^+$ calcd, 630.2741; found, 630.2740.

2-(Adenin-9-yl)-1,5-anhydro-6-*O*-[*N*-(L-alanyl)sulfamoyl]-2,3-dideoxy-D-arabino-hexitol (16). A solution of compound **51** (242 mg, 0.38 mmol, 1.0 equiv) in TFA/H_2O (4 mL, 5:2, v/v) was stirred for 5 h at rt. Next, the volatiles were removed by rotary evaporation and coevaporated (EtOH, 3×) to give a white foam. This foam was further purified by silica gel chromatography (HOAc 1%, MeOH 30% in CH_2Cl_2), and the fractions containing **16** were collected and evaporated to dryness. The residue was further purified by size exclusion column chromatography (2×, 30% H_2O in MeOH). Finally, lyophilization from H_2O yielded 135 mg (0.32 mmol, 87%) of pure **16**.

^{1}H NMR (D_2O): δ 8.38 (s, 1H, 8-H), 8.13 (s, 1H, 2-H), 4.83 (bs, 1H, 2′-H), 4.47−4.34 (m, 3H, 1′-H_A, 6′-H_2), 4.12 (dd, 1H, 6′-H_B, J = 2.6 Hz, 13.1 Hz), 3.88 (q, 1H, α-H, J = 7.2 Hz), 3.79−3.66 (m, 2H, 5′-H, 4′-H), 2.55−2.45 (m, 1H, 3′-H_A), 2.14−2.03 (m, 1H, 3′-H_B), 1.51 (d, 3H, β-H_3, J = 7.2 Hz). ^{13}C NMR (D_2O + dioxane): δ 177.2, 156.0, 152.9, 149.3, 141.7, 118.6, 80.1, 69.1, 69.0, 61.6, 52.1, 51.4, 35.5, 17.2. HRMS for $C_{14}H_{20}N_7O_6S$ $[M - H]^-$ calcd, 414.1196; found, 414.1187.

The synthesis and spectroscopic data of the anhydrohexitol derivatives **17**−**21** can be found in the Supporting Information.

Synthesis of the 2′-Deoxy-2′-fluoroadenosine derivatives (22−26). 3′-*O*-(*tert*-Butyldimethylsilyl)-2′-deoxy-2′-fluoroadenosine (58). 2′-Deoxy-2′-fluoroadenosine (**57**, 1.16 g, 4.31 mmol, 1.0 equiv) was coevaporated with anhydrous pyridine (3×) and then dissolved in anhydrous pyridine (50 mL).[30] Triethylamine (567 mg, 5.60 mmol, 1.3 equiv) was added to the solution followed by monomethoxytrityl chloride (2.26 g, 7.32 mmol, 1.7 equiv). After stirring overnight at rt, the volatiles were evaporated. The residue was dissolved in DMF (50 mL) and *tert*-butyldimethylsilyl chloride (974 mg, 6.46 mmol, 1.5 equiv) and imidazole (733 mg, 10.77 mmol, 2.5 equiv) were added. The mixture was stirred at rt for 1 day and the volatiles were evaporated. Finally, the residue was treated with a 2% solution of *p*-toluenesulfonic acid monohydrate in MeOH/CH_2Cl_2 (150 mL, 2:8, v/v) at 0 °C for 2 h. The solution was then diluted with CH_2Cl_2 (400 mL) and the reaction was quenched with 5% aq $NaHCO_3$ (150 mL). The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 × 100 ml). The organic layers were combined, dried (Na_2SO_4), filtered, and evaporated. The residue was further purified by silical gel chromatography (MeOH 2 → 6% in CH_2Cl_2) to give **58** as a white solid (1.12 g, 67% yield). ^{1}H NMR (DMSO-d_6): δ 8.38 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.39 (s, 2H, Ade-NH_2), 6.23 (dd, 1H, 1′-H, J = 16.6, 3.4 Hz), 5.56 (ddd, 1H, 2′-H, J = 52.4 Hz), 5.30 (t, 1H, 5′-OH), 4.67−4.77 (m, 1H, 3′-H), 3.99 (m, 1H, 4′-H), 3.42−3.78 (m, 2H, 5′-H_2), 0.91 (s, 9H, C$(CH_3)_3$), 0.14 and 0.12 (2 s, 6H, Si$(CH_3)_2$). ^{13}C NMR (DMSO-d_6): δ 156.4, 153.0, 149.2, 140.0, 119.3, 92.5 (J = 189.2 Hz), 85.9 (J = 32.0 Hz), 84.9, 70.2 (J = 15.3 Hz), 60.4, 25.8, 18.0, −4.8, −5.0. HRMS for $C_{16}H_{26}N_5O_3FSi$ $[M + H]^+$ calcd, 384.1867; found, 384.1870.

