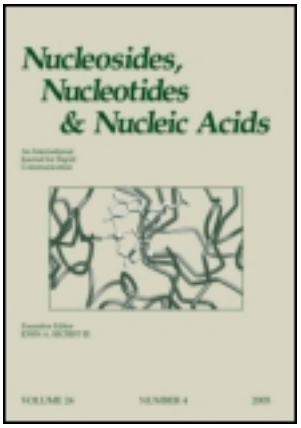

Nucleosides, Nucleotides and Nucleic Acids

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NUCLEOSIDES, NUCLEOTIDES & NUCLEIC ACIDS, 19(7), 1123–1134 (2000)

CONVENIENT SYNTHESES OF 6-METHYLPURINE AND RELATED NUCLEOSIDES

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ABSTRACT: Efficient methods for the synthesis of 6-methylpurine (**3**), 9-(2-deoxy-β-D-*erythro*-pentofuranosyl)-6-methylpurine (**8**), and 6-methyl-9-β-D-ribofuranosylpurine (**5**) are described. Methodology involving the $(Ph_3P)_4Pd$ catalyzed cross-coupling reaction of CH_3ZnBr with several different 6-chloropurine derivatives is described in high yield. This methodology now provides a facile and high-yielding synthesis of **8**, which is needed in significant amounts for studies in cancer gene therapy.

Introduction

For the past few years we have been developing a new approach to the gene therapy of cancer.[1-5] This approach involves the delivery of the gene for the enzyme *E. coli* purine nucleoside phosphorylase (PNP) to a tumor cell. After delivery of the gene, the organism is treated with a non-toxic nucleoside that is specifically susceptible to cleavage by *E. coli* PNP to the base and carbohydrate components. *E. coli* PNP, unlike mammalian PNP, accepts not only 6-oxopurine nucleosides, but also 6-aminopurine nucleosides and related compounds. If the base that is liberated is toxic, and has the proper combination of biological properties, then a specific and selective anticancer effect can be seen. We have had significant success with this approach in several tumor systems in animals, and the toxic bases of most interest to date are 6-methylpurine (6-MeP, **3**)[4,6] and 2-fluoroadenine (2-F-Ade).[4,7] Thus, as a part of that program, we have had to provide large quantities of the non-toxic nucleoside 9-(2-deoxy-β-D-*erythro*-

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pentofuranosyl)-6-methylpurine (6-MePdR, **8**),[8] which has been the most thoroughly studied prodrug, as well as quantities of a variety of other nucleosides containing 6-MeP.

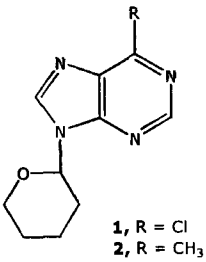
When we first began this research, 6-MeP was commercially available. Several years ago, however, supplies of it were no longer available commercially, and we had to develop a route to it as well as to various nucleosides derived from it. Several routes to certain 6-MeP-containing nucleosides, including 6-MePdR, exist in the literature.[9-13] None of these routes, however, appeared to offer a straightforward and high-yielding approach to this type of compound. The utilization of organozinc chemistry, which has been applied to the synthesis of 6-arylpurines such as 9-benzyl-6-phenylpurine,[14,15] appeared to be appropriate for our purposes. Herein we present the application of this methodology for the synthesis of 6-MeP, 6-MePdR, and 6-methyl-9-β-D-ribofuranosylpurine (6-MePR, **5**). For 6-MePdR, we have included two new synthetic routes from different starting materials. The chemistry described should be applicable to the synthesis of a wide variety of nucleosides containing 6-MeP.

Chemistry

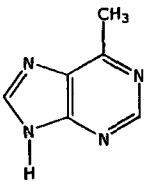
Several syntheses of 6-alkylpurines, including 6-MeP, are available in the literature.[14-18] As we considered possible alternative approaches, it appeared that the use of organozinc reagents, which are stable, safe, and have a tolerance for internally positioned electrophilic functionality, might succeed not only for purines, as had already been found,[14,15] but also for nucleosides. We have discovered that high yields of 6-methylpurine-containing compounds including nucleosides can be obtained by the Pd(0)-catalyzed cross-coupling of an organozinc reagent with a 6-chloropurine derivative.[14]

Synthesis of 6-MeP.

