^{15}N-Multilabeled Adenine and Guanine Nucleosides. Syntheses of [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,NH_2-$^{15}N_3$]-Labeled Adenosine, Guanosine, 2′-Deoxyadenosine, and 2′-Deoxyguanosine

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We report a high-yield route to the following specifically ^{15}N- and ^{13}C-multilabeled nucleosides: [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,NH_2-$^{15}N_3$]-adenosine; [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,NH_2-$^{15}N_3$]-guanosine; [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,NH_2-$^{15}N_3$]-2′-deoxyadenosine; [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,-NH_2-$^{15}N_3$]-2′-deoxyguanosine. In each set, the ^{13}C2 atom functions as a "tag" that allows the ^{15}N1 and ^{15}N3 atoms to be unambiguously differentiated from the untagged versions in ^{15}N NMR of RNA or DNA fragments. The key intermediate of this synthetic strategy for both the adenine and guanine nucleosides is [NH_2,$CONH_2$-$^{15}N_2$]-5-amino-4-imidazolecarboxamide. The [2-^{13}C]-label is added through a ring closure using [^{13}C]-sodium ethyl xanthate (NaS^{13}CSOEt). Enzymatic transglycosylation of either multilabeled 6-chloropurine or multilabeled 2-mercaptohypoxanthine and a final reaction with $^{15}NH_3$ give the adenine and guanine nucleosides. This is the first report of a [3-^{15}N]-labeled guanine nucleoside.

Specific ^{15}N-labeling of DNA fragments with single ^{15}N labels demonstrated its value for NMR studies of nucleic acid structure and interactions.[1−11] However, to maximize the information available from a single NMR experiment and a single synthesis, we have recently expanded these studies to include in one nucleoside as many ^{15}N labels as can be unambiguously distinguished.[12−14] Thus, we have reported synthetic methods for triply labeled [1,7,-NH_2-$^{15}N_3$]-adenosine, [1,7,NH_2-$^{15}N_3$]-guanosine, and their deoxy analogues and a variety of doubly labeled versions. Further, for the guanosine series, we have also reported procedures that include a 2-^{13}C label that functions as a tag so that two otherwise identical ^{15}N-multilabeled nucleosides incorporated into DNA or RNA fragments can be differentiated.[13] We now extend this work to a new family of multilabeled adenine and guanine nucleosides, with and without ^{13}C tags, that includes the N3 atom. Here we report the syntheses of [1,3,NH_2-$^{15}N_3$]- and [2-^{13}C-1,3,NH_2-$^{15}N_3$]-labeled adenosine, guanosine, and their deoxy analogues. While singly [3-^{15}N]-labeled adenine has been described previously,[15−18] the work described here is the first report of any [3-^{15}N]-labeled guanine nucleoside. Incorporation of these nucleosides into DNA and RNA fragments will provide valuable information, particularly regarding interactions with minor groove binding agents.

Synthesis of singly [3-^{15}N]-labeled adenine using nitration of 4-bromoimidazole has been reported by several groups.[15−17] Several years ago, we introduced an alternative method based on a milder azo coupling reaction to introduce the ^{15}N, which gave [NH_2-^{15}N]-5-amino-4-imidazolecarboxamide (AICA).[18] The syntheses reported here begin with this same procedure but incorporate a second ^{15}N as part of the amide functionality. Thus, the ethyl diester of the commercially available imidazole-4,5-dicarboxylic acid is brominated with *N*-bromosuccinimide (NBS) to afford **2** (see Scheme 1). Saponification of **2** followed by a diazocoupling with [β-^{15}N]-4-bromobenzenediazonium ion, generated in situ by diazotization of 4-bromoaniline using $Na^{15}NO_2$, gives the singly labeled azo acid **3** in 83% yield. Our previous route to the amide using ethyl chloroformate[18] worked well with excess NH_3 but gives low yields when only a few equivalents of $^{15}NH_3$ are employed (generated in situ with DBU/$^{15}NH_4$-Cl in CH_3CN). However, use of carbodiimides such as dicyclohexylcarbodiimide (DCC) or 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (DEC) gives the doubly labeled azoamide **4** in 93% yield with only 1.25 equiv of $^{15}NH_4$Cl. Concomitant reduction of the bromo

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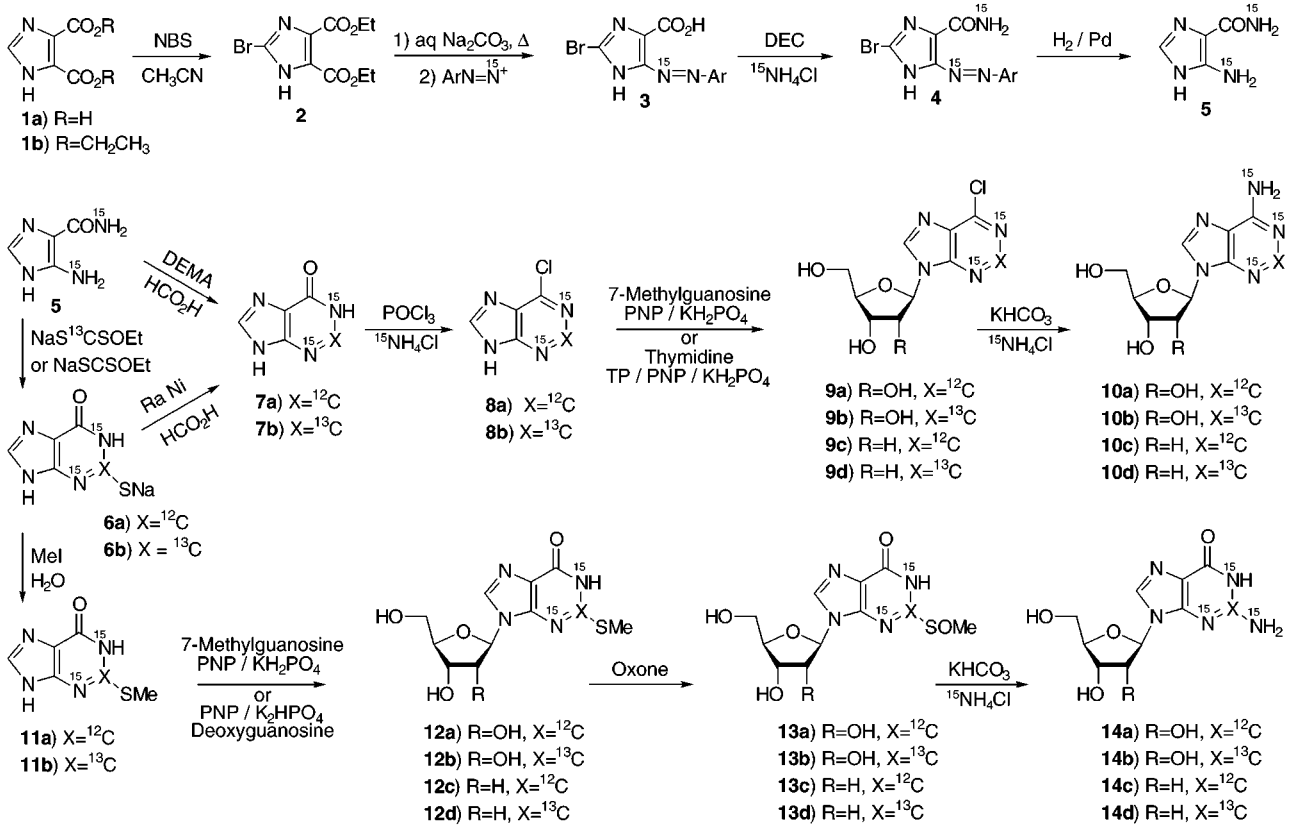
Scheme 1



and azo groups to give the doubly labeled AICA, **5**, is accomplished using excess H_2 with 10% Pd/C in methanol saturated with NH_3. Similar results are obtained using Zn/HCl/Pd/10%C in 2-propanol, but the isolation and purification of **5** by this procedure is more difficult.

Ring closure using diethoxymethyl acetate (DEMA), as we have reported previously,[12,18,19] gives the doubly ^{15}N-labeled hypoxanthine **7a**, from which adenine nucleosides **10a**,**c** are obtained. To introduce the C2 label, and to provide access to guanine nucleosides, we use an alternative ring closure reaction using sodium ethyl xanthate, made from either [^{12}C]- or [^{13}C]-CS_2, to give the mercaptohypoxanthines **6a**,**b**.[20] The latter can be reduced to **7b** using Raney nickel in formic acid. Hypoxanthines **7a**,**b** are then converted to the corresponding 6-chloropurines **8a**,**b**, which readily undergo enzymatic transglycosylation using as sugar donors 7-methylguanosine or thymidine to give **9a**,**b** and **9c**,**d**, respectively. Finally, the third ^{15}N is introduced by chlorine displacement with 2 equiv of $^{15}NH_4Cl/KHCO_3$, to give the four multilabeled adenine nucleosides, **10a**−**d**.

These adenine nucleosides could be converted to the corresponding guanine nucleosides by a five-step rearrangement,[13] but the N1 label would be lost in the process. The mercaptohypoxanthines **6a**,**b** offer an alternate route which preserves this label. Methylation of **6a**,**b** with iodomethane gives the corresponding 2-methylthiohypoxanthines, **11a**,**b**. Enzymatic transglycosylation of **11a**,**b** with purine nucleoside phosphorylase (PNP) in the presence of either 7-methylguanosine or 2′-deoxyguanosine affords the corresponding ribo- or 2′-deoxyribonucleosides, **12a**−**d**. In each case, 10% of the N7 isomer was formed along with the N9 isomer.[21] The regiochemistry observed for PNP couplings normally is exclusively N9, but it has been reported that coupling of the pyrimidopurine M_1G by this method also gives 10% of the N7 isomer.[22] Oxidation of **12a**−**d** to the corresponding sulfoxides **13a**−**d** with Oxone ($2KHSO_5-KHSO_4-K_2SO_4$) in water followed by displacement of the methylsulfoxyl group with [^{15}N]-NH_3 generated from $^{15}NH_4Cl$ in $KHCO_3$ gives the guanine nucleosides **14a**−**d**.