3′-*O*-(*tert*-Butyldimethylsilyl)-2′-deoxy-2′-fluoro-5′-*O*-sulfamoyladenosine (59). Following the procedure used for the synthesis of **39**, compound **58** (505 mg, 1.32 mmol, 1.0 equiv) was sulfamoylated to give **59** (537 mg, 1.16 mmol, 88%). ^{1}H NMR (DMSO-d_6): δ 8.30 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.62 (bs, 2H, SO_2NH_2), 7.39 (bs, 2H, Ade-NH_2), 6.29 (dd, 1H, 1′-H, J = 18.8 Hz, 2.2 Hz), 5.61 (ddd, 1H, 2′-H, J = 55.0 Hz), 4.98−4.83 (m, 1H, 3′-H), 4.34−4.17 (m, 3H, 4′-H, 5′-H_2), 0.91 (s, 9H, Si−C$(CH_3)_3$), 0.17 and 0.15 (2 × s, 6H, Si$(CH_3)_2$). ^{13}C NMR (DMSO-d_6): δ 156.4, 153.0, 149.1, 139.9, 119.3, 92.3 (J = 189.2 Hz), 86.2 (J = 33.6 Hz), 80.4, 70.2 (J = 15.3 Hz), 67.8, 25.6, 17.8, 0.2. HRMS for $C_{16}H_{27}N_6O_5FSSi$ $[M + H]^+$ calcd, 463.1595; found, 463.1591.

5′-*O*-[*N*-(*N*-Boc-L-alanyl)sulfamoyl]-3′-*O*-(*tert*-butyldimethylsilyl)-2′-deoxy-2′-fluoroadenosine Et_3N Salt (60). Following the procedure used for the synthesis of **51**, compound **59** (185 mg, 0.40 mmol, 1.0 equiv) and *N*-Boc-L-alanine *N*-hydroxysuccinimide ester (115 mg, 0.40 mmol, 1.0 equiv) were reacted with DBU (91 mg, 0.6 mmol, 1.5 equiv). The product isolated by column chromatography was dried under vacuum to afford 179 mg of solid **60** (0.24 mmol, 61%). ^{1}H NMR (DMSO-d_6): δ 8.40 (s, 1H, 8-H), 8.15 (s, 1H, 2-H), 7.36 (s, 2H, Ade-NH_2), 6.24 (dd, 1H, 1′-H, J = 16.2 Hz, 4.4 Hz), 6.06 (d, 1H, Ala-NH, J = 7.2 Hz), 5.62 (ddd, 1H, 2′-H, J = 52.8 Hz), 4.76−4.64 (m, 1H, 3′-H), 4.19−3.96 (m, 3H, 4′-H, 5′-H_2), 3.71 (m, 1H, α-H), 3.08 (q, 6H, Et_3NH^+-H_2, J = 11.0 Hz), 1.35 (s, 9H, Boc-C$(CH_3)_3$), 1.16 (t, 9H, Et_3NH^+-H_3), 1.17 (d, 3H, β-H_3), 0.90 (s, 9H, Si−C$(CH_3)_3$), 0.14 and 0.12 (2 × s, 6H, Si$(CH_3)_2$). ^{13}C NMR (DMSO-d_6): δ 177.3, 156.4, 155.1, 153.4, 149.6, 139.9, 119.2, 92.5 (J = 192.3 Hz), 85.3 (J = 32.0 Hz), 82.7, 78.1, 71.1 (J = 15.4 Hz), 66.9, 52.0, 46.1, 28.5, 25.9, 19.9, 18.1, 8.9, −4.9. HRMS for $C_{24}H_{40}N_7O_8FSSi$ $[M + H]^+$ calcd, 634.2490; found, 634.2497.