As a key aspect of our gene therapy program, we have needed to be able to prepare a variety of non-standard nucleosides derived from 6-MeP. The availability of large quantities of 6-MeP allows it to be used as a versatile base for coupling to a variety of activated carbohydrates to prepare such nucleosides. Treatment of 9-tetrahydropyranyl-6-chloropurine (**1**)[19] with CH_3ZnBr in the presence of $(Ph_3P)_4Pd$ in tetrahydrofuran produced 6-methyl-9-(tetrahydropyranyl)purine (**2**) in quantitative yield. Removal of the tetrahydropyranyl group with aqueous acid in tetrahydrofuran gave 6-MeP (**3**) in essentially quantitative yield. By this procedure we have been able to readily prepare **3** in multi-gram quantities.



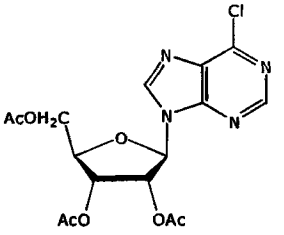
1, R = Cl
2, R = CH_3



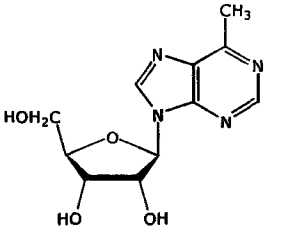
3

Synthesis of 6-MeP-Containing Nucleosides.

The same procedure that was successful on the 9-protected 6-chloropurine derivative above was also effective on 6-chloropurine nucleosides with the carbohydrate hydroxyls protected. For example, treatment of 9-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-6-chloropurine (**4**), readily available from inosine in two steps,[20,21] with CH_3ZnBr under the same conditions produced the corresponding blocked 6-methylpurine nucleoside. Removal of acetyl groups with methanolic ammonia afforded 6-methyl-9-β-D-ribofuranosylpurine (6-MePR, **5**), in nearly quantitative yield.

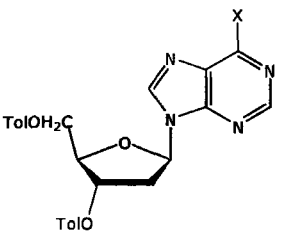


4

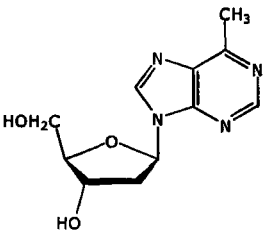


5

Several syntheses of 6-MePdR are available,[8,22-24] though each of them has shortcomings. The organozinc reagent proved to be sufficiently mild that it could even be utilized on a blocked 2′-deoxynucleoside. 6-Chloro-9-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)purine (**6**), is readily prepared from either the commercially available 2′-deoxyinosine[21] or by a coupling reaction between 6-chloropurine and 1-chloro-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose, both also commercially available.[25-28] When **6** is treated with CH_3ZnBr under our standard conditions, it was converted into the 6-methylpurine nucleoside **7** in 92% yield. Removal of the acyl protecting groups with sodium methoxide gave 6-MePdR (**8**) in 84% yield.

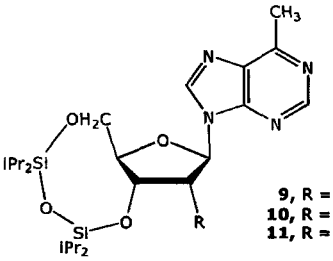


6, X = Cl
7, X = CH_3



8

As a part of our investigations, we also prepared 6-MePdR from 6-MePR by a 2′-deoxygenation procedure.[29] After blocking the *O*-3′ and *O*-5′ hydroxyls of **5** with 1,4-dichloro-1,1,3,3-tetraisopropyldisiloxane to produce **9**, the 2′-hydroxyl of **9** was activated by treatment with thiocarbonyldiimidazole in 1,2-dichloroethane at reflux in the presence of a catalytic amount of DMAP to afford **10**. Reductive cleavage of **10** with triethylsilane and dibenzoyl peroxide in hot toluene gave the blocked 2′-deoxy derivative **11**. Deprotection of the silyl group using tetraethylammonium fluoride provided 6-MePdR (**8**) in 55% yield for the four steps. A similar deoxygenation procedure has been recently reported.[24]



9, R = OH
10, R = C(=S)Im
11, R = H

Summary and Conclusions

The method described herein to prepare 6-MeP and related nucleosides utilizing CH_3ZnBr, which extends the methodology of Gunderson *et al.*,[14,15] offers a simple and high-yielding approach to these compounds. The ability to use this reagent with an appropriately protected ribonucleoside and 2′-deoxyribonucleoside suggests that it will be broadly applicable to nucleosides with a variety of different carbohydrate moieties.