Table 1 shows NMR chemical shifts and coupling constants for the base protons and labeled atoms of the final multilabeled nucleosides **10b**,**d** and **14b**,**d**. The nucleosides without ^{13}C labels had the same ^1H, ^{13}C, and ^{15}N chemical shifts. In the adenine nucleosides **10b**,**d**, the one-bond coupling between H2 and C2 is large (199 Hz) while the smaller two-bond couplings of H2 to N1 and N3 are similar (16 Hz), leading to doublets of apparent triplets for H2. In the amino groups of all four compounds, the one-bond coupling between hydrogen and nitrogen is the normal ~90 Hz. Small (3 Hz) three-bond couplings between the amino hydrogens and the N1 are apparent in the adenine nucleosides but not in the guanine nucleosides.

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(21) The side products we observed were found to return to the free bases **11a**,**b**, upon treatment with acid, or isomerize to the correct nucleosides **12a**−**d**, upon treatment with PNP without a sugar donor. To determine conclusively the identity of the minor products, we prepared singly labeled [7-^{15}N]-2-(methylthio)hypoxanthine by omitting the RaNi desulfurization in a route we have reported previously.[12] The minor product formed upon transglycosylation using this [7-^{15}N]-2-(methylthio)hypoxanthine then was isolated. The N7 linkage was confirmed by the observation of coupling (9 Hz) between the $^{15}N7$ and the C1′.

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Table 1. NMR Chemical Shifts and Coupling Constants for Base Protons and Labeled Atoms for 10b,d and 14b,d[a]

compd	H1	H2	H8	N**H**$_2$	C2[b]	N1[b]	N3[b]	**N**H$_2$[b]
10b		8.12 (dt) 199: H2−C2 16: H2−N1,N3	8.33 (s)	7.36 (dd) 90: H−N 3: H−N1	152.4 (dd) 3: C2−N1/N3 2: C2−N1/N3	236.7 (d) 5: N1−NH$_2$	223.3 (s)	82.9 (d) 4: NH$_2$−N1
10d		8.12 (dt) 199: H2−C2 16: H2−N1,N3	8.32 (s)	7.28 (dd) 90: H−N 3: H−N1	152.4 (br)	236.7 (d) 5: N1−NH$_2$	223.8 (s)	82.5 (d) 4: NH$_2$−N1
14b	10.6 (br)		7.91 (s)	6.53 (d) 90: H−N	154.2 (ddd) 23: C2−NH$_2$ 13: C2−N1 7: C2−N3	152.1 (d) 12: N1−C2	166.8 (t) 6: N3−C2,NH$_2$	74.6 (dd) 23: NH$_2$−C2 4: NH$_2$−N3
14d	11.2 (br)		7.85 (s)	6.70 (d) 88: H−N	155.0 (ddd) 23: C2−NH$_2$ 12: C2−N1 7: C2−N3	157.7 (d) 10: N1−C2	167.1 (t) 6: N3−C2,NH$_2$	75.2 (ddd) 23: NH$_2$−C2 6: NH$_2$−N3 2: NH$_2$−N1

[a] Each entry shows chemical shift (δ) and splitting pattern in parentheses, followed by coupling constants (Hz) and designation of coupled atoms. [b] Proton decoupled.

Table 2. EI Mass Spectral Data for 10a−d[a]

compd	M	M − 30	M − 89	b + 30	b + 2	b + 1	b − 27/28
10a	270 (3)	240 (10)	181 (28)	167 (74)	139 (27)	138 (100)	110 (10)
10b	271 (4)	241 (11)	182 (33)	168 (84)	140 (67)	139 (100)	110 (13)
10c	254 (2)	224 (4)	165 (22)	167 (10)	139 (18)	138 (100)	110 (17)
10d	255 (3)	225 (7)	166 (36)	168 (11)	140 (26)	139 (100)	110 (11)

[a] Entries for significant ions show mass followed by relative abundance in parentheses.

In the proton-decoupled ^{13}C spectra, the one-bond C−N couplings are significant in the guanines (23, 13, and 7 Hz) but are much smaller in adenosine (3 and 2 Hz) and are not resolved in 2′-deoxyadenosine. This difference also occurs in many of the intermediates.

The nitrogen chemical shifts are consistent with literature values.[23] Some couplings among the C2 and the three nitrogens are resolved in the proton-decoupled spectra of these four compounds, while others are not. For example, in adenosine, the largest coupling of N1 is to the amino nitrogen, while, in guanosine, it is to C2. We have previously noted the surprisingly large (23 Hz) coupling between the amino nitrogen and C2 in the guanosines.[13] The ^{1}H, ^{13}C, and ^{15}N NMR spectra for these four compounds are included in the Supporting Information.

Table 2 shows EI MS mass and relative abundance of significant ions for adenine nucleosides **10a**−**d**. The results are consistent with known mass spectral data, which are well-documented.[1,24] The presence of three ^{15}N atoms in **10a** (*m*/*z* 270) and **10c** (*m*/*z* 254) is seen in their molecular ions, which are three units larger than unlabeled adenosine (*m*/*z* 267) and deoxyadenosine (*m*/*z* 251), respectively. The additional ^{13}C atom in **10b** (*m*/*z* 271) and **10d** (*m*/*z* 255) is also evident. Loss of the 5′ group as CH_2O results in ions 30 units smaller than the molecular ions. Fragmentation of the sugar by loss of the 3′, 4′, and 5′ groups gives ions 89 units smaller than the molecular ions, while loss of all but the 1′ group gives ions 30 units larger than the base ions. The base + H ion is known to fragment by successive losses of HCN units.[24] Using singly labeled [1-^{15}N]-deoxyadenosine, we have previously shown conclusively that the first HCN lost includes N1.[1] Not surprisingly, the work reported here demonstrates that this lost HCN also includes C2, since the resulting ion (*m*/*z* 110) is the same in all four cases. The FAB spectra obtained for the guanine nucleosides have as significant ions only the M + 1 and the B + 2 ions (Experimental Section). The presence of three ^{15}N atoms in **14a**,**c** is seen in their molecular ions, M + 1 = 287 and 271, respectively, which are three units larger than unlabeled guanosine (283) and deoxyguanosine (267). The additional ^{13}C atom in **14b** (M + 1 = 288) and **14d** (M + 1 = 272) is also evident. Mass spectra for all eight final products are included in the Supporting Information.

Experimental Section

General Methods. All ^{1}H NMR spectra were acquired at 200 MHz, and ^{13}C NMR spectra, at 50.3 MHz. ^{15}N NMR spectra were acquired at 40.5 MHz, and chemical shifts are reported relative to NH_3 using external 1 M [^{15}N]-urea in DMSO at 25 °C at 77.0 ppm as a reference.[25] Analytical HPLC was carried out with Waters C-18 Nova-Pak cartridges (8 × 100 mm) using a gradient of 2−20% acetonitrile in 0.1 M triethylammonium acetate (TEAA) at a flow rate of 2 mL/min. Preparative reversed-phase HPLC was performed with three Waters Delta-Pak PrepPak cartridges (40 × 100 mm, C_{18} 300 Å, 15 μm) in series at a flow rate of 40 mL/min. UV data were determined from multiwavelength HPLC using a diode array detector and Millennium software.

The [^{15}N]-NH_4Cl and [^{15}N]-$NaNO_2$ were obtained from Isotec Inc., and the [^{13}C]-CS_2 was from Cambridge Isotope Laboratories. Thymidine phosphorylase was obtained from Sigma Chemical Co., and purine nucleoside phosphorylase (PNP) was a gift from Burroughs Wellcome Co. and is also available from commercial sources. General reagents were obtained from Aldrich Chemical Co.

Ethyl Imidazole-4,5-dicarboxylate (1b).[18] A mixture of imidazole-4,5-dicarboxylic acid (**1a**) (97% pure, 24 g, 149 mmol), 1.5 L of absolute ethanol, and 150 mL of H_2SO_4 was refluxed for 2 days under N_2 until a homogeneous solution was obtained. The mixture was concentrated, neutralized with saturated aqueous $NaHCO_3$, and then continuously extracted with CH_2Cl_2. The organic layer was washed twice with 0.1 M NH_4HCO_3, and the combined aqueous layers were backwashed with CH_2Cl_2. The combined organic layers were dried

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over anhydrous $MgSO_4$, filtered, and concentrated to give 26 g (123 mmol, 83%) of pure **1b**: IR 1705 cm^{-1}; UV λ_{max} 257 nm; ^1H NMR ($CDCl_3$) δ 12.10 (br s, 1H), 7.90 (s, 1H), 4.41 (q, J = 7 Hz, 4H), 1.38 (t, J = 7 Hz, 6H); ^{13}C NMR ($CDCl_3$) δ 160.8, 137.7, 129.8, 61.4, 14.1.

Ethyl 2-Bromoimidazole-4,5-dicarboxylate (2).[18] To a mixture of **1b** (26 g, 123 mmol) and 33 g (183 mmol) of *N*-bromosuccinimide (NBS) was added 300 mL of dry acetonitrile under N_2. The resulting solution was stirred in the dark for 24 h and concentrated to dryness. The residue was dissolved in 1.2 L of ethyl acetate, and the solution was washed four times with brine (250 mL), twice with saturated aqueous Na_2SO_3 (100 mL), and twice again with brine (100 mL). The organic layer was dried over $MgSO_4$ and concentrated to dryness. The residue was purified by flash chromatography on silica gel using a gradient of 0−10% CH_3OH in CH_2Cl_2 to give 34 g (117 mmol, 95%) of pure **2**: IR 1750 cm^{-1}; UV λ_{max} 286 ; ^1H NMR ($CDCl_3$) δ 11.00 (br s, 1H), 4.41 (q, J = 7 Hz, 4H), 1.37 (t, J = 7 Hz, 6H); ^{13}C NMR ($CDCl_3$) δ 159.4, 131.8, 120.2, 61.7, 13.9.