5′-*O*-[*N*-(L-Alanyl)sulfamoyl]-2′-deoxy-2′fluoroadenosine (22). Following the procedure used for the synthesis of compound **16**, compound **60** (194 mg, 0.26 mmol, 1.0 equiv) was deprotected to give **22** as a white solid (51 mg, 46% yield). ^{1}H NMR (DMSO-d_6): δ 8.36 (s, 1H, 8-H), 8.16 (s, 1H, 2-H), 7.35 (s, 2H, Ade-NH_2), 6.25 (dd, 1H, 1′-H, J = 17.6, 2.4 Hz), 5.47 (ddd, 1H, 2′-H, J = 53.4 Hz), 4.61−4.37 (m, 1H, 3′-H), 4.27−4.05 (m, 3H, 4′-H, 5′-H_2), 3.49 (q, 1H, α-H, J = 11.1 Hz), 1.30 (d, 3H, β-H_3, J = 10.5 Hz). ^{13}C NMR (DMSO-d_6): δ 173.6, 156.3, 153.0, 149.2, 139.5, 119.1, 93.4 (J = 186.19 Hz), 85.7 (J = 32.0 Hz), 81.3, 69.1 (J = 16.7 Hz), 67.2, 50.9, 17.3. HRMS for $C_{13}H_{18}N_7O_6FS$ $[M + H]^+$ calcd, 420.1101; found, 420.1101.

The synthesis and spectroscopic data of the 2′-deoxy-2′-fluoroadenosine derivatives **23**−**26** can be found in the Supporting Information.

Synthesis of the Altritol Derivatives (9−15). 2′-(Adenin-9-yl)-1′,5′-anhydro-2-deoxy-6′-*O*-monomethoxytrityl-3′,4′-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-D-*altro*-hexitol (67). To a suspension of 2-(adenin-9-yl)-1,5-anhydro-2-deoxy-D-*altro*-hexitol (**65**, 206 mg, 0.73 mmol, 1.0 equiv) in dry pyridine (15 mL) was added monomethoxytrityl chloride (226 mg, 0.73 mmol, 1.0 equiv) and the resulting mixture was stirred overnight.[31] TLC (CH_2Cl_2/MeOH, 9:1 v/v) indicated complete conversion of the starting material **65**, and 1,3-dichloro-1,1,3,3-tetrasiopropyldisiloxane (0.25 mL, 0.80 mmol, 1.1 equiv) was added and the resulting solution was stirred for 3 days until TLC (CH_2Cl_2/MeOH, 9:1, v/v) indicated complete conversion of the monomethoxytritylated intermediate **66**. The volatiles were removed under reduced pressure and the resulting viscous oil was purified using column chromatography (MeOH 0→6%, Et_3N 1% in CH_2Cl_2) to yield pure **67** (465 mg, 0.58 mmol, 80%).

^{1}H NMR (200 MHz, $CDCl_3$): δ 8.58 (s, 1H, 8-H), 8.34 (s, 1H, 2-H), 7.52−7.43 (m, 4H, Ar−H), 7.40−7.14 (m, 10H, Ar−H), 4.75 (s, 1H, 2′-H), 4.43 (d, 1H, 1′-H_A, J = 12.4 Hz), 4.24 (d, 1H, 1′-H_B, J = 12.8 Hz), 4.18−4.05 (m, 2H, 3′-H, 4′-H), 3.93−3.82 (m, 1H, 5′-H), 3.79 (s, 3H, MMTr-O-CH_3), 3.46 (dd, 1H, 6′-H_A, J = 9.8 Hz, 1.8 Hz), 3.29 (dd, 1H, 6′-H_B, J = 4.1 Hz, 9.9 Hz), 1.22−0.65 (m, 28H, TIPDS-CH(CH_3)$_2$).

^{13}C NMR ($CDCl_3$): δ 158.6, 155.5, 153.1, 150.5, 144.7, 140.52, 135.8, 130.5, 128.5, 127.8, 126.8, 117.6, 113.0, 86.0, 76.3, 69.5, 67.9, 64.2, 62.7, 55.8, 55.1, 17.2, 14.0, 12.8. HRMS for $C_{43}H_{58}N_5O_6Si_2$ $[M + H]^+$: calcd, 796.3926; found, 796.3930.