Two different routes to 6-MePdR have recently been published.[22-24] The first route involves the coupling of the sodium salt of 6-MeP with 1-chloro-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranose to produce mainly the 9-β nucleoside, but also

significant amounts of the 9-α and 7β isomers that must be separated from **8**.[22] The other route involves the preparation of 6-MePR followed by its deoxygenation.[24] In considering these routes, along with those mentioned above, it seems clear that our route beginning with 2′-deoxyinosine is the method of choice for the synthesis of 6-MePdR (**8**). It is high-yielding, reproducible on reasonable scale, and provides only the desired isomer. The utilization of a coupling route requires a tedious separation that involves both α/β and 7/9 isomers. The 2′-deoxygenation route is somewhat longer, and utilizes several reagents that are expensive or bring other challenges to the routes, especially on large scale.

In conclusion, we have been able to apply an organozinc-based methodology[14,15] to facilitate the preparation of 6-methylpurine-containing nucleosides. This methodology also succeeds for the introduction of other groups at C-6 of a purine where a new carbon-carbon bond to C-6 is being formed. This research will be presented in subsequent manuscripts.

Experimental

Melting points were determined on a Mel-temp apparatus and are uncorrected. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Nicolet NT 300NB spectrometer operating at 300.635 MHz (^{1}H) or 75.6 MHz (^{13}C). Chemical shifts are expressed in parts per million from tetramethylsilane. The hydrogen-decoupled ^{13}C NMR spectra were assigned by comparison of the J_{CH} values obtained from hydrogen-coupled ^{13}C NMR spectra. When necessary, selective hydrogen decoupling was performed in order to confirm the assignments. Ultraviolet absorption spectra were determined on Perkin-Elmer Lambda 9 spectrometer by dissolving each compound in methanol or water and diluting 10-fold with 0.1 *N* HCl, pH 7 buffer, and 0.1 *N* NaOH. Mass spectra were recorded either on a Varian/MAT 311A double-focusing mass spectrometer in the fast atom bombardment (FAB) mode (glycerol matrix) or by electrospray on a Perkin Elmer SCIEX API3 triple quadrupole spectrometer. HPLC analysis were carried out on a Hewlett-Packard 1100 series liquid chromatograph with a Phenomenex Sphenclone 5 μ ODS (1) column (4.6 mm x 25 cm) with UV monitoring (254 nm). All flash column chromatography used 230-400 mesh silica gel from E. Merck. TLC was done on Analtech precoated (250 μm) silica gel (GF) plates. The compounds **9** and **11** have been previously fully characterized.[24]

6-Methyl-9-(tetrahydro-2-pyranyl)purine (2). A solution of CH_3ZnBr (0.11 mol) was generated by the dropwise addition of $ZnBr_2$ (25 g) in anhydrous THF (100 mL) to a 3 *M* solution of CH_3MgBr (37 mL, 0.11 mol) in THF (100 mL) at –78 °C for 1h. After this solution was allowed to warm to room temperature, a solution of $(Ph_3P)_4Pd$ (3.2 g, 2.7 mmol) in THF (10 mL) was added to it. A solution of **1**[19] (13.23 g, 0.055 mol) in THF (50 mL) was then added at room temperature and the mixture was heated under argon for 2 h at 60 °C. The mixture was cooled down to room temperature and quenched with saturated solution of NH_4Cl. The solvent was concentrated under reduced pressure and the residue was partitioned between $CHCl_3$ and H_2O. The organic phase was dried ($MgSO_4$), evaporated to dryness, and the residue was purified by short flash silica gel column (eluate; 1% MeOH in $CHCl_3$) to give **2** (11.63 g, 97%) as a pale yellow syrup, which was used directly in the next step: HPLC 98.4% (0.01 M $NH_4H_2PO_4$/MeOH, 20 min linear gradient from 10-90%, RT 19.4 min); MS *m/z* 219 $(M+1)^+$, UV λ_{max} (pH 1) 264, λ_{max} (pH 7) 260, λ_{max} (pH 13) 260 nm; ^{1}H NMR ($CDCl_3$) δ 8.89 (1H, s, H-8), 8.4 (1H, s, H-2), 5.80 (1H, dd, H-1′, J = 2.8, J = 10.1 Hz), 4.19 (1H, m, H-5′$_a$), 3.80 (1H, m, H-5′$_b$), 2.87 (3H, s, 6-CH_3), 2.18-1.66 (6H, m, H-2′$_{a,b}$, H-3′$_{a,b}$, and H-4′$_{a,b}$).