[5-^{15}N]-5-((4-Bromophenyl)azo)-2-bromo-4-imidazolecarboxylic Acid (3).[18] A suspension of **2** (25 g, 86 mmol) and Na_2CO_3 (24 g) in 850 mL of water was stirred at 100 °C for 36 h and then cooled to 0 °C. To a cold solution of 4-bromoaniline (97% pure, 13.4 g, 78 mmol) in 78 mL of 10% HCl and 350 mL of water was added dropwise to a cold solution of $Na^{15}NO_2$ (5.7 g, 79 mmol) in 100 mL of water. After 20 min, when the 4-bromoaniline had reacted completely (HPLC), a cold solution of Na_2CO_3 (11 g) in 125 mL of water was added slowly with stirring. This cold mixture was then added slowly to the cold aqueous solution of **2**. After 5 min, the initially yellow solution became red, and a thick precipitate formed. Stirring was continued for 2 h, after which the solid was filtered out and dissolved in 600 mL of aqueous Na_2CO_3 (10 g). A black residue remained that contained no product. The aqueous solution was washed with CH_2Cl_2 (5 × 100 mL) and then acidified with concentrated HCl to afford a yellow precipitate. This precipitate was collected by filtration, washed with cold water (3 × 40 mL), and dried under vacuum over P_2O_5 to give 27 g (71 mmol, 83%) of pure **3**: IR 1720 cm^{-1}; UV λ_{max} 235 nm, 357 nm, λ_{min} 270 nm; ^1H NMR (DMSO-d_6) δ 13.50 (br s, 1H), 7.79 (s, 4H); ^{13}C NMR (DMSO-d_6) 160.3, 151.3 (d, J = 6 Hz), 132.7, 125.2, 124.5 (br s), 123.1; ^{15}N NMR (DMSO-d_6) δ 486 (br s).

[5,CONH$_2$-^{15}N$_2$]-5-((4-Bromophenyl)azo)-2-bromo-4-imidazolecarboxamide (4). To a mixture of **3** (9.5 g, 25 mmol), hydroxybenzotriazole (3.5 g, 25 mmol), and $^{15}NH_4Cl$ (1.8 g, 32 mmol) in a 2 L flask was added 1.4 L of anhydrous CH_3CN under N_2. 1-(3-Dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (DEC) (5.5 g, 29 mmol) was dissolved in 450 mL of anhydrous CH_3CN in a 500 mL round-bottom flask under N_2. Both flasks were kept for 2 h at −15 °C in a freezer after which the DEC solution was cannulated to the 2 L flask kept at −15 °C with stirring. During the transfer, 5.5 mL of DBU (36 mmol) was added. Portions of water (3 × 20 mL) were added after 4, 6, and 8 h, and stirring was continued at −10 °C for 36 h. The solvent was then evaporated, and the resulting orange precipitate was suspended in 400 mL of H_2O and allowed to stand overnight at 4 °C. The orange precipitate was collected by filtration. The filtrate was acidified with concentrated HCl until the pH was 5 and then extracted with CH_2Cl_2 (2 × 25 mL). The orange filter cake was dissolved in 3 L of warm aqueous Na_2CO_3 (60 g), leaving a dark brown residue that was discarded. The red solution was washed with CH_2Cl_2 (3 × 100 mL) and then neutralized with concentrated HCl. The resulting yellow precipitate was filtered out and washed with H_2O (2 × 50 mL). Small amounts of additional product were recovered from the combined CH_2Cl_2 layers by evaporation and solubilization in warm aqueous Na_2CO_3 followed by acidification. The combined products were dried under vacuum over P_2O_5 to afford 8.8 g (24 mmol, 93%) of pure **4**: IR 1663 cm^{-1}; UV λ_{max} 248 nm, 399 nm, λ_{min} 275 nm; ^1H NMR (DMSO-d_6) δ 8.2−7.2 (m); ^{13}C NMR (DMSO-d_6) 159.4 (d, J = 18 Hz), 151.1 (d, J = 5 Hz), 149.1, 132.5, 131.0, 125.0, 124.6, 123.1; ^{15}N NMR (DMSO-d_6) 470, 109; HRMS m/z 372.8957 (calcd for $C_{10}H_7ON_3{}^{15}N_2Br_2$: 372.8958). Anal. Calcd for $C_{10}H_7N_3{}^{15}N_2OBr_2 \cdot 0.25CH_3OH$: C, 32.14; H, 2.11; N, 18.29. Found: C, 32.26; H, 2.09; N, 18.01.

[NH$_2$,CONH$_2$-^{15}N$_2$]-5-Amino-4-imidazolecarboxamide (AICA) (5). To a mixture of 4.4 g (12 mmol) of **4**, 2.5 g of 10% Pd/C, and 0.20 g of 1% Pt/C in a 1 L round-bottom flask was cannulated under N_2 0.95 L of a saturated solution of NH_3(g) in methanol. The resulting mixture was stirred for 10 min at which point H_2 was bubbled through the solution for 6 h from balloons. The mixture was then filtered through a bed of Celite to give a clear, pale yellow solution. After evaporation of the solvent, the residue was dissolved in 40 mL of H_2O, washed with ether (3 × 20 mL), and then applied to a reversed phase preparative column which was eluted with water. Formic acid (2 mL/50 mL HPLC fraction) was added to fractions containing the product. Evaporation of the solvent gave 1.8 g (10 mmol, 88%) of the formate salt of **5**: mp 147−8 °C; UV λ_{max} 230 nm (shoulder), 267 nm; ^1H NMR (DMSO-d_6) δ 8.14 (s, 1H), 7.19 (s, 1H), 6.72 (d, J = 88 Hz, 2H); ^{13}C NMR (DMSO-d_6) 164.7 (d, J = 16 Hz), 163.3, 145.8 (d, J = 16 Hz), 129.6, 107.5; ^{15}N NMR (DMSO-d_6) 98, 50; HRMS m/z 128.0483 (calcd for $C_4H_6ON_2{}^{15}N_2$: 128.0482). Anal. Calcd for $C_4H_6ON_2{}^{15}N_2 \cdot H_2O$: C, 32.87; H, 5.52; N, 38.34. Found: C, 33.08; H, 5.55; N, 38.39.

[^{13}C]-Sodium Ethyl Xanthate (NaS^{13}CSOEt). To a mixture of 2.1 g of NaOH (53 mmol) dissolved by sonication in 200 mL of absolute ethanol was added 4 g (52 mmol) of [^{13}C]-CS_2 (97−99%), and the pale yellow solution was stirred overnight at room temperature. The solvent was evaporated, and the white residue was dried under vacuum to afford 7.6 g (52 mmol, 99%) of [^{13}C]-sodium ethyl xanthate: UV λ_{max} 300 nm; ^1H NMR (DMSO-d_6) δ 4.21 (dq, J_1 = 4 Hz, J_2 = 7 Hz, 2H), 1.16 (t, J = 7 Hz, 3H); ^{13}C NMR (DMSO-d_6) δ 229.8, 66.0, 14.5.

[2-^{13}C-1,3-^{15}N$_2$]-2-Mercaptohypoxanthine (6b). A mixture of 2.6 g of **5** (20 mmol) and 3.4 g (23 mmol) of [^{13}C]-sodium ethyl xanthate in 80 mL of anhydrous *N*,*N*-dimethylformamide was stirred under N_2 at room temperature for 20 min and then refluxed for 4 h. The clear solution turned dark green, and a white precipitate formed. The solution was allowed to cool, and 160 mL of CH_3CN was added. The precipitate was filtered out and washed with CH_3CN (3 × 50 mL) to afford 4.6 g of crude product. The filtrate was evaporated to dryness, and the residue was purified by reversed phase preparative HPLC using aqueous NH_4HCO_3 (pH = 8) to afford an additional 0.3 g of product. The combined products were used without further purification to make **7b** or **11b**. Pure analytical samples were prepared by solubilization in water and precipitation with 10% HCl: mp > 300 °C; UV λ_{max} 280 nm; ^1H NMR (DMSO-d_6) δ 13.60 (br s, 1H), 13.23 (d, J = 96 Hz, 1H), 12.16 (d, J = 92 Hz, 1H), 8.05 (s, 1H); ^{13}C NMR (DMSO-d_6) 173.2 (C2), 154.1 (d, J = 8 Hz), 149.5 (d, J = 16 Hz), 141.2, 111.3 (d, J = 8 Hz); ^{15}N NMR (DMSO-d_6) 178 (d, J = 11 Hz), 148 (d, J = 14 Hz); HRMS m/z 171.0087 (calcd for $C_4{}^{13}CH_4ON_2{}^{15}N_2S$: 171.0080). Anal. Calcd for $C_4{}^{13}CH_4ON_2{}^{15}N_2S \cdot 0.6H_2O$: C, 33.00; H, 2.88; N, 30.79. Found: C, 32.95; H, 2.78; N, 30.64.

[1,3-^{15}N$_2$]-Hypoxanthine (7a). To a mixture of 9.5 g (55 mmol) of the formate salt of **5** and 200 mL of anhydrous DMF under N_2 was added 9 mL of diethoxymethyl acetate (DEMA). The resulting suspension was stirred for 15 min at room temperature at which point 2.25 mL of formic acid was added and the mixture was heated at 135 °C for 4 h. The suspension was concentrated to a gray solid, resuspended in 100 mL of refluxing acetonitrile, and then chilled. The solid was collected by filtration and dried in a vacuum desiccator over P_2O_5 to give 7.0 g (51 mmol, 92%) of pure **7a**: mp > 300 °C; UV λ_{max} 250 nm; ^1H NMR (D_2O/NaOD) δ 8.04 (t, J = 13 Hz, 1H), 7.84 (s, 1H); ^{13}C NMR (D_2O/NaOD) 170.3 (t, J = 4 Hz), 162.9 (d, J = 6 Hz), 153.9, 153.6 (d, J = 2 Hz), 127.1; ^{15}N NMR (D_2O/NaOD) 231, 220; HRMS m/z 138.0330 (calcd for $C_5H_4ON_2{}^{15}N_2$: 138.0326). Anal. Calcd for $C_5H_4ON_2{}^{15}N_2 \cdot 0.125H_2O$: C, 42.79; H, 3.05; N, 40.57. Found: C, 42.65; H, 2.88; N, 40.21.

