Briefly, 5×10^4 L1210 and FM3A, or 7.5×10^4 Raji, Molt/4, and MT-4 cells were suspended in growth medium and added to microplate wells in the presence of varying concentrations of the test compounds. The cells were then allowed to proliferate for 48 h (L1210 and FM3A), 72 h (Raji and Molt/4F), or 120 h (MT-4) at 37 °C in a humidified CO_2-controlled atmosphere. At the end of the incubation period, the cells were counted in a Coulter counter (L1210, FM3A, Raji, Molt/4F). Cell viability was determined by the trypan blue dye exclusion method. The IC_{50} value was determined as the concentration of test compound required to inhibit tumor cell proliferation by 50%.

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Registry No. 1, 36413-91-9; 5, 6138-90-5; 6a, 139870-94-3; 6b, 495-02-3; 6c, 57782-96-4; 6d, 35266-82-1; 6e, 40267-83-2; 6f, 57782-99-7; 7a, 139870-95-4; 7b, 30825-46-8; 7c, 139871-03-7; 7d, 139372-91-1; 7e, 139871-10-6; 7f, 139871-14-0; 8, 115-22-0; (±)-9a, 139870-96-5; (±)-9b, 139871-01-5; (±)-9c, 139871-05-9; (±)-9d, 139871-08-2; (±)-9e, 139871-12-8; (±)-9f, 139871-16-2; (±)-10a, 139870-97-6; (±)-10b, 139871-02-6; (±)-10c, 139871-06-0; (±)-10d, 139871-09-3; (±)-10e, 139871-13-9; (±)-10f, 139871-17-3; 11a, 139870-98-7; 11b, 139871-00-4; 11c, 139871-04-8; 11d, 139871-07-1; 11e, 139871-11-7; 11f, 139871-15-1; (±)-12, 31946-92-6; 13, 139870-99-8; (±)-14, 98066-12-7; 2,6-dichlorophenol, 87-65-0; 7-hydroxy-2*H*-1-benzopyran-2-one, 93-35-6; 3-methoxyphenol, 150-19-6; phenol, 108-95-2; 2-naphthol, 135-19-3; 4-methoxyphenol, 150-76-5.

(19) De Clercq, E.; Balzarini, J.; Torrence, P. F.; Mertes, M. P.; Schmidt, C. L.; Shugar, D.; Barr, P. J.; Jones, A. S.; Verhelst, G.; Walker, R. T. Thymidylate synthetase as target enzyme for the inhibitory activity of 5-substituted-2′-deoxyuridines on mouse Leukemia L 1210 cell growth. *Mol. Pharmacol.* 1981, *19*, 321–330.

Synthesis and Anti-HIV Activity of 9-[*c*-4,*t*-5-Bis(hydroxymethyl)cyclopent-2-en-*r*-1-yl]-9*H*-adenine[1]

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The synthesis and in vitro anti-HIV activity of two new racemic nucleoside analogues are described; namely, 9-[*c*-4,*t*-5-bis(hydroxymethyl)cyclopent-2-en-*r*-1-yl]-9*H*-adenine (12) and its guanine analogue 18. While the latter (18) showed no activity, the therapeutic index of the former (12) was 200 and comparable to that (400) of carbovir. One enantiomer of 12 may be viewed as an analogue of carbocyclic oxetanocin and the other as an analogue of carbovir. Hence, these results indicate that one or both of the individual enantiomers of 12 could serve as candidates or lead compounds for the development of anti-AIDS agents.

Introduction

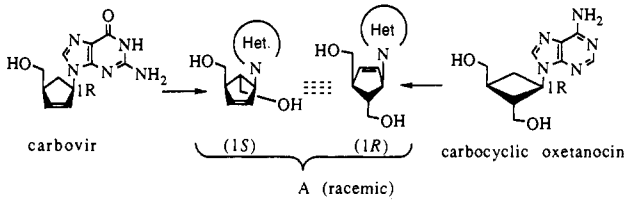
The identification of a retrovirus, referred to as human immunodeficiency virus (HIV), as the etiological agent of human acquired immunodeficiency syndrome (AIDS),[2] has aroused much interest in creating drugs for the treatment of this lethal disease. Although several nucleoside derivatives have been reported to exhibit in vitro anti-HIV activity, to the best of our knowledge, only 3′-azido-3′-deoxythymidine (AZT) and 2′,3′-dideoxyinosine (DDI) have been used clinically. However, clinical studies have indicated substantial toxicities associated with the administration of these two compounds.[3]

In order to create new drugs, we chose as the lead compounds, carbovir[4] and carbocyclic oxetanocin,[5] which were previously reported to exhibit significant anti-HIV activity. The former was pursued as a prospective chemotherapeutic agent against AIDS. From this standpoint, we have chosen the racemic compounds of the 5′-hydroxymethyl derivative of carbovir (A: Het. = 9-adenyl) and its analogue (A: Het. = 9-guanyl) as the target molecules. One enantiomer of A can be regarded as an analogue of carbovir, while the other can be regarded as an analogue of carbocyclic oxetanocin (Scheme I).

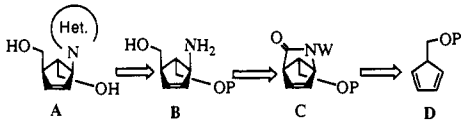
Chemistry

We have previously elaborated the shortest synthetic route to *cis*-4-(hydroxymethyl)cyclopent-2-enylamine from the bicycloamide prepared from the Diels–Alder reactions of cyclopentadiene with either tosyl cyanide[6] or chloro-

Scheme I



Scheme II



W : an electron-withdrawing group
P : a protecting group

† Tohoku University.
‡ Cancer Chemotherapy Centre.

(1) Part XXIII of Synthesis of Nucleosides and Related Compounds. For Part XXII: Katagiri, N.; Nomura, M.; Muto, M.; Kaneko, C. Synthesis of Nucleosides and Related Compounds. XXII. Carbocyclic Analogues of Thymidine and Related Compounds from 2-Azabicyclo[2.2.1]hept-5-en-3-ones. *Chem. Pharm. Bull.* 1991, *39*, 1682–1688.
(2) Popovic, M.; Sarngadharan, M. G.; Read, E.; Gallo, R. C. Detection, Isolation, and Continuous Production of Cytopathic Retroviruses (HTLV-III) from Patients with AIDS and Pre-AIDS. *Science* 1984, *224*, 497–500 and references cited therein.