6-Methylpurine (3). A solution of **2** (11.6 g, 0.053 mol) in THF (50 mL) and 2 *N* HCl (50 mL) was stirred for 72 h at room temperature. The volatiles were evaporated under reduced pressure and the concentrated aqueous solution was applied to Dowex 50 W (H^+) column. The column was washed with H_2O until no UV absorbing fractions were observed, then eluted with 2.5% NH_4OH. The collected fractions were evaporated and the residue was purified by flash silica gel column (eluate; 20% MeOH in $CHCl_3$) to give **3** (7.52 g, 98%) as a pale yellow solid that was crystallized from MeOH: mp 235-236 °C (lit.[16] 236 °C), HPLC 100% (0.01 M $NH_4H_2PO_4$/MeOH, 40:60, RT 3.4 min); MS *m/z* 135.1 $(M+1)^+$, UV λ_{max} (pH 1) 263, λ_{max} (pH 7) 261, λ_{max} (pH 13) 270 nm; ^{1}H NMR (DMSO-d_6) δ 13.40 (1H, br s, 9-NH), 8.75 (1H, s, H-2), 8.53 (1H, s, H-8), 2.72 (3H, s, 6-CH_3).

6-Methyl-9-(β-D-ribofuranosyl)purine (5). A solution of $(PPh_3)_4Pd$ (0.224 g,.0.21 mmol) in THF (5 mL) was added to a solution of CH_3ZnBr (8.5 mmol, generated as above) in THF (20 mL) at room temperature. A solution of **4** (1.75 g, 4.24 mmol) in THF (10 mL) was added at room temperature and the mixture was stirred for 1 h at 55 °C. After an aqueous work up, the residue obtained by evaporation of the dried organic

phase was dissolved in MeOH saturated with NH_3 (15 mL) and stirred for 4 h at room temperature. The solvent was evaporated and the residue was purified by silica gel chromatography (eluate; 6% EtOH in $CHCl_3$) to give (0.9 g, 95%) of **5**[8] as a colorless solid that was crystallized from hot ethanol: mp 204 °C (lit.[8] 208-209 °C); HPLC 99.9% (0.01 M $NH_4H_2PO_4$/MeOH, 20 min linear gradient from 10-90%, RT 9.2 min); MS *m/z* 267.1 $(M+1)^+$; UV λ_{max} (pH 1) 264, λ_{max} (pH 7) 260, λ_{max} (pH 13) 261 nm; ^{1}H NMR (DMSO-d_6-D_2O) δ. 8.79 (1H, s, H-2), 8.77 (1H, s, H-8), 6.01 (1H, d, H-1′, J = 5.8 Hz), 4.63 (1H, t, H-2′, J = 5.4 Hz), 4.20 (1H, dd, H-3′, J = 3.5, J = 4.8 Hz), 4.02 (1H, dd, H-4′, J = 3.5, J = 7.1 Hz), 3.71 (1H, dd, H-5′a, J = 3.8, J = 12.2 Hz), 3.64 (1H, dd, H-5′a, J = 4.0, J = 12.2 Hz), 2.76 (1H, s, 6-CH_3).