[2-^{13}C-1,3-^{15}N$_2$]-Hypoxanthine (7b). A stirred mixture of crude **6b** (4.8 g) and 10 g of Raney nickel (50% water suspension) in 250 mL of H_2O was heated at 60 °C for 10 min, and 3.5 mL of formic acid was added. The mixture was refluxed for 30 min, 10 g of EDTA was added, and the mixture was

refluxed for an additional 2 h. The hot reaction mixture was filtered, and the Raney nickel filter cake was washed with boiling water to give a blue solution which was combined with the filtrate, concentrated, and purified by reversed phase preparative HPLC using a gradient of 0−20% CH_3CN in H_2O. Appropriate fractions of **7b** were concentrated to dryness and dried in a vacuum desiccator over P_2O_5 to afford 2.6 g (19 mmol, 95% from **5**) of pure **7b**: mp > 300 °C; UV λ_{max} 250 nm; ^{1}H NMR (D_2O/NaOD) δ 8.04 (dt, J_1 = 13 Hz, J_2 = 195 Hz, 1H), 7.84 (s, 1H); ^{13}C NMR (D_2O/NaOD) 170.4, 163.3 (dd, J_1 = 2 Hz, J_2 = 6 Hz), 154.3 (C2), 152.0, 127.5 (d, J = 7 Hz); ^{15}N NMR (D_2O/NaOD) 228 (d, J = 2 Hz), 220; HRMS *m*/*z* 139.0362 (calcd for $C_4{}^{13}CH_4ON_2{}^{15}N_2$: 139.0359). Anal. Calcd for $C_4{}^{13}CH_4ON_2{}^{15}N_2$: C, 43.17; H, 2.90; N, 40.28. Found: C, 43.11; H, 2.80; N, 40.13.

[1,3-$^{15}N_2$]-6-Chloropurine (8a). To a mixture of **7a** (7.0 g, 51 mmol) and *N*,*N*-dimethylaniline (DMA) (18 mL, 0.14 mol) under N_2 was added $POCl_3$ (175 mL, 1.9 mol). The resulting mixture was heated at 130 °C for 30 min and monitored by HPLC. The solution was then allowed to cool and was concentrated to a black gum. The gum was dissolved in 150 mL of NH_4OH (30%) with cooling (−20 °C). The aqueous solution was filtered through a bed of Celite and washed once with 100 mL of ethyl acetate and then twice with 50 mL of ether. The NH_3 was evaporated, and the residue was dissolved in 80 mL of water, acidified to pH 2 with concentrated HCl (with cooling in an ice bath), and then continuously extracted with ether for 5 days. The ether layer was evaporated, and the residue was dissolved in 20 mL of NH_4OH (30%). This aqueous solution was concentrated to 10 mL and applied to a reversed phase preparative column. Elution with a gradient of 0−10% CH_3CN in water gave product fractions that were concentrated to a white solid and dried in a vacuum desiccator over P_2O_5 to give 7.0 g (45 mmol, 88%) of pure **8a**: mp > 300 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.76 (dd, J_1 = 15 Hz, J_2 = 16 Hz, 1H), 8.71 (s, 1H); ^{13}C NMR (DMSO-d_6) 154.1 (dd, J_1 = 2 Hz, J_2 = 4 Hz), 151.5 (br s), 147.7 (dd, J_1 = 3 Hz, J_2 = 5 Hz), 146.3, 129.3; ^{15}N NMR (DMSO-d_6) 273, 256; HRMS *m*/*z* 155.9980 (calcd for $C_5H_3N_2{}^{15}N_2Cl$: 155.9987). Anal. Calcd for $C_5H_3N_2{}^{15}N_2Cl{\cdot}0.125H_2O$: C, 37.81; H, 2.06; N, 35.28. Found: C, 37.76; H, 1.87; N, 35.70.

[2-^{13}C-1,3-$^{15}N_2$]-6-Chloropurine (8b). The same procedure used to prepare **8a**, except for starting with **7b** instead of **7a**, was used for **8b**: mp > 300 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.73 (dt, J_1 = 15 Hz, J_2 = 209 Hz, 1H), 8.68 (s, 1H); ^{13}C NMR (DMSO-d_6) 154.1, 151.4 (dd, J_1 = 3 Hz, J_2 = 4 Hz, C2), 147.7 (dd, J_1 = 3 Hz, J_2 = 5 Hz), 146.2, 129.3 (dd, J_1 = 3 Hz, J_2 = 12 Hz); ^{15}N NMR (DMSO-d_6) 273 (d, J = 4 Hz), 256; HRMS *m*/*z* 157.0015 (calcd for $C_4{}^{13}CH_3N_2{}^{15}N_2Cl$: 157.0020). Anal. Calcd for $C_4{}^{13}CH_3N_2{}^{15}N_2Cl{\cdot}0.25H_2O$: C, 37.06; H, 2.18; N, 34.58. Found: C, 37.30; H, 1.84; N, 34.79.

7-Methylguanosine. A suspension of guanosine (41 g, 140 mmol) and dimethyl sulfate (28 mL, 296 mmol) in *N*,*N*-dimethylacetamide (350 mL) was stirred at room temperature for 6 h. The pH of the homogeneous solution was adjusted to 10 with NH_4OH (30%), and the solution was poured into 900 mL of chilled acetone (0 °C). The white precipitate was filtered out, suspended in absolute ethanol (400 mL), refiltered, suspended in dry ether (400 mL), and finally filtered again. The white product was dried in a vacuum desiccator over P_2O_5 to give 40 g (130 mmol, 93%) of 7-methylguanosine, which was used without further purification.

[1,3-$^{15}N_2$]-6-Chloro-9-(β-D-*erythro*-pentofuranosyl)purine (9a). To a suspension of **8a** (3.4 g, 22 mmol) and 7-methylguanosine (13 g, 44 mmol) in aqueous K_2HPO_4 (70 mL, 0.02 M) was added 6 N KOH until the pH was 7.4. To this mixture was added purine nucleoside phosphorylase (500 μL, 2.1 units/μL). The mixture was kept at 43 °C with gentle agitation for 7 days. The reaction mixture was then filtered and the filter cake extracted using sonication with DMF (2 × 80 mL) and then H_2O (100 mL). All solutions were combined and concentrated under vacuum just until a white precipitate started to appear (60 mL). Water was added (60 mL), and 40 mL of the warm solution was filtered directly onto a reversed phase preparative column which was eluted with 0−10% CH_3CN in water. Appropriate fractions were combined and concentrated to a white solid, which was dried in a vacuum desiccator over P_2O_5 to give 5.8 g (20.1 mmol, 92%) of pure **9a**: mp 167−8 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.94 (s, 1H), 8.80 (t, J = 15 Hz, 1H), 6.04 (d, J = 5 Hz, 1H), 5.59 (d, J = 5 Hz, 1H), 5.27 (d, J = 5 Hz, 1H), 5.11 (t, J = 5 Hz, 1H), 4.59 (q, J = 5 Hz, 1H), 4.20 (q, J = 4 Hz, 1H), 3.99 (q, J = 4 Hz, 1H), 3.80−3.50 (m, 2 H); ^{13}C NMR (DMSO-d_6) 151.6 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 149.3 (dd, J_1 = 3 Hz, J_2 = 5 Hz), 145.8 (t, J = 9 Hz), 131.4 (t, J = 2 Hz), 88.2, 85.7, 74.0, 70.1, 61.0; ^{15}N NMR (DMSO-d_6) 275, 251; HRMS *m*/*z* 288.0407 (calcd for $C_{10}H_{11}O_4N_2{}^{15}N_2Cl$: 288.0410). Anal. Calcd for $C_{10}H_{11}O_4N_2{}^{15}N_2Cl{\cdot}0.125H_2O$: C, 41.27; H, 3.90; N, 19.94. Found: C, 41.49; H, 3.76; N, 19.63.

[2-^{13}C-1,3-$^{15}N_2$]-6-Chloro-9-(β-D-*erythro*-pentofuranosyl)-purine (9b). The same procedure used to prepare **9a**, except for starting with **8b** rather than **8a**, was used for **9b**: mp 166−168 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.93 (s, 1H), 8.80 (dt, J_1 = 15 Hz, J_2 = 210 Hz, 1H), 6.04 (d, J = 5 Hz, 1H), 5.56 (d, J = 5 Hz, 1H), 5.24 (d, J = 5 Hz, 1H), 5.08 (t, J = 5 Hz, 1H), 4.58 (q, J = 5 Hz, 1H), 4.20 (q, J = 4 Hz, 1H), 4.00 (q, J = 4 Hz, 1H), 3.80−3.50 (m, 2 H); ^{13}C NMR (DMSO-d_6) 151.7 (dd, J_1 = 2 Hz, J_2 = 4 Hz, C2), 149.3 (dd, J_1 = 3 Hz, J_2 = 5 Hz), 145.8, 131.4 (d, J_1 = 2 Hz), 88.3, 85.7, 74.1, 70.1, 61.0; ^{15}N NMR (DMSO-d_6) 275 (d, J = 4 Hz), 251 (d, J = 2 Hz); HRMS *m*/*z* 289.0432 (calcd for $C_9{}^{13}CH_{11}O_4N_2{}^{15}N_2Cl$: 289.0443). Anal. Calcd for $C_9{}^{13}CH_{11}O_4N_2{}^{15}N_2Cl$: C, 41.46; H, 3.83; N, 19.34. Found: C, 41.58; H, 3.70; N, 19.68.