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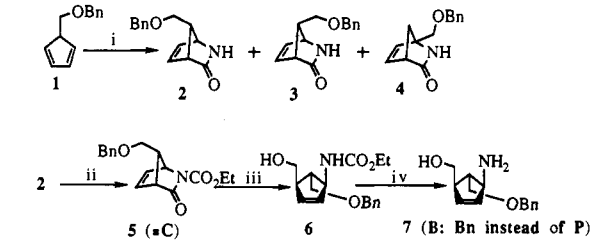
Table I. In Vitro Antiviral Activity of 12 and 18 against HIV-1 in MT-4 Cells[a]

compd	ED_{50}, μg/mL	ID_{50}, μg/mL	TI
12	0.20 ± 0.05	40 ± 7.0	200
18	>100	>100	
DDC	0.011 ± 0.05	27 ± 5.4	2450

[a] MT-4 cells were exposed to HIV-1 ($HTLVIII_B$) at an m. o. i. of 0.002, and cultured for 6 days in the presence of various concentrations of drug. Control cells were treated similarly but not exposed to the virus. Cell proliferation was assessed by the XTT [2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino)-carbonyl]-2*H*-tetrazolium hydroxide] method as described in Weislow et al.[14] The effective dose, 50% (ED_{50}), represents the concentration of compound that increases formazan production in infected cultures to 50% of untreated, uninfected cell controls. The inhibitory dose, 50% (ID_{50}), represents the toxic concentration of drug that reduces formazan production in uninfected cultures to 50% determined by simple linear interpolation from the data. The therapeutic index (TI) was determined by dividing the ID_{50} by the ED_{50}.

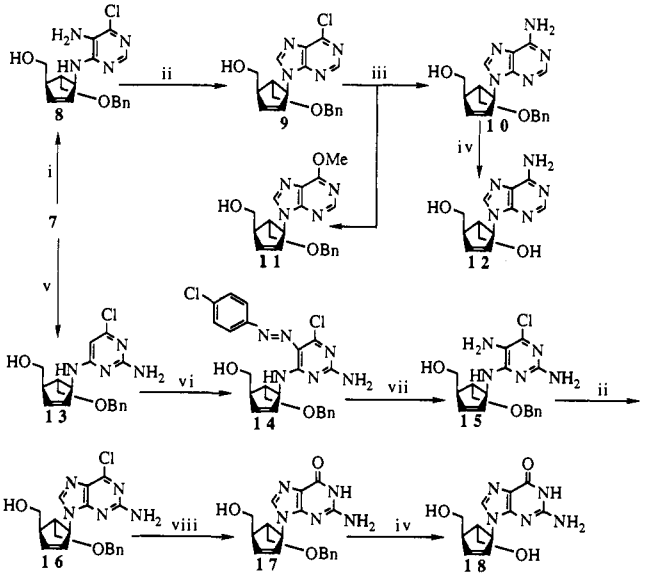
sulfonyl isocyanate[7] through three steps: introduction of an electron-withdrawing group (W) at the nitrogen atom, reductive amide bond cleavage reaction by treatment with sodium borohydride in methanol, and deblocking of the N-substituent (cf. D → C → B in Scheme II).[8]

Scheme III[a]



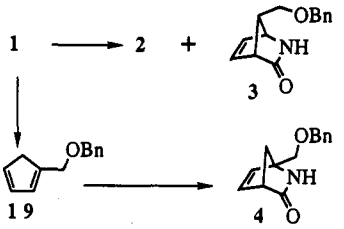
[a] Reagents and conditions: (i) TsCN, ether, room temperature, 12 h, then H_2O, reflux; (ii) lithium diisopropylamide (LDA) (1 equiv), hexamethylphosphoroamide (HMPA), Et_2O, then $ClCO_2Et$ (1 equiv), −78 °C; (iii) sodium borohydride ($NaBH_4$), MeOH; (iv) KOH, aqueous MeOH.

Scheme IV[a]



[a] Reagents and conditions: (i) 5-amino-4,6-dichloropyrimidine, Et_3N, heat; (ii) $(EtO)_3CH$, concentrated HCl; (iii) NH_3, MeOH; (iv) BCl_3, −78 °C; (v) 2-amino-4,6-dichloropyrimidine, Et_3N, heat; (vi) *p*-chlorophenyldiazonium chloride; (vii) Zn/AcOH; (viii) aqueous KOH, MeOH, heat.

Scheme V



Based on the above methodology, the following synthetic scheme for the synthesis of the key precursor *c*-4,*t*-5-bis(hydroxymethyl)-*r*-1-cyclopent-2-enylamine (B: H instead of P) was elaborated. Thus, 5-[(benzyloxy)methyl]cyclopenta-1,3-diene (1)[9] was reacted with tosyl cyanide, and

(3) (a) Richman, D. D.; Fischl, M. A.; Grieco, M. H.; Gottlieb, M. S.; Volberding, P. A.; Laskin, O. L.; Leedom, J. M.; Groopman, J. E.; Mildvan, D.; Hirsch, M. S.; Jackson, G. G.; Durack, D. T.; Phil, D.; Nusinoff-Lehrman, S. The Toxicity of Azidothymidine (AZT) in the Treatment of Patients with AIDS and AIDS-Related Complex. *N. Engl. J. Med.* **1987**, *317*, 192–197. (b) Shirae, H.; Kobayashi, K.; Irie, Y.; Yasuda, N.; Yokozeki, K. Production of 2′,3′-Dideoxyadenosine and 2′,3′-Dideoxyinosine from 2′,3′-Dideoxyuridine and the Corresponding Purine Bases by Resting Cells of Escherichia coli AJ 2595. *Appl. Environ. Microbiol.* **1989**, *55*, 419–424.

(4) Vince, R.; Hua, M. Synthesis and Anti-HIV Activity of Carbocyclic 2′,3′-Didehydro-2′,3′-dideoxy-2,6-Disubstituted Purine Nucleosides. *J. Med. Chem.* **1990**, *33*, 17–21 and references cited therein.