9-(2-Deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)-6-methylpurine (7). 6-Chloro-9-(2-deoxy-3,5-di-*O*-*p*-toluoyl-β-D-*erythro*-pentofuranosyl)purine (**6**,[21,25-28] 1.29 g, 2.8 mmol) in THF (10 mL) was added to a solution of CH_3ZnBr (5.61 mmol, generated as above) and Pd $(PPh_3)_4$ (0.16 g, 0.14 mmol) in THF (20 mL) at room temperature. The mixture was stirred for 4.5 h at 55 °C. Aqueous work up and silica gel chromatography (eluate; 2% EtOH in $CHCl_3$) gave (1.13 g, 92%) of **7**[22] as a colorless foam that was utilized in the next step: MS *m/z* 487 $(M+1)^+$, 493.4 $(M+Li)^+$; UV λ_{max} (pH 1) 245, λ_{max} (pH 7) 245, λ_{max} (pH 13) 235 nm; ^{1}H NMR ($CDCl_3$) δ 8.80 (1H, s, H-8), 8.19 (1H, s, H-2), 7.98 (2H, d, *ortho*-toluoyl, J = 8.1 Hz), 7.90 (2H, d, *ortho*-toluoyl, J = 8.1 Hz), 7.30 (2H, d, *meta*-toluoyl, J = 7.9 Hz), 7.22 (2H, d, *meta*-toluoyl, J = 7.9 Hz), 6.59 (1H, dd, H-1′, J = 5.7, J = 8.1 Hz), 5.84 (1H, dt, H-3′, J = 6.3, J = 2.2 Hz), 4.78 (1H, dd, H-5′$_a$, J = 5.1, J = 13.0 Hz), 4.69-4.63 (2H, m, H-4′ and H-5′$_b$), 3.21 (1H, ddd, H-2′$_a$, J = 6.3, J = 8.4, J = 14.1 Hz), 2.88 (4H, m, H-2′$_b$ and 6-CH_3), 2.42 (3H, s, CH_3- toluoyl), 2.39 (3H, s, CH_3- toluoyl);^{13}C-NMR ($CDCl_3$) δ 166.12 [C(O)], 165.92 [C(O)], 159.62 (C-6), 152.34 (C-2), 150.04 (C-4), 144.52 (C-1 toluoyl), 144.13 (C-1 toluoyl), 141.88 (C-8), 133.64 (C-5), 130.00 (C-2 toluoyl), 129.80 (C-6 toluoyl), 129.62 (C-3 toluoyl), 129.28 (C-5 toluoyl), 126.67 (C-4 toluoyl), 126.41 (C-4 toluoyl), 84.86 (C-1′), 83.09 (C-4′), 75.09 (C-3′), 63.94 (C-5′), 37.89 (C-2′), 21.72 (*P*-CH_3-toluoyl), 21.66 (*P*-CH_3-toluoyl), 19.48 (6-CH_3).

9-(2-Deoxy-β-D-*erythro*-pentofuranosyl)6-methylpurine (8). Deprotection of **7**. After treatment of **7** (1 g, 2.05 mmol) with 1 M MeONa (20 mL) in anhydrous MeOH (20 mL) at 0 °C, the mixture was stirred for 2 h at room temperature and neutralized with

Dowex 50 W (H^+). The resin was filtered off and the filtrate was evaporated *in vacuo*. The residue was purified by flash silica gel column (eluate; 10% MeOH in $CHCl_3$) to give 0.45 g, 84% of **8**[8] as a white solid that was crystallized from acetone: HPLC 99.3% (0.01 M $NH_4H_2PO_4/CH_3CN$, 95:5, RT 10.35 min); mp 153-155 °C (lit.[8] 153 °C); UV λ_{max} (pH 1) 264, λ_{max} (pH 7) 261, λ_{max} (pH 13) 261 nm; ^{1}H NMR (DMSO-d_6) δ 8.78 (1H, s, H-2), 8.72 (1H, s, H-8), 6.46 (1H, t, H-1′, J = 6.9 Hz), 5.36 (1H, d, 3′-OH, J = 4.1 Hz), 5.01 (1H, t, 5′-OH, J = 5.6 Hz), 4.47 (1H, m, H-3′), 3.94 (1H, ddd, H-4′, J = 3.0, J = 4.7, J = 7.6 Hz), 3.62 (1H, m, H-5′b), 3.52 (1H, m, H-5′b); Anal. Calcd. for $C_{11}H_{14}O_3N_4$; C 52.79, H 5.64, N 22.37; found C 52.68, H 5.61, N 22.48. Deprotection of **11**. To a solution of **11** (0.350 g, 0.71 mmol) in CH_3CN (10 mL) at room temperature was added Et_4NF•x H_2O (0.250 g, 1.78 mmol) in one portion. The reaction mixture was stirred for 30 min at room temperature. The solvent was evaporated and the residue was purified over a flash silica gel column (eluate 10 % MeOH in $CHCl_3$) to give (0.163 g, 92%) of **8** as a white solid.