[1,3-$^{15}N_2$]-6-Chloro-9-(2-deoxy-β-D-*erythro*-pentofuranosyl)purine (9c). To a suspension of **8a** (3.5 g, 22 mmol) and thymidine (13 g, 44 mmol) in aqueous K_2HPO_4 (90 mL, 0.02 M) was added 6 N KOH until the pH was 7.0. To this mixture was added purine nucleoside phosphorylase (400 μL, 2.1 units/μL) and thymidine phosphorylase (dThd Pase, 800 units). The mixture was kept at 43 °C with gentle agitation for 3 days. Sodium chloride (15 g) was added, and the reaction mixture was continuously extracted with CH_2Cl_2 for 48 h at which time HPLC analysis indicated that all the **8a** and **9c** had been extracted as well as small amounts of thymidine and thymine. The organic phase (white suspension) was concentrated and purified by reversed phase preparative chromatography using a gradient of 0−15% CH_3CN in water. The fractions containing **8a** (0.26 g, 5% recovered yield) and **9c** (5.1 g, 19 mmol, 83%) were collected separately, concentrated, and then dried in a vacuum desiccator over P_2O_5; mp > 147−8 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.90 (s, 1H), 8.78 (t, J = 16 Hz, 1H), 6.47 (t, J = 6 Hz, 1H), 5.38 (d, J = 4 Hz, 1H), 4.98 (t, J = 5 Hz, 1H), 4.45 (m, 1H), 3.90 (q, J = 3 Hz, 1H), 3.58 (m, 2H), 2.80−2.70 (m, 1H), 2.50−2.40 (m, 1H); ^{13}C NMR (DMSO-d_6) 151.6 (t, J = 4 Hz), 151.3 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 149.2 (dd, J_1 = 3 Hz, J_2 = 4 Hz), 145.8, 131.4 (d, J = 2 Hz), 88.1, 84.2, 70.4, 61.3; ^{15}N NMR (DMSO-d_6) 275, 251; HRMS *m*/*z* 272.0454 (calcd for $C_{10}H_{11}O_3N_2{}^{15}N_2Cl$: 272.0460). Anal. Calcd for $C_{10}H_{11}O_3N_2{}^{15}N_2Cl$: C, 44.05; H, 4.07; N, 20.55. Found: C, 44.00; H, 4.04; N, 20.45.

[2-^{13}C-1,3-$^{15}N_2$]-6-Chloro-9-(2-deoxy-β-D-*erythro*-pentofuranosyl)purine (9d). The same procedure used to prepare **9c**, except for starting with **8b** rather than **8a**, was used for **9d**: mp 146−148 °C; UV λ_{max} 267 nm; ^{1}H NMR (DMSO-d_6) δ 8.88 (s, 1H), 8.78 (dt, J_1 = 15 Hz, J_2 = 210 Hz, 1H), 6.46 (t, J = 6 Hz, 1H), 5.36 (d, J = 4 Hz, 1H), 4.95 (t, J = 5 Hz, 1H), 4.44 (m, 1H), 3.89 (q, J = 3 Hz, 1H), 3.58 (m, 2H), 2.80−2.70 (m, 1H), 2.50−2.40 (m, 1H); ^{13}C NMR (DMSO-d_6) 151.5 (t, J = 3 Hz, C2), 149.1 (d, J = 3 Hz), 145.7, 131.3 (d, J = 11 Hz), 88.1, 84.2, 70.4, 61.3; ^{15}N NMR (DMSO-d_6) 275 (d, J = 4 Hz), 251 (d, J = 2 Hz); HRMS *m*/*z* 273.0483 (calcd for $C_9{}^{13}CH_{11}O_3N_2{}^{15}N_2Cl$: 273.0494). Anal. Calcd for $C_9{}^{13}CH_{11}O_3N_2{}^{15}N_2Cl$: C, 43.89; H, 4.05; N, 20.47. Found: C, 43.74; H, 3.98; N, 20.50.

[1,3,NH$_2$-$^{15}N_3$]-Adenosine (10a). A mixture of **9a** (5.5 g, 19 mmol), [^{15}N]-NH_4Cl (2.2 g, 40 mmol), and $KHCO_3$ (6.1 g, 60 mmol) in DMSO (14 mL) was sealed in a 100 mL flask and kept in an oven at 80 °C for 7 days. The cooled (0 °C) reaction vial was opened carefully, the contents were diluted with 70 mL of water, and the pH was adjusted to 7 with glacial acetic acid. The product was purified by reversed phase preparative

chromatography using a gradient of 0−20% CH_3CN in water. Appropriate fractions were concentrated to dryness, and the solid was dried in a vacuum desiccator over P_2O_5 to afford 4.6 g (17 mmol, 89%) of pure **10a**: mp 233−4 °C; UV λ_{max} 260 nm; ^{1}H NMR (DMSO-d_6) δ 8.33 (s, 1H), 8.12 (t, J = 15 Hz, 1H), 7.34 (dd, J_1 = 2 Hz, J_2 = 90 Hz, 2H), 5.86 (d, J = 5 Hz, 1H), 5.43 (2H), 5.18 (d, J = 4 Hz, 1H), 4.60 (q, J = 5 Hz, 1H), 4.12 (m, 1H), 3.95 (m, 1H), 3.78−3.40 (2H); ^{13}C NMR (DMSO-d_6) 156.2 (dt, J_1 = 2 Hz, J_2 = 16 Hz), 152.4 (br s), 149.1 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 140.0, 119.4, 88.0, 86.0, 73.5, 70.7, 61.8; ^{15}N NMR (DMSO-d_6) 237, 223, 83; HRMS *m*/*z* 270.0873 (calcd for $C_{10}H_{13}O_4N_2{}^{15}N_3$: 270.0879). Anal. Calcd for $C_{10}H_{13}O_4N_2{}^{15}N_3$: C, 44.45; H, 4.85; N, 25.92. Found: C, 44.57; H, 4.91; N, 26.18.

[2-^{13}C-1,3,NH$_2$-^{15}N$_3$]-Adenosine (10b). The same procedure used to prepare **10a**, except for starting with **9b** rather than **9a**, was used for **10b**: mp 233−234 °C; UV λ_{max} 260 nm; ^{1}H NMR (DMSO-d_6) δ 8.33 (s, 1H), 8.12 (dt, J_1 = 16 Hz, J_2 = 199 Hz, 1H), 7.36 (dd, J_1 = 3 Hz, J_2 = 90 Hz, 2H), 5.87 (d, J = 6 Hz, 1H), 5.40 (2H), 5.18 (1H), 4.60 (t, J = 5 Hz, 1H), 4.13 (dd, J_1 = 3 Hz, J_2 = 4 Hz, 1H), 3.96 (q, J = 3 Hz, 1H), 3.78−3.40 (m, 2H); ^{13}C NMR (DMSO-d_6) 156.1 (dt, J_1 = 3 Hz, J_2 = 19 Hz), 152.4 (br s, C2), 149.1 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 140.0, 119.4, 88.0, 86.0, 73.5, 70.7, 61.8; ^{15}N NMR (DMSO-d_6) 237 (d, J = 5 Hz), 223, 83 (d, J = 4 Hz); HRMS *m*/*z* 271.0906 (calcd for $C_9{}^{13}CH_{13}O_4N_2{}^{15}N_3$: 271.0912). Anal. Calcd for $C_9{}^{13}CH_{13}O_4N_2{}^{15}N_3$·0.25 H_2O: C, 43.56; H, 4.94; N, 25.40. Found: C, 43.37; H, 4.83; N, 25.57.

[1,3,NH$_2$-^{15}N$_3$]-2′-Deoxyadenosine (10c). A mixture of **9c** (4.8 g, 18 mmol), [^{15}N]-NH_4Cl (1.93 g, 36 mmol), and $KHCO_3$ (5.5 g, 54 mmol) in DMSO (12 mL) was sealed in a 100 mL flask and kept in an oven at 80 °C for 7 days. The cooled (0 °C) reaction vial was opened carefully, the contents were diluted with 66 mL of water, and the pH was adjusted to 7 with glacial acetic acid. The product was purified by reversed phase preparative chromatography using a gradient of 0−20% CH_3CN in water. Appropriate fractions were concentrated to dryness, and the solid was dried in a vacuum desiccator over P_2O_5 to afford 4.3 g (17 mmol, 94%) of pure **10c**: mp 190−1 °C (dec); UV λ_{max} 260 nm; ^{1}H NMR (DMSO-d_6) δ 8.33 (s, 1H), 8.13 (t, J = 16 Hz, 1H), 7.33 (dd, J_1 = 3 Hz, J_2 = 90 Hz, 2H), 6.34 (t, J = 7 Hz, 1H), 5.31 (d, J = 4 Hz, 1H), 5.26 (t, J = 5 Hz, 1H), 4.40 (m, 1H), 3.88 (m, 1H), 3.72−3.38 (m, 2H), 2.73 (m, 1H), 2.25 (ddd, J_1 = 3 Hz, J_2 = 6 Hz, J_3 = 11 Hz, 1H); ^{13}C NMR (DMSO-d_6) 156.1 (dt, J_1 = 4 Hz, J_2 = 21 Hz), 152.4, 148.9 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 139.6, 119.3, 88.1, 84.0, 71.1, 62.0; ^{15}N NMR (DMSO-d_6) 237, 224, 82; HRMS *m*/*z* 254.0925 (calcd for $C_{10}H_{13}O_3N_2{}^{15}N_3$: 254.0929). Anal. Calcd for $C_{10}H_{13}O_3N_2{}^{15}N_3$·0.875$H_2O$: C, 44.48; H, 5.51; N, 25.94. Found: C, 44.33; H, 5.31; N, 26.19.