(5) For antiviral activity, see: (a) Norbeck, D. W.; Kern, E.; Hayashi, S.; Rosenbrook, W.; Sham, H.; Herrin, T.; Plattner, J. J.; Clement, R.; Swanson, R.; Shipkowitz, N.; Hardy, D.; Marsh, K.; Arnett, G.; Shannon, W.; Broder, S.; Mitsuya, H. Cyclobut-A and Cyclobut-G: Broad-Spectrum Antiviral Agents with Potential Utility for the Therapy of AIDS. *J. Med. Chem.* **1990**, *33*, 1281–1285. For synthesis, see: (b) Ichikawa, Y.; Narita, A.; Shiozawa, A.; Hayashi, Y.; Narasaka, K. Enantio- and Diastereo-selective Synthesis of Carbocyclic Oxetanocin Analogues. *J. Chem. Soc., Chem. Commun.* **1989**, 1919–1921. (c) Honjo, M.; Maruyama, T.; Sato, T.; Horii, T. Synthesis of the Carbocyclic Analogue of Oxetanocin A. *Chem. Pharm. Bull.* **1989**, *37*, 1413–1415. (d) Slusarchyk, W. A.; Young, M. G.; Bisacchi, G. S.; Hockstein, D. R.; Zahler, R. Synthesis of SQ-33,054, A Novel Cyclobutane Nucleoside with Potent Antiviral Activity. *Tetrahedron Lett.* **1989**, *30*, 6453–6456. (e) Katagiri, N.; Sato, H.; Kaneko, C. Highly Stereoselective Synthesis of Carbocyclic Analogues of Oxetanocin. *Chem. Pharm. Bull.* **1990**, *38*, 228–290. (f) Hsiao, C.; Hannick, S. M. Efficient Synthesis of Protected (2S,3S)-2,3-Bis(hydroxymethyl)cyclobutanone, Key Intermediates for the Synthesis of Chiral Carbocyclic Analogues of Oxetanocin. *Tetrahedron Lett.* **1990**, *46*, 6609–6612.

(6) Jagt, J. C.; van Leusen, A. M. Diels–Alder Cycloadditions or Sulfonyl Cyanides with Cyclopentadiene. Synthesis of 2-Azabicyclo[2.2.1]hepta-2,5-dienes. *J. Org. Chem.* **1974**, *39*, 564–566. See also, Daluge, S.; Vince, R. Synthesis of Carbocyclic Aminonucleosides. *J. Org. Chem.* **1978**, *43*, 2311–2320.

(7) Malpass, J. R.; Tweddle, N. Reaction of Chlorosulphonyl Isocyanate with 1,3-Dienes. Control of 1,2- and 1,4-Addition Pathways and the Synthesis of Aza- and Oxabicyclic Systems. *J. Chem. Soc., Perkin Trans. 1* **1977**, 874–884.

(8) For reviews, see: (a) Katagiri, N. A New Method for the Highly Stereoselective Synthesis of Nucleosides...Creation of New Dienophiles and Use of Reductive Retrograde Aldol Reaction. *Yuki Gosei Kagaku Kyokaishi* **1989**, *47*, 707–721. (b) Kaneko, C.; Katagiri, N.; Nomura, M.; Sato, H. A New Method for the Stereoselective Synthesis of Nucleosides by Means of Sodium Borohydride Mediated C-C or C-N Bond Cleavage Reaction. *Isr. J. Chem.* **1991**, *31*, 247–259.

the products were treated with water to give three bicycloamides (2, 3, and 4) in the respective yields of 25, 12, and 17% (Scheme III). These products could be separated readily by silica gel column chromatography. Their structures were determined by analysis of their NMR spectra as *syn*-2, *anti*-3, and the regioisomeric adducts 4.[10] Thus, due to the anisotropic effect of C–C double bond, the proton at the 7-position of the anti-adduct 3 was observed at higher field (δ 2.92) than that (δ 3.21) of the syn adduct 2. The LDA-assisted ethoxycarbonylation of 2 with ethoxycarbonyl chloride afforded the carbamate 5. The reductive amide bond cleavage reaction of 5 gave the *cis*-1,4-disubstituted cyclopent-2-ene 6 as a sole product. The ethoxycarbonyl group was removed by basic hydrolysis to give the amine 7.

According to the general method,[11] 7 was subjected to reaction with 5-amino-4,6-dichloropyrimidine to give the pyrimidinylamino derivative 8 (Scheme IV). Ring closure with triethyl orthoformate afforded the 9-substituted 6-chloropurine 9. The latter was converted to one of the target molecules (12 = A: Het. = 9-adenyl) by treatment with ammonia in methanol followed by deblocking of the benzyl group with BCl_3 (9 → 10 → 12). The synthesis of the second target molecule (18 = A: Het. = 9-guanyl) was also accomplished in the usual manner.[12] Thus, the condensation of 7 with 2-amino-4,6-dichloropyrimidine followed by treatment with the *p*-chlorobenzenediazonium chloride afforded the 5-[(*p*-chlorophenyl)azo]pyrimidine 14. Reduction of the latter with zinc and acetic acid, followed by ring closure with triethyl orthoformate gave the 9-substituted 2-amino-6-chloropurine 16. Replacement of the chlorine with hydroxyl group by aqueous NaOH and subsequent deblocking of benzyl group with BCl_3 afforded 18.

All reactions except the cycloaddition reaction (i in Scheme III) proceeded in high yields. The low yield (25%) of 2 was due to concomitant formation of the *anti*-3 (12%) and the regioisomeric adducts 4 (17%). The formation of 4 was best explained by assuming a thermally allowed 1,5-hydrogen shift of 1 to 19 (Scheme V). This assumption was verified by the fact that quantitative conversion of 1 to 19 was observed when 1 was kept in THF at room temperature for 1 day (note that the cycloaddition reaction of 1 with tosyl cyanide required 12 h at room temperature).