9-[2-*O*-((1-Imidazolyl)thiocarbonyl-3,5-*O*-tetraisopropyldisiloxane-1,3-diyl-β-D-ribofuranosyl)]6-methylpurine (10). A mixture of **9**[24] (0.36 g, 0.76 mmol), 1,1′-thiocarbonyldiimidazole (0.2 g, 1.0 mmol) and DMAP (3.6 mg, 0.03 mmol) in $ClCH_2CH_2Cl$ (10 mL) was heated for 12 h at reflux, by which time the starting material was completely consumed (TLC). The solvent was removed *in vacuo* and the residue was purified by flash silica gel column chromatography (eluate, 30% hexane in ethyl acetate) to give (425 mg, 90%) of **10** as a pale yellow syrup: MS *m/z* 619 $(M+1)^+$, UV λ_{max} (pH 1) 263, λ_{max} (pH 7) 269, λ_{max} (pH 13) 261 nm; ^{1}H NMR ($CDCl_3$) δ 8.80 (1H, s, H-2), 8.79 (1H, s, Im), 8.40 (1H, s, H-8), 8.39 (1H, d, Im, J = 1.1 Hz), 7.09 (1H, dd, Im, J = 0.9, J = 1.8 Hz), 6.45 (1H, dd, H-2′, J = 1, J = 5.6 Hz), 6.19 (1H, d, H-1′, J = 1.1 Hz), 5.52 (1H, dd, H-3′, J = 5.6, J = 8.6 Hz), 4.21-4.06 (3H, m, H-4′, H-5′a, and H-5′b), 2.88 (3H, s, 6-CH_3), 1.25-0.98 (28H, m, *i*-Pr).

9-[2-Deoxy-3,5-*O*-(1,1,3,3-tetraisopropyl)-1,3-disiloxanediyl-β-D-*erythro*-pentofuranosyl]-6-methylpurine (11). To a solution of **10** (0.575 g, 0.93 mmol) in dry toluene (5 mL) was added Et_3SiH (7 mL, 43 mmol) and the mixture was heated at 110 °C. Benzoyl peroxide (0.135 g, 0.55 mmol) in dry dioxane (2 mL) was added to the mixture immediately, and twice more at 30-minute intervals for each addition. The mixture was stirred for a total of 1.5 h at the above temperature, cooled to room

temperature, and the solvent was evaporated under reduced pressure. The residue was purified by flash silica gel column (eluate; cyclohexane : ethyl acetate, 1:2) to give (370 mg, 82%) of **11** as a white solid; HPLC 98% (0.01M $NH_4H_2PO_4$/MeOH, 40:60, RT 3.41 min). Spectral data were in accord with literature values.[24]

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REFERENCES

1. Sorscher, E. J.; Peng, S., Bebok, Z.; Allan, P. W.; Bennett, L. L., Jr; Parker, W. B. Tumor Cell Bystander Killing in Colonic Carcinoma Utilizing the *Escherichia Coli* DeoD Gene to Generate Toxic Purines. *Gene Therapy*, **1994**, *1*, 233-238.
2. Parker, W. B.; King, S. A.; Allan, P. W.; Bennett, L. L., Jr.; Secrist, J. A., III; Montgomery, J. A.; Gilbert, K. S.; Waud, W. R.; Wells, A. H.; Gillespie, G. Y.; Sorscher, E. J. *In Vivo* Gene Therapy of Cancer with *E. coli* Purine Nucleoside Phosphorylase. *Human Gene Therapy*, **1997**, *8*, 1637-1644.
3. Hughes, B. W.; King, S. A.; Allan, P. W.; Parker, W. B.; Sorscher, E. J. Cell to Cell Contact is Not Required for Bystander Cell Killing by *E. coli* Purine Nucleoside Phosphorylase. *J. Biol. Chem.*, **1998**, *273*, 2322-2328.
4. Hughes, B. W.; Wells, A. H.; Bebok, Z.; Gadi, V. K.; Garver, R. I., Jr.; Parker, W. B.; Sorscher, E. J. Bystander Killing of Melanoma Cells Using the Human Tyrosinase Promoter to Express the *Escherichia coli* Purine Nucleoside Phosphorylase Gene. *Cancer Res.*, **1995**, *55*, 3339-3345.
5. Secrist, J. A. III; Parker, W. B.; Allan, P. W.; Bennett, L. L., Jr.; Waud, W. R.; Truss, J. W.; Fowler, A. T.; Montgomery, J. A.; Ealick, S. E.; Wells, A. H.; Gillespie, G. Y.; Gadi, V. K.; Sorscher, E. J. Gene Therapy of Cancer: Activation of Nucleoside Prodrugs with *E. Coli* Purine Nucleoside Phosphorylase. *Nucleosides Nucleotides*, **1999**, *18*, 745-757.
6. Bennett, L. L., Jr.; Vail, M. H.; Chumley, S.; Montgomery, J. A. Activity of