[2-^{13}C-1,3,NH$_2$-^{15}N$_3$]-2′-Deoxyadenosine (10d). The same procedure used to prepare **10c**, except for starting with **9d** rather than **9c**, was used for **10d**: mp 191−192 °C; UV λ_{max} 260 nm; ^{1}H NMR (DMSO-d_6) δ 8.32 (s, 1H), 8.12 (dt, J_1 = 16 Hz, J_2 = 199 Hz, 1H), 7.28 (dd, J_1 = 3 Hz, J_2 = 90 Hz, 2H), 6.33 (t, J = 7 Hz, 1H), 5.29 (d, J = 4 Hz, 1H), 5.28 (t, J = 5 Hz, 1H), 4.40 (m, 1H), 3.87 (m, 1H), 3.72−3.38 (2H), 2.73 (m, 1H), 2.25 (ddd, J_1 = 3 Hz, J_2 = 6 Hz, J_3 = 11 Hz, 1H); ^{13}C NMR (DMSO-d_6) 156.1 (d, J = 20 Hz), 152.4 (C2), 148.9 (dd, J_1 = 2 Hz, J_2 = 5 Hz), 139.6, 119.3, 88.1, 84.0, 71.1, 62.0; ^{15}N NMR (DMSO-d_6) 237 (d, J = 5 Hz), 224, 83 (d, J = 4 Hz); HRMS *m*/*z* 255.0952 (calcd for $C_9{}^{13}CH_{13}O_3N_2{}^{15}N_3$: 255.0963). Anal. Calcd for $C_9{}^{13}CH_{13}O_3N_2{}^{15}N_3$·0.5$H_2O$: C, 45.45; H, 5.34 N, 26.51. Found: C, 45.06; H, 5.24; N, 26.46.

Sodium Ethyl Xanthate (NaSCSOEt). The same procedure described above was used except unlabeled CS_2 was employed.

[1,3-^{15}N$_2$]-2-Mercaptohypoxanthine (6a). A mixture of 2.6 g (20 mmol) of [NH_2,$CONH_2$-^{15}N$_2$]-5-amino-4-imidazolecarboxamide (**5**) and 3.4 g (24 mmol) of sodium ethyl xanthate in 80 mL of anhydrous *N*,*N*-dimethylformamide was stirred under N_2 at room temperature for 20 min and then refluxed for 4 h. The clear solution turned dark green, and a white precipitate formed. The solution was allowed to cool, and 160 mL of CH_3CN was added. The precipitate was collected by filtration and was washed with CH_3CN (3 × 50 mL) to afford 4.6 g of crude product. The filtrate was evaporated to dryness, and the residue was purified by reversed phase preparative HPLC using aqueous NH_4HCO_3 (pH = 8) to afford an additional 0.3 g of product. The combined products were used without further purification to make **11a**. Pure analytical samples of **6a** were prepared by solubilization in water and precipitation with 10% HCl: mp > 300 °C; UV λ_{max} 280 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 13.54 (br s, 1H), 13.10 (br s, 1H), 12.19 (d, J = 92 Hz, 1H), 8.06 (s, 1H); ^{13}C NMR (DMSO-d_6) 173.4 (dd, J_1 = 11 Hz, J_2 = 15 Hz), 153.5 (d, J = 9 Hz), 149.1 (d, J = 19 Hz), 141.6, 110.4 (dd, J_1 = 3 Hz, J_2 = 9 Hz); ^{15}N NMR (DMSO-d_6) 178, 148; HRMS *m*/*z* 170.0040 (calcd for $C_5H_4ON_2{}^{15}N_2S$: 170.0047). Anal. Calcd for $C_5H_4ON_2{}^{15}N_2S$·0.66H_2O: C, 32.96; H, 2.95; N, 30.76. Found: C, 33.32; H, 2.62; N, 30.76.

[1,3-^{15}N$_2$]-2-(Methylthio)hypoxanthine (11a). To a solution of 4 g of crude **6a** in 150 mL of water was added 1.4 mL (22 mmol) of iodomethane. The mixture was stirred vigorously in darkness at room temperature for 24 h. A white precipitate formed, and the starting material disappeared. The mixture was concentrated to 50 mL, and the precipitate was collected by filtration. Additional product was obtained from the filtrate by preparative reversed phase HPLC using a gradient of 0−20% CH_3CN in H_2O, giving a total of 3.2 g (17 mmol, 85% from **5**) of pure **11a**: mp 290−1 °C (dec); UV λ_{max} 262 nm; ^{1}H NMR (D_2O/NaOD) δ (ppm) 7.71 (s, 1H), 2.52 (s, 3H); ^{13}C NMR (D_2O) 169.9 (d, J = 4 Hz), 164.2 (m), 152.9, 124.5, 16.4; ^{15}N NMR (D_2O/NaOD) 227, 213; HRMS *m*/*z* 184.0210 (calcd for $C_6H_6ON_2{}^{15}N_2S$: 184.0203). Anal. Calcd for $C_6H_6ON_2{}^{15}N_2S$·0.66H_2O: C, 36.73; H, 3.77; N, 28.56. Found: C, 36.93; H, 3.53; N, 28.30.

[2-^{13}C-1,3-^{15}N$_2$]-2-(Methylthio)hypoxanthine (11b). The same procedure used to prepare **11a**, except for starting with **6b** rather than **6a**, was used to prepare **11b**: ^{1}H NMR (D_2O/NaOD) δ (ppm) 7.72 (s, 1H), 2.53 (d, J = 4 Hz, 3H); ^{13}C NMR (D_2O) 169.8 (d, J = 4 Hz), 168.5, 164.4 (C2), 152.3 (dd, J_1 = 3 Hz, J_2 = 29 Hz, 124.2 (d, J = 5 Hz), 16.3 (d, J = 9 Hz); ^{15}N NMR (D_2O/NaOD) 227, 213; HRMS *m*/*z* 185.0243 (calcd for $C_5{}^{13}CH_6ON_2{}^{15}N_2S$: 185.0237). Anal. Calcd for $C_5{}^{13}CH_6ON_2{}^{15}N_2S$·0.75$H_2O$: C, 36.26; H, 3.81; N, 28.20. Found: C, 36.40; H, 3.63; N, 27.96.

[1,3-^{15}N$_2$]-2-(Methylthio)inosine (12a). To a suspension of **11a** (3.3 g, 18 mmol) and 7-methylguanosine (12 g, 41 mmol) in aqueous K_2HPO_4 (70 mL, 0.02 M) was added 6 N potassium hydroxide until the pH was 7.4. To this mixture was added purine nucleoside phosphorylase (500 μL, 2.1 units/μL). The mixture was kept at 43 °C with gentle agitation for 7 days. The reaction mixture was then filtered and the filter cake extracted by sonication with DMF (2 × 80 mL) and then H_2O (100 mL). All solutions were combined and concentrated under vacuum until a white precipitate first appeared (∼60 mL). Then 60 mL of water was added, and 40 mL portions of the warm solution were filtered directly onto the reversed phase preparative column and eluted with 0−10% CH_3CN in water. Appropriate fractions were combined, concentrated, and dried in a vacuum desiccator over P_2O_5 to give 4.8 g (15 mmol, 83%) of pure **12a**: mp 245−6 °C (dec); UV λ_{max} 263 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 12.6 (br d, J = 83 Hz, 1H), 8.20 (s, 1H), 5.83 (d, J = 6 Hz, 1H), 5.46 (d, J = 6 Hz, 1H), 5.20 (d, J = 4 Hz, 1H), 4.98 (t, J = 5 Hz, 1H), 4.52 (q, J = 5 Hz, 1H), 4.12 (q, J = 4 Hz, 1H), 3.90 (q, J = 4 Hz, 1H), 3.57 (m, 2H), 2.55 (s, 3H); ^{13}C NMR (DMSO-d_6) 157.7 (d, J = 7 Hz), 156.8 (d, J = 9 Hz), 148.5 (d, J = 6 Hz), 138.1 (m), 121.2 (d, J = 6 Hz), 87.3, 85.5, 73.9, 70.3, 61.4, 13.2; ^{15}N NMR (DMSO-d_6) 206, 171; HRMS (FAB) *m*/*z* 317.0706 (calcd for $C_{11}H_{15}O_5N_2{}^{15}N_2S$: 317.0704). Anal. Calcd for $C_{11}H_{14}O_5N_2{}^{15}N_2S$·$H_2O$: C, 39.51; H, 4.82; N, 16.76. Found: C, 39.78; H, 4.77; N, 16.97.

[2-^{13}C-1,3-^{15}N$_2$]-2-(Methylthio)inosine (12b). The same procedure used to prepare **12a**, except for starting with **11b** rather than **11a**, was used to prepare **12b**: ^{1}H NMR (DMSO-d_6) δ (ppm) 12.6 (br s, 1H), 8.20 (s, 1H), 5.82 (d, J = 5 Hz, 1H), 5.44 (br s, 1H), 5.21 (br s, 1H), 4.99 (br s, 1H), 4.53 (t, J = 5 Hz, 1H), 4.12 (t, J = 4 Hz, 1H), 3.90 (q, J = 3 Hz, 1H) 3.57 (m, 2H), 2.54 (d, J = 5 Hz, 3H); ^{13}C NMR (DMSO-d_6) 157.9

(dd, $J_1 = 2$ Hz, $J_2 = 10$ Hz), 148.5 (d, $J = 5$ Hz), 138.1, 121.2 (m), 87.3, 85.5, 73.9, 70.4, 61.4, 13.3; ^{15}N NMR (DMSO-d_6) 206, 173); HRMS (FAB) *m/z* 318.0753 (calcd for $C_{10}{}^{13}CH_{15}O_5N_2{}^{15}N_2S$: 318.0737). Anal. Calcd for $C_{10}{}^{13}CH_{14}O_5N_2{}^{15}N_2S\cdot H_2O$: C, 39.40; H, 4.81; N, 16.71. Found: C, 39.64; H, 4.65, N, 17.01.