Biological Evaluation and Discussion

The inhibitory effect of 12 and 18 on HIV-1 induced cytopathogenicity in MT-4 cells is shown in Table I. As a reference compound, 2′,3′-dideoxycytidine (DDC) was also included. DDC has been identified as one of the most potent and selective inhibitors of HIV replication.[13]

Compound 12 showed significant protection of MT-4 cells from the cytopathic effects of HIV-1 (HTLV-III_B). The in vitro anti-HIV activity (defined as 50% of cytopathic effect) was confirmed in several independent experiments. Its therapeutic index (ratio of 50% inhibitory dose, ID_{50}, to 50% antiviral effective dose, ED_{50}) was 200. However, compound 18 showed neither significant in vitro anti-HIV activity nor toxicity at concentrations as high as 100 μg/mL. Replacement of the adenine heterocycle by guanine abolished activity. The lack of activity of this compound may be due to the inability of cellular enzymes to convert it to the triphosphate, or because of poor binding to HIV-1 reverse transcriptase.

Since, as demonstrated recently by Vince and Brownell,[15] only the (−)-isomer (1*R*) of carbovir exhibited anti-HIV activity, it is logical to predict that the anti-HIV activity of 12 is most likely exerted exclusively by the enantiomer (1*S*) that is analogous to the naturally occurring β-D-nucleosides. Quite recently, Bisacchi and his co-workers[16] have reported that only the (1*R*)-enantiomer of carbocyclic oxetanocin shows antiviral activity, and its absolute structure corresponds to oxetanocin itself. Hence, if 12 is regarded as a homomethylene derivative of carbocyclic oxetanocin, only the (1*R*)-isomer should have anti-HIV activity (Scheme I). Therefore, from the standpoint of structure–activity relationship, it is important to clarify if only one or both enantiomers of 12 exhibits potent anti-HIV activity. In order to create a new potential antiretroviral agent in the treatment of AIDS patients, two lines of work are now in progress in our laboratory: 1) The resolution of racemic 12 into its D- and L-enantiomers for the evaluation against HIV and 2) EPC (enantiomerically pure compounds) synthesis of each enantiomer.

Experimental Section

Chemistry. Melting points were determined on a Yanagimoto melting-point apparatus and are uncorrected. ^1H NMR spectra were obtained with a JEOL JNM-PMX 60 or JEOL JNM-GX 500 spectrometer with tetramethylsilane (TMS) as an internal standard. IR spectra were recorded on a JASCO A-102 spectrometer. Electron ionization high-resolution mass spectra (EI-HRMS) were obtained on a JEOL JMS-DX-303 mass spectrometer. Wakogel (C-200) and Merck Kiesel-gel 60F 254 were employed for silica gel column and thin-layer chromatography (TLC), respectively. The ratio of mixtures of solvents for chromatography are shown as volume/volume. Microanalytical results are indicated by atomic symbols and are within ±0.4% of the theoretical values.

Reaction of 5-[(Benzyloxy)methyl]cyclopentadiene (1) with *p*-Toluenesulfonyl Cyanide. To a suspension of cyclopentadienylthallium[9] (13.5 g, 50 mmol) in anhydrous ether (15

(9) (a) Corey, E. J.; Koelliker, U.; Neuffer, J. Methoxymethylation of Thallous Cyclopentadienide. A Simplified Preparation of a Key Intermediate for the Synthesis of Prostaglandins. *J. Am. Chem. Soc.* **1971**, *93*, 1489–1490. (b) Corey, E. J.; Ravindranathan, T.; Terashima, S. A New Method for the 1,4-Addition of the Methylenecarbonyl Unit (–CH_2CO–) to Dienes. *ibid.* **1971**, *93*, 4326–4327. (c) Corey, E. J.; Albonico, S. M.; Koelliker, U.; Schaaf, T. K.; Varma, R. K. New Reagents for Stereoselective Carbonyl Reduction. An Improved Synthetic Route to the Primary Prostaglandins. *ibid* **1971**, *93*, 1491–1493. (d) Corey, E. J.; Ensley, H. E. Preparation of an Optically Active Prostaglandin Intermediate via Asymmetric Induction. *ibid* **1975**, *97*, 6908–6909.

(10) Syn and anti are defined relative to the newly formed C–C double bond.

(11) Shealy, Y. F.; Clayton, J. D. Synthesis of Carbocyclic Analogs of Purine Ribonucleosides. *J. Am. Chem. Soc.* **1969**, *91*, 3075–3083.

(12) Shealy, Y. F.; Clayton, J. D. Carbocyclic Analogs of Guanosine and 8-Azaguanosine. *J. Pharm. Sci.* **1973**, *62*, 1432–1434.

(13) Mitsuya, H.; Broder, S. Inhibition of the in vitro Infectivity and Cytopathic Effect of Human T-Lymphotrophic Virus Type III/Lymphadenopathy-Associated Virus (HTLV-III/LAV) by 2′,3′-Dideoxynucleosides. *Proc. Natl. Acad. Sci. U.S.A.* **1986**, *83*, 1911–1915.

(14) Weislow, O. S.; Kiser, R.; Fine, D. L.; Bader, J.; Shoemaker, R. H.; Boyd, M. R. New Soluble-Formazan Assay for HIV-1 Cytopathic Effects: Application to High-Flux Screening of Synthetic and Natural Products for AIDS-Antiviral Activity. *J. Natl. Cancer Inst.* **1989**, *81*, 577–586.

(15) Vince, R.; Brownell, J. Resolution of Racemic Carvovir and Selective Inhibition of Human Immunodeficiency Virus by the (−) Enantiomer. *Biochem. Biophys. Res. Commun.* **1990**, *168*, 912–916.

(16) Bisacchi, G. S.; Braitman, A.; Cianci, C. W.; Clark, J. M.; Field, A. K.; Hagen, M. E.; Hockstein, D. R.; Malley, M. F.; Mitt, T.; Slusarchyk, W. A.; Sundeen, J. E.; Terry, B. J.; Tuomari, A. V.; Weaver, E. R.; Young, M. G.; Zahler, R. Synthesis and Antiviral Activity of Enantiomeric Forms of Cyclobutyl Nucleoside Analogues. *J. Med. Chem.* **1991**, *34*, 1415–1421.

mL) cooled at −20 °C was added a solution of benzyl chloromethyl ether (7.8 g, 50 mmol) cooled at −20 °C under argon atmosphere, and the mixture was stirred for 7 h. After the temperature of the mixture was raised to 0 °C, the precipitate was removed by filtration and washed thoroughly with ether (100 mL). The filtrate and washing were combined and to this solution was added *p*-toluenesulfonyl cyanide (9.0 g, 50 mmol). The mixture was stirred at room temperature in a vacuum equipment connected to an aspirator with continuous evaporation of the solvent. After concentration to ca. 25 mL, the mixture was kept stirring for 12 h. After dilution of the mixture with 100 mL of chloroform, 10 g of silica gel was added and the whole was stirred for 3 h. The residue obtained after removal of the silica gel followed by concentration in vacuo was subjected to silica gel column chromatography. Elution with a mixture of hexane and ethyl acetate afforded **4** (4:1), **3** (3:1), and **2** (1:1) in yields of 1.5 g (12%), 2.8 g (25%), and 2.0 g (17%), respectively.