Adenosine Analogs Against a Cell Culture Line Resistant to 2-Fluoroadenine. *Biochem. Pharmacol.*, **1966**, *15*, 1719-1728.

7. a) Philips, F. S.; Sternberg, S. S.; Hamilton, L.; Clarke, D. A. The Toxic Effects of 6-Mercaptopurine and Related Compounds. *Ann. N. Y. Acad. Sci.*, **1954**, *60*, 283. b) Clarke, D. A.; Philips, F. S.; Sternberg, S. S.; Stock, C. C. Effects of 6-Mercaptopurine and Analogs on Experimental Tumors. *ibid.*, **1954**, *60*, 235.
8. Montgomery, J. A.; Hewson, K. Analogs of 6-Methyl-9-β-D-ribofuranosylpurine. *J. Med. Chem.*, **1968**, *11*, 48-52.
9. Yamane, A.; Matsuda, A.; Ueda, T. Reaction of 6-Methylsulfonypurine Riboside with Carbon Nucleophiles and the Synthesis of 6-Alkylpurine Nucleosides. *Chem. Pharm. Bull.*, **1980**, *28*, 150-156.
10. Yamane, A.; Inoue, H.; Ueda, T. Introduction of Carbon Substituents into Pyrimidine and Purine Nucleosides by Sulfur Extrusion. *Chem. Pharm. Bull.*, **1980**, *28*, 157-162.
11. Vorbruggen, M.; Krolikiewicz, K. C-Substitution of Nucleosides with the Aid of the Eschenmoser Sulfide Contraction. *Angew. Chem. Int. Engl.*, **1976**, *15*, 689-690.
12. Evans, B.; Wolfenden, R. A Potential Transition State Analog for Adenosine Deaminase. *J. Amer. Chem. Soc.*, **1970**, *92*, 4751-4752.
13. Van Aerschot, A. A.; Mamos, P.; Weyns, N. J.; Ikeda, S.; De Clercq, E.; Herdewijn, P. A. Antiviral Activity of C-Alkylated Purine Nucleosides Obtained by Cross-Coupling with Tetraalkyltin Reagents. *J. Med. Chem.*, **1993**, *36*, 2938-2942.
14. Gundersen, L.-L.; Bakkestuen, A. K.; Aasen, A. J.; Øverås, H.; Rise, F. 6-Halopurines in Palladium-Catalyzed Coupling with Organotin and Organozinc Reagents. *Tetrahedron*, **1994**, *50*, 9743-9756.
15. Gundersen, L.-L.; Langli, G.; Rise, F. Regioselective Pd-Mediated Coupling between 2,6-Dichloropurines and Organometallic Reagents. *Tetrahedron Lett.*, **1995**, *36*, 1945-1948.
16. Taylor, E. C.; Martin, S. F. A General Method for Alkylation and Alkenylation of Heterocycles. *J. Amer. Chem. Soc.*, **1974**, *96*, 8095-8102.
17. Dvořáková, H.; Dvořák, D.; Holý, A. Coupling of 6-Chloropurines with Organocuprates Derived from Grignard Reagents: A Convenient Route to *sec* and *tert* 6-Alkylpurines. *Tetrahedron Lett.*, **1996**, *37*, 1285-1288.
18. Gundersen, L.-L. 6-Chloropurines and Organostannanes in Palladium Catalyzed Cross Coupling Reactions. *Tetrahedron Lett.*, **1994**, *35*, 3155-3158.