2-(Adenin-9-yl)-1,5-anhydro-2-deoxy-3,4-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-D-*altro*-hexitol (68). To an ice-cooled solution of 2% *p*-toluenesulfonic acid monohydrate in MeOH/CH_2Cl_2 (200 mL, v/v) was added 3.80 g (4.77 mmol, 1.0 equiv) of **67**. After 1 h at 0 °C, TLC (CH_2Cl_2/MeOH, 9:1, v/v) indicated complete conversion of **67** to a lower R_f compound. The reaction was quenched by the addition of solid $NaHCO_3$ and the reaction mixture was adsorbed onto 10 g of silica by rotary evaporation and subjected to column chromatography with gradient elution (MeOH 0 → 10% in CH_2Cl_2) to give solid **68** (2.15 g, 4.10 mmol, 86%).

^{1}H NMR (DMSO-d_6): δ 8.32 (s, 1H, 8-H), 8.12 (s, 1H, 2-H), 7.26 (bs, Ade-NH_2), 4.85−4.72 (m, 2H, 3′-H, 6′-OH), 4.68−4.60 (m, 1H, 2′-H), 4.24−4.08 (m, 2H, 1′-H_2), 4.10−3.92 (m, 1H, 4′-H), 3.80−3.55 (m, 3H, 6′-H_2, 5′-H), 1.20−0.75 (m, 28H, TIPDS-C*H*(CH_3)$_2$).

^{13}C NMR (DMSO-d_6): δ 156.2, 152.7, 149.9, 139.7, 118.5, 77.5, 69.5, 67.9, 63.3, 59.9, 55.0, 17.1, 13.1, 12.7, 12.4. HRMS for $C_{23}H_{42}N_5O_5Si_2$ $[M+H]^+$ calcd, 524.2724; found, 524.2726.

2-(Adenin-9-yl)-1,5-anhydro-2-deoxy-6-*O*-sulfamoyl-3,4-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-D-*altro*-hexitol (69). Following the procedure used for the synthesis of **39**, compound **68** (1.50 g, 2.86 mmol, 1.0 equiv) was sulfamoylated to give **69** (1.24 g, 2.06 mmol, 72%) as a white solid.

^{1}H NMR (DMSO-d_6): δ 8.23 (s, 1H, 8-H), 8.13 (s, 1H, 2-H), 4.86 (bs, 1H, 3′-H), 4.65 (bs, 1H, 2′-H), 4.40−4.14 (m, 4H, 6′-H_2, 1′-H_2), 3.96−3.78 (m, 2H, 4′-H, 5′-H), 1.25−0.75 (m, 28H, 4 × TIPDS-C*H* (CH_3)$_2$).

^{13}C NMR (DMSO-d_6): δ 156.3, 152.8, 149.9, 139.3, 118.5, 74.4, 68.9, 68.1, 67.9, 63.4, 55.1, 12.4, 12.7, 13.3, 17.1. HRMS for $C_{23}H_{42}N_6O_7SSi_2$ $[M + H]^+$ calcd, 603.2452; found, 603.2458.

2-(Adenin-9-yl)-1,5-anhydro-2-deoxy-6-*O*-[(*N*-Boc-L-alanyl)sulfamoyl]-3,4-*O*-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-D-*altro*-hexitol (70). Following the procedure used for the synthesis of **51**, compound **69** (300 mg, 0.50 mmol, 1.0 equiv) was reacted with *N*-Boc-L-alanine *N*-hydroxysuccinimide ester (280 mg, 0.98 mmol, 2.0 equiv) and DBU (114 mg, 1.05 mmol, 2.1 equiv) to give 349 mg of **70** (0.45 mmol, 90%).