1-[(Benzyloxy)methyl]-2-azabicyclo[2.2.1]hept-5-en-3-one (4): colorless oil; EI-HRMS (*m/z*) calcd for $C_{14}H_{15}NO_2$ (M^+) 229.1102, found 229.1098; IR ($CHCl_3$) 3450, 1715 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 2.20 (2 H, m, 7-H), 3.27 (1 H, m, 4-H), 3.83 (2 H, s, CH_2OBn), 4.63 (2 H, s, CH_2Ph), 5.3–5.7 (1 H, br s, NH), 6.71 (2 H, m, 5- and 6-H), 7.37 (5 H, s, Ph).

7-*anti*-[(Benzyloxy)methyl]-2-azabicyclo[2.2.1]hept-5-en-3-one (3): colorless needles, mp 70–75 °C (Et_2O–hexane); EI-HRMS (*m/z*) calcd for $C_{14}H_{15}NO_2$ (M^+) 229.1102, found 229.1128; IR ($CHCl_3$) 3450, 1710 cm^{-1}; ^1H NMR ($CDCl_3$, 500 MHz) δ 2.92 (1 H, m, 7-H), 3.08 (1 H, br s, 4-H), 3.49 (1 H, dd, *J* = 10 and 6 Hz, C*H*HOBn), 3.69 (1 H, dd, *J* = 10 and 9 Hz, CH*H*OBn), 4.25 (1 H, m, 1-H), 4.50 (2 H, s, CH_2Ph), 5.18 (1 H, br s, NH), 6.71 (1 H, m, 5-H), 6.85 (1 H, dd, *J* = 6 and 2 Hz, 6-H), 7.30 (5 H, m, Ph).

7-*syn*-[(Benzyloxy)methyl]-2-azabicyclo[2.2.1]hept-5-en-3-one (2): colorless oil; EI-HRMS (*m/z*) calcd for $C_{14}H_{15}NO_2$ (M^+) 229.1102, found 229.1106; IR ($CHCl_3$) 3450, 1710 cm^{-1}; ^1H NMR ($CDCl_3$, 500 MHz) δ 3.08 (1 H, br s, 4-H), 3.21 (1 H, m, 7-H), 3.39 (1 H, dd, *J* = 10 and 8 Hz, C*H*HOBn), 3.43 (1 H, dd, *J* = 10 and 7 Hz, CH*H*OBn), 4.27 (1 H, m, 1-H), 4.44 (2 H, s, CH_2Ph), 6.21 (1 H, br s, NH), 6.49 (1 H, m, 5-H), 6.85 (1 H, dd, *J* = 5 and 2 Hz, 6-H), 7.30 (5 H, s, Ph).

7-*syn*-[(Benzyloxy)methyl]-2-(ethoxycarbonyl)-2-azabicyclo[2.2.1]hept-5-en-3-one (5). A solution of **2** (1.26 g, 5.52 mmol) in anhydrous tetrahydrofuran (10 mL) was added dropwise to a mixture of 10% LDA–hexane suspension (10 mL) and anhydrous tetrahydrofuran under argon atmosphere with stirring at −78 °C. After being stirred at −78 °C for 1 h, ethyl chloroformate (0.70 mL, 7.34 mmol) was added to the mixture, and the reaction temperature was raised gradually to room temperature. After being kept for 12 h, the solvent was evaporated in vacuo. The residue was subjected to silica gel (60 g) column chromatography. Elution with hexane–ethyl acetate (3:1) gave 983 mg (59%) of colorless oil of **5**: EI-HRMS (*m/z*) calcd for $C_{13}H_{14}O$ ($M^+ - C_4H_5NO_3$) 186.1044, found 186.1027; IR ($CHCl_3$) 1790, 1761, 1710 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.32 (3 H, t, *J* = 7 Hz, CH_2CH_3), 3.13 (1 H, m, 7-H), 3.28 (1 H, m, 4-H), 3.2–3.7 (2 H, m, CH_2OBn), 4.26 (2 H, q, *J* = 7 Hz, CH_2CH_3), 4.46 (2 H, s, CH_2Ph), 5.00 (1 H, m, 1-H), 6.48 (1 H, m, 5-H), 6.73 (1 H, dd, *J* = 6 and 3 Hz, 6-H), 7.31 (5 H, s, Ph).

Ethyl (±)-*N*-[5α-[(Benzyloxy)methyl]-4β-(hydroxymethyl)cyclopent-2-en-1β-yl]carbamate (6). To a stirred solution of **5** (1.132 g, 3.76 mmol) in absolute methanol (25 mL) was added under ice-cooling finely powdered $NaBH_4$ (714 mg, 18.80 mmol) portionwise. After stirring the mixture for 1 h at room temperature, the excess of the reducing reagent was destroyed by addition of AcOH–MeOH (1:1), and the solvent was evaporated under a reduced pressure. The residue was chromatographed on silica gel. Elution with hexane–AcOEt afforded **6** in 83% yield (954 mg), colorless oil: EI-HRMS (*m/z*) calcd for $C_{17}H_{24}NO_4$ (M^+ + 1) 306.1705, found 306.1754; IR ($CHCl_3$) 3450, 1715 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 1.23 (3 H, t, *J* = 7 Hz, CH_2CH_3), 2.3–3.0 (2 H, m, 4- and 5-H), 3.2–3.9 (5 H, m, CH_2OBn, OH and CH_2OH), 4.11 (2 H, q, *J* = 7 Hz, CH_2CH_3), 4.3–4.5 (1 H, m, 1 H), 4.57 (2 H, s, CH_2Ph), 4.8–5.2 (1 H, br d, *J* = 10 Hz, NH), 5.67 (2 H, s, 2- and 3-H), 7.32 (5 H, s, Ph).