[1,3-$^{15}N_2$]-2-(Methylthio)-2′-deoxyinosine (12c). To a suspension of **11a** (2.5 g, 14 mmol) and 2′-deoxyguanosine (4.3 g, 15 mmol) in aqueous K_2HPO_4 (70 mL, 0.02 M) was added 6 N potassium hydroxide until the pH was 8.4. To this mixture was added purine nucleoside phosphorylase (400 μL, 2.1 units/μL). The flask was sealed and the mixture kept at 43 °C with gentle agitation for 2 days. The reaction mixture was then filtered and the filter cake was extracted by sonication and heating with 2% NH_4OH (4 × 50 mL). All solutions were combined and concentrated under vacuum until a white precipitate first formed. The mixture was heated and the warm solution was filtered directly onto the reversed phase preparative column which was then eluted with 0−5% CH_3CN in 0.25 M NH_4HCO_3. Unreacted **11a** eluted with 2′-deoxyguanosine, so fractions containing both products were combined, evaporated, heated again with PNP, and then purified. Final fractions containing pure product were combined and concentrated to a white solid, which was dried in a vacuum desiccator over P_2O_5 to give 3.1 g (10 mmol, 71%) of pure **12c**: mp 245−6 °C (dec); UV λ_{max} nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 8.13 (s, 1H), 6.26 (t, $J = 7$ Hz, 1H), 5.32 (br s, 1H), 4.95 (br s, 1H), 4.38 (m, 1H), 3.83 (m, 1H), 3.52 (m, 2H), 2.67 (m, 1H), 2.53 (s, 3H), 2.26 (ddd, $J_1 = 2$ Hz, $J_2 = 6$ Hz, $J_3 = 12$ Hz, 1H); ^{13}C NMR (DMSO-d_6) 158.2 (dd, $J_1 = 2$ Hz, $J_2 = 9$ Hz), 157.9 (d, $J = 8$ Hz), 148.3 (d, $J = 5$ Hz), 137.5, 121.2 (dd, $J_1 = 2$ Hz, $J_2 = 7$ Hz), 87.8, 83.4, 70.8, 61.7, 13.2 (d, $J = 4$ Hz); ^{15}N NMR (DMSO-d_6) 206, 171; HRMS (FAB) *m/z* 301.0762 (calcd for $C_{11}H_{15}O_4N_2{}^{15}N_2S$: 301.0754). Anal. Calcd for $C_{11}H_{14}O_4N_2{}^{15}N_2S\cdot 0.75H_2O$: C, 42.10; H, 4.98; N, 17.85. Found: C, 41.92; H, 4.92; N, 18.04.

[2-^{13}C-1,3-$^{15}N_2$]-2-(Methylthio)-2′-deoxyinosine (12d). The same procedure used to prepare **12c**, except for starting with **11b** rather than **11a**, was used to prepare **12d**: ^{1}H NMR (DMSO-d_6) δ (ppm) 12.5 (br s, 1H), 8.17 (s, 1H), 6.27 (t, $J = 7$ Hz, 1H), 5.32 (br s, 1H), 4.91 (br s, 1H), 4.38 (br m, 1H), 3.83 (m, 1H), 3.52 (m, 2H), 2.67 (m, 1H), 2.54 (d, $J = 5$ Hz, 3H), 2.26 (ddd, $J_1 = 2$ Hz, $J_2 = 6$ Hz, $J_3 = 12$ Hz, 1H); ^{13}C NMR (DMSO-d_6) 157.6 (dd, $J_1 = 2$ Hz, $J_2 = 10$ Hz, C2), 156.9 (d, $J = 8$ Hz), 148.2 (d, $J = 5$ Hz), 137.9, 121.2 (dd, $J_1 = 2$ Hz, $J_2 = 7$ Hz), 87.8, 83.3, 70.7, 61.7, 13.2 (d, $J = 4$ Hz); ^{15}N NMR (DMSO-d_6) 206, 171; HRMS (FAB) *m/z* 302.0793 (calcd for $C_{10}{}^{13}CH_{15}O_4N_2{}^{15}N_2S$: 302.0788). Anal. Calcd for $C_{10}{}^{13}CH_{14}O_4N_2{}^{15}N_2S\cdot H_2O$: C, 41.37; H, 5.05; N, 17.55. Found: C, 41.21; H, 4.67; N, 17.85.

[1,3-$^{15}N_2$]-2-(Methylsulfoxyl)inosine (13a). To a cold stirred mixture of 4.5 g (14 mmol) of **12a** in 700 mL of H_2O was added dropwise a solution of 3.8 g (7.5 mmol, 1.2 eq) of Oxone ($2KHSO_5-KHSO_4-K_2SO_4$) in 100 mL H_2O over 2 h. The mixture was stirred vigorously for an additional 2 h at 0 °C. The mixture became a clear solution, and Na_2SO_3 (0.26 g, 2 mmol) was added. The solution was concentrated, and the diastereoisomeric mixture was purified by reversed phase preparative HPLC using 0−20% CH_3CN in H_2O to give 4.5 g (13.5 mmol, 96%) of pure **13a**: UV λ_{max} 258 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 8.43 (s, 1H), 5.88 (d, $J = 6$ Hz, 1H), 5.48 (br s, 1H), 5.21 (br s, 1H), 4.97 (br s, 1H), 4.52 (q, $J = 3$ Hz, 1H), 4.14 (t, $J = 4$ Hz, 1H), 3.93 (q, $J = 4$ Hz, 1H) 3.57 (m, 2H), 2.95 (s, 3H); ^{13}C NMR (DMSO-d_6) 160.8 (d, $J = 8$ Hz), 156.9 (d, $J = 8$ Hz), 147.6 (d, $J = 5$ Hz), 140.1, 124.2 (d, $J = 7$ Hz), 87.4, 85.7 (d, $J = 15$ Hz), 74.1, 70.3 (m), 61.3; ^{15}N NMR (DMSO-d_6) 213, 174 (br s); HRMS (FAB) *m/z* 333.0638 (calcd for $C_{11}H_{15}O_6N_2{}^{15}N_2S$: 333.0653). Anal. Calcd for $C_{11}H_{14}O_6N_2{}^{15}N_2S\cdot 0.5H_2O$: C, 38.71; H, 4.43; N, 16.42. Found: C, 38.81; H, 4.49; N, 16.32.

[2-^{13}C-1,3-$^{15}N_2$]-2-(Methylsulfoxyl)inosine (13b). The same procedure used to prepare **13a**, except for starting with **12b** rather than **12a**, was used to prepare **13b**: ^{1}H NMR (DMSO-d_6) δ (ppm) 8.44 (s, 1H), 5.88 (d, $J = 6$ Hz, 1H), 5.53 (br s, 1H), 5.26 (br s, 1H), 5.02 (br s, 1H), 4.53 (br s, 1H), 4.14 (t, $J = 4$ Hz, 1H), 3.94 (q, $J = 3$ Hz, 1H) 3.60 (m, 2H), 2.96 (d, $J = 5$ Hz, 3H); ^{13}C NMR (DMSO-d_6) 160.9 (C2), 157.0 (d, $J = 8$ Hz), 147.6, 139.9, 124.2, 87.4 (d, $J = 4$ Hz), 85.8, 74.0 (d, $J = 8$ Hz), 70.4 (d, $J = 2$ Hz), 61.3; ^{15}N NMR (DMSO-d_6) 213, 176 (br s); HRMS (FAB) *m/z* 334.0688 (calcd for $C_{10}{}^{13}CH_{15}O_6N_2{}^{15}N_2S$: 334.0687). Anal. Calcd for $C_{10}{}^{13}CH_{14}O_6N_2{}^{15}N_2S\cdot H_2O$: C, 37.60; H, 4.59; N, 15.95. Found: C, 37.63; H, 4.46; N, 15.86.

[1,3-$^{15}N_2$]-2-(Methylsulfoxyl)-2′-deoxyinosine (13c). To a cold stirred mixture of 3.5 g (12 mmol) of **12c** in 650 mL of H_2O was added dropwise a cold solution of 4.1 g (6.6 mmol, 1.15 equiv) of Oxone ($2KHSO_5-KHSO_4-K_2SO_4$) in 100 mL of H_2O over 2 h. The mixture was stirred vigorously for an additional 2 h at 0 °C. The mixture became a clear solution, and Na_2SO_3 (0.26 g, 2 mmol) was added. The solution was stirred for 10 min, and the pH was adjusted to 6.7 with $NaHCO_3$. The resulting solution was concentrated, and the diastereoisomeric mixture was purified by reversed phase preparative HPLC using a gradient of 0−20% CH_3CN in H_2O to obtain 3.3 g of nearly pure **13c**, which was used without further purification: UV λ_{max} 258 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 7.99 (s, 1H), 6.25 (dd, $J_1 = 6$ Hz, $J_2 = 8$, 1H), 5.32 (d, $J = 4$ Hz, 1H), 5.04 (br m, 1H), 4.37 (br m, 1H), 3.83 (br m, 1H), 3.52 (br m, 2H), 2.75−2.55 (br m, 1H), 2.68 (s, 3H), 2.19 (ddd, $J_1 = 2$ Hz, $J_2 = 6$ Hz, $J_3 = 13$ Hz, 1H); ^{13}C NMR (DMSO-d_6) 166.6, 166.0, 149.2 (d, $J = 5$ Hz), 137.0, 124.7, 87.8, 83.5, 71.0, 62.0, 48.6, 39.7; ^{15}N NMR (DMSO-d_6) 242 (d, 15 Hz), 204 (d, 13 Hz).