^{1}H NMR (CD_3OD) δ 8.47 (s, 1H, 8-H), 8.21 (s, 1H, 2-H), 4.90−4.70 (partly obscured by D_2O peak, 2H, 2′-H, 3′-H), 4.63−4.48 (m, 2H, 6′-H_2), 4.47−4.26 (m, 2H, 1′-H_2), 4.16−3.75 (m, 3H, α-H, 4′-H, 5′-H), 1.30−0.76 (m, 40H, 4 × -C*H*(CH_3)$_2$, Boc-$(CH_3)_3$, Ala-CH_3). ^{13}C NMR (CD_3OD): δ 153.8, 153.2, 152.0, 142.0, 119.5, 75.7, 72.6, 70.5, 68.9, 64.8, 57.3, 28.7, 18.0, 17.6, 15.1, 14.4, 14.1. HRMS for $C_{33}H_{56}N_7O_{10}SSi_2$ $[M + H]^+$ calcd, 774.3348; found, 774.3334.

2-(Adenin-9-yl)-1,5-anhydro-2-deoxy-6-*O*-[(L-alanyl)sulfamoyl]-D-*altro*-hexitol (9). A solution of compound **70** (330 mg, 0.43 mmol, 1.0 equiv) in TFA/H_2O (5 mL, 5:2 v/v) was stirred for 4 h at rt. Next, the volatiles were removed under reduced pressure and the residue was coevaporated three times with EtOH and once with EtOH + Et_3N (1 mL). The residue was dried (overnight, P_2O_5, vacuum) and THF (20 mL) and $Et_3N\cdot 3HF$ (430 μL, 2.64 mmol, 18 equiv) were added. The solution was stirred overnight (rt), after which the volatiles were removed by rotary evaporation. The residue was purified by silica gel column chromatography (MeOH 30% in CH_2Cl_2) and further purified by Sephadex LH-20 column chromatography. Fractions containing **9** were combined and lyophilized from H_2O, giving 145 mg (0.31 mmol, 71%) of pure **9** as a white solid. ^{1}H NMR (D_2O): δ 8.44 (s, 1H, 8-H), 8.18 (s, 1H, 2-H), 4.74−4.68 (m, 1H, 2′-H), 4.48−4.24 (m, 5H, 1′-H_2, 3′-H, 6′-H_2), 4.07 (dt, 1H, 5′-H, J = 3.1 Hz, J = 6.4 Hz), 3.88 (q, 1H, α-H_3, J = 7.2 Hz), 3.77 (dd, 1H, 4′-H, J = 3.2 Hz, J = 9.5 Hz), 1.52 (d, 3H, β-H_3, J = 7.2 Hz). ^{13}C NMR (D_2O + dioxane): δ 177.3, 156.1, 153.2, 149.7, 141.7, 118.5, 74.5, 69.2, 68.1, 64.3, 64.2, 55.7, 52.1, 17.2. HRMS for $C_{14}H_{22}N_7O_7S$ $[M + H]^+$ calcd, 432.1302; found, 432.1290.

The synthesis and spectroscopic data of the altritol derivatives **10**−**15** can be found in the Supporting Information.

Biological Evaluation−**MLR.** Human peripheral blood mononuclear cells (PBMCs) were isolated from buffy coats, obtained from healthy blood donors by Ficoll (Lymphoprep, Axis-Shield PoC AS, Oslo, Norway) density-gradient centrifugation. The cells at the Ficoll-plasma interface were washed three times and used as "Responder" cells. RPMI 1788 (ATCC, No. CCL-156) cells were treated with mitomycin C (Kyowa, Nycomed, Brussel, Belgium) and used as "Stimulator" cells. Responder cells (0.12×10^6), stimulator cells (0.045×10^6), and compounds (in different concentrations) were cocultured for 6 days in RPMI 1640 medium (BioWhittaker, Lonza, Belgium) supplemented with 10% fetal calf serum, 100 U/mL Geneticin (Gibco, LifeTechnologies, U.K.). Cells were cultured in triplicate in flat-bottomed 96-well microtiter tissue culture plates (TTP, Switzerland). After five days, cells were pulsed with 1 μCi of methyl-^{3}H thymidine (MP Biomedicals, U.S.A.), harvested 18 h later on glass filter paper and counted. Proliferation values were expressed as counts per minute (cpm) and converted to % inhibition with respect to a blank MLR test (identical but without added compound). The IC_{50} was determined from a graph with at least four points, each derived from the mean of 3−6 experiments.