19. Robins, R. K.; Godefroi, E. F.; Taylor, E. C.; Lewis, L. R.; Jackson, A. Purine Nucleosides. I. The Synthesis of Certain 6-Substituted-9-(tetrahydro-2-pyranyl)-purines as Models of Purine Deoxynucleosides. *J. Amer. Chem. Soc.*, **1961**, *83*, 2574-2579.

20. Buck, I. M.; Reese, C. B. An Unambiguous Synthesis of Adenylosuccinic Acid and its Constituent Nucleoside. *J. Chem. Soc. Perkin Trans. 1*, **1990**, 2937-2942.

21. Robins, M. J.; Basom G. L. Nucleic Acid Related Compounds. 8. Direct Conversion of 2′-Deoxyinosine to 6-Chloropurine 2′-Deoxyriboside and Selected 6-Substituted Deoxynucleosides and Their Evaluation As Substrates of Adenosine Deaminase. *Can. J. Chem.*, **1973**, *51*, 3161-3169.

22. Anderson-McKay, J. E.; Both, G. W.; Simpson, G. W. Stereoselective Synthesis of 6-Methyl-9-(2-Deoxy-β-D-*erythro*-pentofuranosyl)purine. *Nucleosides Nucleotides*, **1996**, *15*, 1307-1313.

23. Holý, A.; Vortruba, I. Facile Preparation of Purine and Pyrimidine 2-Deoxy-β-D-ribonucleosides by Biotransformation on Encapsulated Cells. *Nucleic Acids Symp. Ser.*, **1987**, *18*, 69.

24. Boryzki, J. A Novel Approach to Synthesis of 2′-Deoxy-β-D-ribonucleosides *via* Transglycosylation of 6-Oxopurine Ribonucleosides. *Nucleosides Nucleotides*, **1998**, *17*, 1547-1556.

25. Kazimierczuk, Z.; Cottam, H. B.; Revankar, G. R.; Robins, R. K. Synthesis of 2′-Deoxytubercidin, 2′-Deoxyadenosine, and Related 2′-Deoxynucleosides via a Novel Direct Stereospecific Sodium Salt Glycosylation Procedure. *J. Amer. Chem. Soc.*, **1984**, *106*, 6379-6382.

26. Hanna, N. B.; Ramasamy, K.; Robins, R. K.; Revankar, G. R. A Convenient Synthesis of 2′-Deoxy-6-thioguanosine, *Ara*-Guanine, *Ara*-6-Thioguanine and Certain Related Purine Nucleosides by the Stereospecific Sodium Salt Glycosylation Procedure. *J. Heterocycl. Chem.*, **1988**, *25*, 1899-1903.

27. Hildebrand, C.; Wright, G. E. Sodium Salt Glycosylation in the Synthesis of Purine 2′-Deoxyribonucleosides: Studies of Isomer Distribution. *J. Org. Chem.*, **1992**, *57*, 1808-1813.

28. Seela, F.; Winter, H. Synthesis of 6-Substituted Purine N^7-(2-Deoxy-β-D-ribonucleosides) via Anion Glycosylation and Anomerization during the N^7/N^9-Glycosyl Transfer. *Nucleosides Nucleotides*, **1995**, *14*, 129-142.

29. a) Barton, D. H. R.; McCombie, S. W. A New Method for the Deoxygenation of Secondary Alcohols. *J. Chem. Soc. Perkin Trans. 1*, **1975**, 1574-1585. b) Barton, D. H. R.; Jang, D. O.; Jaszberenyi, J. C. Radical Mono- and Dideoxygenations with the Triethylsilane + Benzoyl Peroxide System. *Tetrahedron Lett.*, **1991**, *32*, 7187-7190. c) Barton, D. H. R.; Jang, D. O.; Jaszberenyi, J. C. Tris(trimethylsilyl)silane and Diphenylsilane in the Radical Chain Dideoxygenation of 1,6-Anhydro-D-glucose: A Comparative Study. *Tetrahedron Lett.*, **1992**, *33*, 6629-6632.

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