[2-^{13}C-1,3-$^{15}N_2$]-2-(Methylsulfoxyl)-2′-deoxyinosine (13d). The same procedure used to prepare **13c**, except for starting with **12d** rather than **12c**, was used to prepare **13d**: ^{1}H NMR (DMSO-d_6) δ (ppm) 7.99 (s, 1H), 6.25 (dd, $J_1 = 6$ Hz, $J_2 = 8$ Hz, 1H), 5.33 (br s, 1H), 5.06 (br m, 1H), 4.37 (br m, 1H), 3.83 (br m, 1H) 3.52 (br m, 2H), 3.35 (s, 1H), 2.75−2.55 (br m, 1H), 2.68 (d, $J = 4$ Hz, 3H), 2.19 (ddd, $J_1 = 2$ Hz, $J_2 = 6$ Hz, $J_3 = 13$ Hz, 1H); ^{13}C NMR (DMSO-d_6) 166.7, 166.0 (C2), 149.2 (d, $J = 5$ Hz), 137.0, 124.7, 87.8, 83.5, 71.1, 62.0, 39.7; ^{15}N NMR (DMSO-d_6) 242 (d, 15 Hz), 204 (d, 13 Hz).

[1,2,3-$^{15}N_3$]-Guanosine (14a). A mixture of **13a** (3.3 g, 9.9 mmol), [^{15}N]-NH_4Cl (1.7 g, 30 mmol), and $KHCO_3$ (2 g, 20 mmol) in anhydrous DMSO (32 mL) was sealed in a 100 mL vial and was kept at 78 °C for 14 days. The cooled (0 °C) reaction vial was opened carefully, and the contents were concentrated under vacuum to 10 mL, diluted with 90 mL of hot water, and purified in two parts by reversed phase preparative chromatography using a gradient of 0−10% CH_3CN in water. The combined product fractions were concentrated to dryness, and the solid was dried in a vacuum desiccator over P_2O_5 to give 2.3 g (8.0 mmol, 81%) of pure **14a**: mp 248−9 °C (dec); UV λ_{max} 253 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 10.65 (br s, 1H), 7.93 (s, 1H), 6.45 (d, $J = 89$ Hz, 2H), 5.69 (d, $J = 5$ Hz, 1H), 5.38 (br s, 1H), 5.11 (br s, 1H), 5.03 (br s, 1H), 4.38 (br s, 1H), 4.07 (br s, 1H), 3.86 (q, $J = 3$ Hz, 1H), 3.56 (m, 2H); ^{13}C NMR (DMSO-d_6) 156.9 (d, $J = 11$ Hz), 153.7 (ddd, $J_1 = 7$ Hz, $J_2 = 13$ Hz, $J_3 = 23$ Hz), 151.4 (dd, $J_1 = 3$ Hz, $J_2 = 7$ Hz), 135.7 (m), 116.7 (dd, $J_1 = 2$ Hz, $J_2 = 8$ Hz), 86.4, 85.3, 73.7, 70.4, 61.5; ^{15}N NMR (DMSO-d_6) 166 (d, $J = 5$ Hz), 148, 74 (d, $J = 5$ Hz); MS (FAB) *m/z* 287 (61%, M + 1); 155 (100%, b + 2); HRMS (FAB) *m/z* 287.0903 (calcd for $C_{10}H_{14}O_5N_2{}^{15}N_3$: 287.0906). Anal. Calcd for $C_{10}H_{13}O_5N_2{}^{15}N_3\cdot 0.25H_2O$: C, 41.31; H, 4.68; N, 24.09. Found: C, 41.00; H, 4.90; N, 24.21.

[2-^{13}C-1,2,3-$^{15}N_3$]-Guanosine (14b). The same procedure used to prepare **14a**, except for starting with **13b** rather than **13a**, was used to prepare **14b**: ^{1}H NMR (DMSO-d_6) δ (ppm) 10.6 (br s, 1H), 7.91 (s, 1H), 6.53 (d, $J = 90$ Hz, 2H), 5.68 (d, $J = 6$ Hz, 1H), 5.5−4.8 (br m, 3H), 4.40 (t, $J = 5$ Hz, 1H), 4.08 (dd, $J_1 = 3$ Hz, $J_2 = 5$ Hz, 1H), 3.86 (q, $J = 4$ Hz, 1H), 3.57 (m, 2H); ^{13}C NMR (DMSO-d_6) 157.7 (d, $J = 11$ Hz), 154.2 (ddd, $J_1 = 7$ Hz, $J_2 = 13$ Hz, $J_3 = 23$ Hz, C2), 151.4 (dd, $J_1 = 3$ Hz, $J_2 = 7$ Hz), 135.6, 116.8 (dd, $J_1 = 2$ Hz, $J_2 = 7$ Hz), 86.5, 85.3, 73.7, 70.5, 61.5; ^{15}N NMR (DMSO-d_6) 167 (t, $J = 6$ Hz), 152 (d, $J = 12$ Hz), 75 (dd, $J_1 = 4$ Hz, $J_2 = 23$ Hz); MS (FAB) *m/z* 288 (100%, M + 1); 156 (99%, b + 2); HRMS (FAB) *m/z*

288.0941 (calcd for $C_9{}^{13}CH_{14}O_5N_2{}^{15}N_3$: 288.0940). Anal. Calcd for $C_9{}^{13}CH_{13}O_5N_2{}^{15}N_3 \cdot H_2O$: C, 39.35; H, 4.95; N, 22.95. Found: C, 39.56; H, 4.91; N, 23.15.

[1,2,3-$^{15}N_3$]-2′-Deoxyguanosine (14c). A mixture of nearly pure **13c** (3.2 g), [^{15}N]-NH_4Cl (1.7 g, 30 mmol), and $KHCO_3$ (2 g, 20 mmol) in anhydrous DMSO (32 mL) was sealed in a 100 mL vial and was kept at 78 °C for 14 days. The cooled (0 °C) reaction vial was opened carefully, and the contents were concentrated under vacuum to 10 mL, diluted with 90 mL of hot water, and purified in two parts by reversed phase preparative chromatography using a gradient of 0−10% CH_3CN in water. The combined product fractions were concentrated to dryness, and the solid was dried in a vacuum desiccator over P_2O_5 to give 2.3 g (8.5 mmol, 71% from **12c**) of pure **14c**: mp > 300 °C (dec); UV λ_{max} 253 nm; ^{1}H NMR (DMSO-d_6) δ (ppm) 10.6 (br d, J = 68 Hz, 1H), 7.91 (s, 1H), 6.44 (d, J = 89 Hz, 2H), 6.11 (dd, J_1 = 6 Hz, J_2 = 7 Hz, 1H), 5.24 (d, J = 4 Hz, 1H), 4.93 (t, J = 5 Hz, 1H), 4.32 (br m, 1H), 3.79 (br m, 1H), 3.51 (br m, 2H), 2.6−2.4 (br m, 1H), 2.18 (ddd, J_1 = 3 Hz, J_2 = 6 Hz, J_3 = 13 Hz, 1H); ^{13}C NMR (DMSO-d_6) 156.9 (d, J = 11 Hz), 153.7 (ddd, J_1 = 8 Hz, J_2 = 13 Hz, J_3 = 23 Hz), 150.9 (dd, J_1 = 3 Hz, J_2 = 8 Hz), 135.4, 116.7 (dd, J_1 = 2 Hz, J_2 = 8 Hz), 87.6, 82.7, 70.8, 61.8, 39.6; ^{15}N NMR (DMSO-d_6) 167 (d, J = 6 Hz), 148, 74 (d, J = 5 Hz); MS (FAB) m/z 271 (37%, M + 1); 155 (100%, b + 2); HRMS (FAB) m/z 271.0945 (calcd for $C_{10}H_{14}O_4N_2{}^{15}N_3$: 271.0957). Anal. Calcd for $C_{10}H_{13}O_4N_2{}^{15}N_3 \cdot 0.5H_2O$: C, 43.01; H, 5.05; N, 25.08. Found: C, 43.31; H, 5.02; N, 25.29.

[2-^{13}C-1,2,3-$^{15}N_3$]-2′-Deoxyguanosine (14d). The same procedure used to prepare **14c**, except for starting with **13d** rather than **13c**, was used to prepare **14d**: ^{1}H NMR (DMSO-d_6) δ (ppm) 11.2 (br), 7.85 (s, 1H), 6.70 (d, J = 88 Hz, 2H), 6.11 (dd, J_1 = 6 Hz, J_2 = 8 Hz, 1H), 5.7−4.8 (br s, 2H), 4.33 (br m, 1H), 3.80 (br m, 1H), 3.52 (br m, 2H), 2.6−2.4 (m, 1H), 2.17 (ddd, J_1 = 3 Hz, J_2 = 6 Hz, J_3 = 13 Hz, 1H); ^{13}C NMR (DMSO-d_6) 158.9 (d, J = 11 Hz), 155.0 (ddd, J_1 = 7 Hz, J_2 = 12 Hz, J_3 = 23 Hz, C2), 151.0 (dd, J_1 = 3 Hz, J_2 = 7 Hz), 135.1, 116.8 (dd, J_1 = 2 Hz, J_2 = 8 Hz), 87.7, 82.8, 70.9, 61.9, 39.6; ^{15}N NMR (DMSO-d_6) 167 (t, J = 6 Hz), 158 (d, J = 10 Hz), 75 (ddd, J_1 = 2 Hz, J_2 = 6 Hz, J_3 = 23 Hz); MS (FAB) m/z 272 (37%, M + 1); 156 (100%, b + 2); HRMS (FAB) m/z 272.0983 (calcd for $C_9{}^{13}CH_{14}O_4N_2{}^{15}N_3$: 272.0990). Anal. Calcd for $C_9{}^{13}CH_{13}O_4N_2{}^{15}N_3 \cdot H_2O$: C, 41.52; H, 5.23; N, 24.22. Found: C, 41.36; H, 5.15; N, 24.22.

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Supporting Information Available: ^{1}H, ^{13}C, and ^{15}N NMR spectra for **10b**,**d** and **14b**,**d**, as well as mass spectra for **10a**−**d** and **14a**−**d**. This material is available free of charge via the Internet at http://pubs.acs.org.

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