Statistics. Basic data analysis was done in Microsoft Excel 2003 (Microsoft Corporation, Redmond, WA). Two-factorial ANOVA was done using Graphpad Prism 4.03 (Graphpad Software, San Diego, CA). Where applicable, data are represented as the mean ± SD. P values <0.05 were considered statistically significant.

Acknowledgment. We thank Luc Baudemprez for assistance in NMR recording, Roger Busson for help with interpretation of NMR data and helpful discussions, and Jef Rozenski and Stephanie Vandenwaeyenberg for recording of MS spectra.

Supporting Information Available: Synthetic procedures and analytical data for the preparation of compounds **10**−**15**, **17**−**21**, **23**−**26**, and **28**−**32** and for the corresponding intermediate compounds. HPLC purity data of final compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

References

(1) Ochsner, U. A.; Sung, X.; Jarvis, T.; Critchley, I.; Janjic, N. *Expert Opin. Invest. Drugs* **2007**, *16*, 573–593.
(2) Ataide, S. F.; Ibba, M. *Chem. Biol.* **2006**, *1*, 285–297.
(3) Kim, S.; Lee, S. W.; Choi, E. C.; Choi, S. Y. *Appl. Microbiol. Biotechnol.* **2003**, *61*, 278–288.
(4) Schimmel, P.; Tao, J. S.; Hill, J. M. *FASEB J.* **1998**, *12*, 1599–1609.
(5) Tao, J. S.; Schimmel, P. *Expert Opin. Invest. Drugs* **2000**, *9*, 1767–1775.
(6) Hill, J. M. Aminoacyl sulfamides for the treatment of hyperproliferative disorders Cubist Pharmaceuticals, Inc., *U.S. Patent 5,824,657*, 1998.
(7) Laske, R.; Schonenberger, H.; Holler, E. *Arch. Pharm.* **1989**, *322*, 857–862.
(8) Lee, J.; Kim, S. E.; Lee, J. Y.; Kim, S. Y.; Kang, S. U.; Seo, S. H.; Chun, M. W.; Kang, T.; Choi, S. Y.; Kim, H. O. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 1087–1092.
(9) Landeka, I.; Filipic-Rocak, S.; Zinic, B.; Weygand-Durasevic, I. *Biochim. Biophys. Acta* **2000**, *1480*, 160–170.
(10) Bernier, S.; Akochy, P. M.; Lapointe, J.; Chenevert, R. *Bioorg. Med. Chem.* **2005**, *13*, 69–75.
(11) Heacock, D.; Forsyth, C. J.; Shiba, K.; Musier-Forsyth, K. *Bioorg. Chem.* **1996**, *24*, 273–289.
(12) Bernier, S.; Dubois, D. Y.; Habegger-Polomat, C.; Gagnon, L. P.; Lapointe, J.; Chenevert, R. *J. Enzym. Inhib.* **2005**, *20*, 61–67.
(13) Shue, Y.-K.; Zydowksi, T. M. Aminoacyl adenylate mimics as novel antimicrobial and antiparasitic agents Cubist Pharmaceuticals, Inc., *U.S. Patent 5,726,195*, 1998.
(14) Ochsner, U. A.; Young, C. L.; Stone, K. C.; Dean, F. B.; Janjic, N.; Critchley, I. *Antimicrob. Agents Chemother.* **2005**, *49*, 4253–4262.
(15) Muguruma, H.; Yano, S.; Kakiuchi, S.; Uehara, H.; Kawatani, M.; Osada, H.; Sone, S. *Clin. Cancer Res.* **2005**, *11*, 8822–8828.
(16) Woo, J.-T.; Kawatani, M.; Kato, M.; Shinki, T.; Yonezawa, T.; Kanoh, N.; Nakagawa, H.; Takami, M.; Lee, K. H.; Stern, P. H.; Nagai, K.; Osada, H. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 4729–4734.
(17) Ruan, B.; Bovee, M. L.; Sacher, M.; Stathopoulos, C.; Poralla, K.; Francklyn, C. S.; Söll, D. *J. Biol. Chem.* **2005**, *280*, 571–577.
(18) Kawamura, T.; Liu, D.; Towle, M. J.; Kageyama, R.; Tsukahara, N.; Wakabayashi, T.; Littlefield, B. A. *J. Antibiot. (Tokyo)* **2003**, *56*, 709–715.
(19) von der Haar, F.; Gabius, H.-J.; Cramer, F. *Angew. Chem., Int. Ed. Engl.* **1983**, *20*, 217–302.
(20) Budisa, N. *Angew. Chem., Int. Ed.* **2004**, *43*, 6426–6463.
(21) Igloi, G.; von der Haar, F.; Cramer, F. *Biochemistry* **1978**, *17*, 3459–3486.
(22) Lavrik, O. I.; Moor, N. A. *Mol. Biol.* **1984**, *18*, 979–999.
(23) Rabinovitz, A.; Finkleman, R. L.; Reagan, R. L.; Breitman, T. R. *J. Bacteriol.* **1969**, *99*, 336–338.
(24) Castro-pichel, J.; Garcia-lopez, M. T.; De las Heras, F. G. *Tetrahedron* **1987**, *43*, 383–389.
(25) Okada, M.; Iwashita, S.; Koizumi, N. *Tetrahedron Lett.* **2000**, *41*, 7047–7051.