(±)-5α-[(Benzyloxy)methyl]-4β-(hydroxymethyl)cyclopent-2-en-1β-ylamine (7). To a solution of **6** (510 mg) in MeOH (10 mL) was added 10 N aqueous KOH (10 mL), and the mixture was refluxed for 20 h. After evaporation of the solvent in vacuo, the product was extracted with ethyl acetate and dried over $MgSO_4$. The residue obtained after filtration and concentration was subjected to silica gel column chromatography. Elution with AcOEt–MeOH (1:1) gave 336 mg (86%) of product as colorless oil: EI-HRMS (*m/z*) calcd for $C_{14}H_{19}NO_2$ ($M^+ - 1$) 232.1338, found 232.1333; IR ($CHCl_3$) 3380 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 3.3–3.9 (5-H, m, CH_2OH, CH_2OBn, and 4-H), 4.56 (2 H, s, CH_2Ph), 5.67 (2 H, s, 2- and 3-H), 7.33 (5 H, s, Ph).

(±)-5-Amino-4-[[5′α-[(benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]amino]-6-chloropyrimidine (8). To a solution of **7** (424 mg, 1.81 mmol) in EtOH (20 mL) was added 5-amino-4,6-dichloropyrimidine (596 mg, 3.63 mmol) and triethylamine (550 mg, 5.51 mmol). The mixture was heated at 100 °C in a sealed tube for 48 h. After evaporation of the solvent in vacuo, the residue was chromatographed on silica gel. Elution with hexane–ethyl acetate (1:1) gave 536 mg (82%) of the product: colorless oil; EI-HRMS (*m/z*) calcd for $C_{18}H_{21}ClN_4O_2$ (M^+), $C_{18}H_{21}ClN_4O_2$ (M^+ + 2) 360.1352, 362.1322, found: 360.1331, 362.1331; IR ($CHCl_3$) 3370, 1580 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 2.16 (1 H, m, 5′-H), 2.77 (1 H, m, 4′-H), 3.3–4.0 (7 H, m, CH_2OH, CH_2OBn, NH_2, and OH), 4.54 (2 H, s, CH_2Ph), 5.03 (1 H, m, 1′-H), 5.73 (3 H, m, 2′-, 3′-H and NH), 7.29 (5 H, s, Ph), 7.98 (1 H, s, 2-H).

(±)-9-[5′α-[(Benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]-6-chloropurine (9). To a solution of **8** (517 mg, 1.43 mmol) in triethyl orthoformate (7.9 mL, 47.6 mmol) was added, under ice-cooling and stirring, 12 N HCl (0.10 mL), and the mixture was stirred at room temperature for 12 h. The residue obtained after evaporation of the solvent in vacuo was dissolved in THF (23 mL), and to the solution was added 0.5 N HCl (30 mL) with stirring and cooling. After being stirred for 2 h at room temperature, the mixture was neutralized with aqueous 1 N NaOH and concentrated in vacuo. The product was taken up in AcOEt and dried on $MgSO_4$. The residue obtained after evaporation of the solvent was chromatographed on silica gel. Elution with hexane–ethyl acetate (1:1) gave 462 mg (86%) of **9**: colorless oil; EI-HRMS (*m/z*) calcd for $C_{19}H_{19}ClN_4O_2$ (M^+), $C_{19}H_{19}ClN_4O_2$ (M^+ + 2) 370.1196, 372.1165, found 370.1158, 372.1138; IR ($CHCl_3$) 3450, 1590, 1560 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 2.64 (1 H, m, 5′-H), 2.88 (1 H, m, 4′-H), 3.23 (1 H, br s, OH), 3.72 (2 H, d, *J* = 6 Hz, CH_2OH), 3.80 (2 H, m, CH_2OBn), 4.51 (2 H, s, CH_2Ph), 5.58 (1 H, m, 1′-H), 5.77 (1 H, ddd, *J* = 5, 1, and 1 Hz, 3′-H), 6.08 (1 H, ddd, *J* = 5, 2, and 2 Hz, 2′-H), 7.25 (5 H, s, Ph), 8.24 (1 H, s, purine H), 8.66 (1 H, s, purine H).

(±)-9-[5′α-[(Benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]adenine (10). Ammonia gas was passed, under ice-cooling, to a solution of **9** (244 mg) in absolute methanol (25 mL) for 1 h, and the whole mixture was heated in a sealed tube at 50 °C for 20 h. The residue obtained after evaporation of the solvent was chromatographed on silica gel. Elution with ethyl acetate–methanol (8:1) gave at first 180 mg (77%) of the adenine derivative **10** and then 21 mg (9%) of 6-methoxypurine derivative **11**.

10: colorless needles, mp 171–173 °C (CH_2Cl_2); IR (Nujol) 3450 cm^{-1}; ^1H NMR (DMSO-d_6, 500 MHz) δ 2.44 (1 H, dddd, *J* = 5, 5, 5, and 5 Hz, 5′-H), 2.72 (1 H, m, 4′-H), 3.52 (1 H, ddd, *J* = 11, 5, and 5 Hz, C*H*HOH), 3.61 (1 H, ddd, *J* = 11, 5, and 5 Hz, CH*H*OH), 3.64 (2 H, dd, *J* = 9 and 5 Hz, C*H*HOBn), 3.66 (2 H, dd, *J* = 9 and 5 Hz, CH*H*OBn), 4.46 (1 H, d, *J* = 12 Hz, C*H*HPh), 4.49 (1 H, d, *J* = 12, Hz, CH*H*Ph), 4.77 (1 H, t, *J* = 5 Hz, OH), 5.52 (1 H, ddd, *J* = 6, 2, and 2 Hz, 1′-H), 5.79 (1 H, ddd, *J* = 6, 2, and 2 Hz, 3′-H), 6.66 (1 H, dd, *J* = 6, and 2 Hz, 2′-H), 7.17 (2 H, br s, NH_2), 7.28 (5 H, m, Ph), 8.05 (1 H, s, purine H), 8.13 (1 H, s, purine H). Anal. ($C_{19}H_{21}N_5O_2$) C, H, N.