(26) Fleming, J. J.; Fiori, K. W.; Du Bois, J. *J. Am. Chem. Soc.* **2003**, *125*, 2028–2029.
(27) Peterson, E. M.; Brownell, J.; Vince, R. *J. Med. Chem.* **1992**, *35*, 3991–4000.
(28) Ferreras, J. A.; Ryu, J. S.; Di Lello, F.; Tan, D. S.; Quadri, L. E. N. *Nat. Chem. Biol.* **2005**, *1*, 29–32.
(29) Verheggen, I.; Van Aerschot, A.; Toppet, S.; Snoeck, R.; Janssen, G.; Balzarini, J.; De Clercq, E.; Herdewijn, P. *J. Med. Chem.* **1993**, *36*, 2033–2040.
(30) Pankiewicz, K. W.; Krzeminski, J.; Ciszewski, L. A.; Ren, W.-Y.; Watanabe, K. A. *J. Org. Chem.* **1992**, *57*, 553–559.
(31) Allart, B.; Busson, R.; Rozenski, J.; Van Aerschot, A.; Herdewijn, P. *Tetrahedron* **1999**, *55*, 6527–6546.
(32) Changelian, P. S.; Flanagan, M. E.; Ball, D. J.; Kent, C. R.; Magnuson, K. S.; Martin, W. H.; Rizzuti, B. J.; Sawyer, P. S.; Perry, B. D.; Brissette, W. H.; McCurdy, S. P.; Kudlacz, E. M.; Conklyn, M. J.; Elliott, E. A.; Koslov, E. R.; Fisher, M. B.; Strelevitz, T. J.; Yoon, K.; Whipple, D. A.; Sun, J. M.; Munchhof, M. J.; Doty, J. L.; Casavant, J. M.; Blumenkopf, T. A.; Hines, M.; Brown, M. F.; Lillie, B. M.; Subramanyam, C.; Shang-Poa, C.; Milici, A. J.; Beckius, G. E.; Moyer, J. D.; Su, C. Y.; Woodworth, T. G.; Gaweco, A. S.; Beals, C. R.; Littman, B. H.; Fisher, D. A.; Smith, J. F.; Zagouras, P.; Magna, H. A.; Saltarelli, M. J.; Johnson, K. S.; Nelms, L. F.; Des Etages, S. G.; Hayes, L. S.; Kawabata, T. T.; Finco-Kent, D.; Baker, D. L.; Larson, M.; Si, M. S.; Paniagua, R.; Higgins, J.; Holm, B.; Reitz, B.; Zhou, Y. J.; Morris, R. E.; O'Shea, J. J.; Borie, D. C. *Science* **2003**, *302*, 875–878.
(33) Slinker, B. K. *J. Mol. Cell. Cardiol.* **1998**, *30*, 723–731.
(34) Tomasz, J. 2′,3′-Isopropylideneacetals of D-ribonucleosides In *Nucleic Acid Chemistry*; Townsend, L. B., Tipson, R. S., Eds.; John Wiley & Sons: New York, 1978; pp 765−769.
(35) Zhu, X. F.; Williams, H. J.; Scott, A. I. *J. Chem. Soc., Perkin Trans. 1* **2000**, *1*, 2305–2306.

JM8000746