11: colorless oil; EI-HRMS (*m/z*) calcd for $C_{20}H_{22}N_4O_3$ (M^+) 366.1692, found 366.1717; IR ($CHCl_3$) 3450, 1600, 1580 cm^{-1}; ^1H NMR ($CDCl_3$, 60 MHz) δ 2.5–3.1 (2 H, m, 4′-H and 5′-H), 3.71 (2 H, d, *J* = 6 Hz, CH_2OH), 3.80 (2 H, m, CH_2OBn), 4.17 (3 H, s, OMe), 4.52 (2 H, s, CH_2Ph), 5.51 (1 H, m, 1′-H), 5.75 (1 H, ddd, *J* = 6, 2, and 2 Hz, 3′-H), 6.07 (1 H, ddd, *J* = 6, 2, and 2 Hz, 2′-H), 7.27 (5 H, s, Ph), 7.94 (1 H, s, purine H), 8.46 (1 H, s, purine H).

(±)-9-[4′β,5′α-Bis(hydroxymethyl)cyclopent-2′-en-1′β-yl]-adenine (12). To a solution of **10** (154 mg, 0.44 mmol) in anhydrous CH_2Cl_2 (30 mL) cooled at −78 °C was added portionwise,

under argon atmosphere, 1 N BCl_3–CH_2Cl_2 solution (13 mL, 13 mmol), and the mixture was stirred for 3 h. After the reaction, a mixture of methanol–CH_2Cl_2 (1:1, 18 mL) was slowly added and the temperature of the mixture was raised to room temperature. The solvent was evaporated in vacuo, and MeOH (7 mL) was added and after 10 min, the solvent was evaporated. This procedure was repeated four times. Ten milliliters of methanol was added to the residue obtained after evaporation of the solvent, and to this mixture Amberlite IRA-45 (2 g) was added and the whole was stirred for 1 h at room temperature. After filtration to remove the resin, the filtrate was evaporated. The residue thus obtained was chromatographed on silica gel. Elution with $CHCl_3$–MeOH (10:1) afforded 95 mg (83%) of **12**.

12: colorless prisms, mp 179–180 °C (MeOH); EI-HRMS (m/z) calcd for $C_{12}H_{15}N_5O_2$ (M^+) 261.1225, found 261.1212; UV (MeOH) λ_{max} 261 nm (ϵ: 14200); ^{1}H NMR (DMSO-d_6, 500 MHz) δ 2.23 (1 H, dddd, J = 5, 5, 5 and 5 Hz, 5′-H), 2.70 (1 H, m, 4′-H), 3.51 (1 H, ddd, J = 10, 5, and 5 Hz, C*H*HOH), 3.57 (1 H, ddd, J = 10, 5, and 5 Hz, CH*H*OH), 3.61 (2 H, dd, J = 5 and 5 Hz, CH_2OH), 4.78 (1 H, t, J = 5 Hz, OH), 4.82 (1 H, t, J = 5 Hz, OH), 5.43 (1 H, m, 1′-H), 5.79 (1 H, m, 3′-H), 6.06 (1 H, m, 2′-H), 7.18 (2 H, br s, NH_2), 8.05 (1 H, s, purine H), 8.13 (1 H, s, purine H). Anal. ($C_{12}H_{15}N_5O_2$) C, H, N.

(±)-2-Amino-4-[[5′α-[(benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]amino]-6-chloropyrimidine (13). To a solution of **7** (373 mg, 1.60 mmol) in ethanol (40 mL) was added 2-amino-4,6-dichloropyrimidine (324 mg, 1.92 mmol) and triethylamine (485 mg, 4.80 mmol), and the mixture was heated at 100 °C in a sealed tube. After 40 h, the solvent was removed in vacuo, and the residue was chromatographed on a silica gel column (hexane–AcOEt: 1:1) to give 462 mg (80%) of the product: colorless oil; EI-HRMS (m/z) calcd for $C_{18}H_{21}ClN_4O_2$ (M^+), $C_{18}H_{21}ClN_4O_2$ (M^+ + 2) 360.1352, 362.1322, found 360.1323, 362.1306; IR ($CHCl_3$) 3550, 3450, 1610, 1580, 1570 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 2.11 (1 H, m, 5′-H), 2.64 (1 H, m, 4′-H), 3.35 (1 H, br s, OH), 3.4–3.7 (4 H, m, CH_2OH and CH_2Bn), 4.54 (2 H, s, CH_2Ph), 4.65 (1 H, m, 1′-H), 5.21 (2 H, br s, NH_2), 5.41 (1 H, br d, J = 10 Hz, NH), 5.70 (2 H, s, 2′- and 3′-H), 5.79 (1 H, s, 5-H), 7.28 (5 H, s, Ph).

(±)-2-Amino-4-[[5′α-[(benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]amino]-5-[(4′-chlorophenyl)azo]-6-chloropyrimidine (14). To a solution of **13** (1.20 g), AcONa (6.6 g) in a mixture of water (17 mL), and methanol (10 mL) was added, under ice-cooling and stirring, the diazonium solution prepared from *p*-chloroaniline (510 mg, 4.0 mmol) and $NaNO_2$ (303 mg, 4.40 mmol) in water (4 mL) in 3 N HCl (8 mL), and the whole mixture was stirred for 6 h at room temperature. The precipitate obtained after evaporation of the solvent and addition of water was collected by filtration to give 1.34 g (80%) of the product: mp 202–205 °C (CH_2Cl_2–hexane); EI-HRMS (m/z) calcd for $C_{24}H_{24}Cl_2N_6O_2$ (M^+, M^+ + 2, M^+ + 4) 498.1337, 500.1307, 502.1337, found 498.1380, 500.1310, 502.1324; IR (Nujol) 3450, 1570 cm^{-1}; ^{1}H NMR (DMSO-d_6, 60 MHz) δ 2.23 (1 H, m, 5′-H), 2.67 (1 H, m, 4′-H), 3.4–4.1 (5 H, m, CH_2OH, CH_2Bn, and OH), 4.51 (2 H, s, CH_2Ph), 5.20 (1 H, m, 1′-H), 5.82 (2 H, m, 2′- and 3′-H), 7.26 (5 H, s, Ph), 7.3–8.0 (6 H, m, NH_2 and *p*-Cl-Ph), 10.33 (1 H, br d, J = 8 Hz, NH).

(±)-2,5-Diamino-4-[[5′α-[(benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]amino]-6-chloropyrimidine (15). To a solution of **14** (1336 mg, 2.03 mmol) in a mixture of ethanol and water (1:1, 80 mL) was added zinc powder (1.74 g, 2.03 mmol) and acetic acid (0.85 mL), and the mixture was refluxed for 3 h. After filtration, the filtrate was concentrated in vacuo. The residue was chromatographed on a silica gel column (hexane–AcOEt 1:2) to give 592 mg (59%) of the product: mp 121–122 °C (CH_2Cl_2–hexane); EI-HRMS (m/z) calcd for $C_{18}H_{22}ClN_5O_2$ (M^+, M^+ + 2) 375.1461, 377.1431, found 375.1474, 377.1468; IR ($CHCl_3$) 3520, 1610, 1560 cm^{-1}; ^{1}H NMR ($CDCl_3$, 60 MHz) δ 2.12 (1 H, m, 5′-H), 2.72 (1 H, m, 4′-H), 3.0 (3 H, br s, NH_2 and OH), 3.1–3.9 (4 H, m, CH_2OH and CH_2OBn), 4.53 (2 H, s, CH_2Ph), 4.68 (2 H, br s, NH_2), 4.74 (1 H, m, 1′-H), 5.70 (2 H, s, 2′- and 3′-H), 5.74 (1 H, br d, J = 9 Hz, NH), 7.27 (5 H, s, Ph).

(±)-2-Amino-9-[5′α-[(benzyloxy)methyl]-4′β-(hydroxymethyl)cyclopent-2′-en-1′β-yl]-6-chloropurine (16). To a suspension of the above product (206 mg, 0.55 mmol) in triethyl orthoformate (3 mL) was added, under ice-cooling, 12 N HCl (0.14 mL), and the mixture was stirred at room temperature for 14 h. The residue obtained after evaporation of the solvent was dissolved in THF (3 mL) and, to this solution, 0.5 N HCl (3 mL) was added. After stirring for 6 h, the mixture was neutralized by the addition of 2 N aqueous KOH. The residue obtained after evaporation of the solvent was chromatographed on a silica gel column ($CHCl_3$–MeOH 50:1) to give 195 mg (92%) of **16**: colorless oil; EI-HRMS (m/z) calcd for $C_{19}H_{20}ClN_5O_2$ (M^+, M^+ + 2) 385.1305, 387.1275, found 385.1325, 387.1293; IR ($CHCl_3$) 3450, 1610, 1570 cm^{-1}; ^{1}H NMR ($CDCl_3$, 60 MHz) δ 2.5–3.1 (2 H, m, 4′- and 5′-H), 3.70 (2 H, d, J = 6 Hz, CH_2OH), 3.8–4.1 (3 H, m, CH_2OBn and OH), 4.55 (2 H, s, CH_2Ph), 5.27 (2 H, br s, NH_2), 5.38 (1 H, m, 1′-H), 5.75 (1 H, m, 3′-H), 6.06 (1 H, m, 2′-H), 7.32 (5 H, s, Ph), 7.86 (1 H, s, 8-H).

(±)-9-[5α-[(Benzyloxy)methyl]-4β-(hydroxymethyl)-cyclopent-2-en-1β-yl]guanine (17). To a solution of **16** (123 mg, 0.32 mmol) in dioxane (3 mL) was added 0.5 N KOH (4 mL), and the mixture was refluxed for 5 h. The residue obtained after evaporation of the solvent in vacuo was chromatographed on a silica gel column ($CHCl_3$–MeOH 20:1) to give 92 mg (78%) of **17**: mp 109–111 °C (MeOH–AcOEt); EI-HRMS (m/z) calcd for $C_{19}H_{21}N_5O_3$ (M^+) 367.1645, found 367.1626; IR (Nujol) 3400 cm^{-1}; ^{1}H NMR (DMSO-d_6, 500 MHz) δ 2.30 (1 H, dddd, J = 6, 6, 6 and 6 Hz, 5′-H), 2.68 (1 H, m, 4′-H), 3.49 (1 H, ddd, J = 10, 5, and 5 Hz, C*H*HOH), 3.59 (1 H, ddd, J = 10, 5 and 5 Hz, CH*H*OH), 3.61 (1 H, dd, J = 10 and 5 Hz, C*H*HOBn), 3.64 (1 H, dd, J = 10 and 5 Hz, CH*H*OBn), 4.48 (2 H, s, CH_2Ph), 4.76 (1 H, t, J = 5 Hz, OH), 5.28 (1 H, m, 1′-H), 5.72 (1 H, m, 3′-H), 6.01 (1 H, m, 2′-H), 6.39 (2 H, br s, NH), 7.2–7.3 (5 H, m, Ph), 7.65 (1 H, s, 8-H).

(±)-9-[4′β,5′α-Bis(hydroxymethyl)cyclopent-2′-en-1′β-yl]guanine (18). The deblocking of benzyl group of **17** (76 mg) by BCl_3 was carried out under exactly the same condition as described for the synthesis of **12** from **10** to give 51 mg (89%) of **18**: mp 256 °C dec (from MeOH–AcOEt); EI-HRMS (m/z) calcd for $C_{12}H_{15}N_5O_3$ (M^+) 277.1174, found 277.1177; IR (Nujol) 3400 cm^{-1}; ^{1}H NMR (DMSO-d_6, 500 MHz) δ 2.16 (1 H, dddd, J = 5, 5, and 5 Hz, 5′-H), 2.67 (1 H, m, 4′-H), 3.4–3.6 (4 H, m, CH_2OH and CH_2OBn), 4.74 (1 H, t, J = 5 Hz, OH), 4.78 (1 H, t, J = 5 Hz, OH), 5.19 (1 H, m, 1′-H), 5.71 (1 H, m, 3′-H), 6.00 (1 H, m, 2′-H), 6.42 (2 H, br s, NH_2), 7.62 (1 H, s, 8-H), 10.58 (1 H, br s, NH): UV (MeOH) 254 and 273 nm (ϵ 12200 and 8300). Anal. ($C_{12}H_{15}N_5O_3$) C, H, N.

Registry No. **1**, 39939-07-6; **2**, 140440-30-8; **3**, 140440-31-9; **4**, 140440-32-0; **5**, 140440-33-1; **6**, 140440-34-2; **7**, 140440-35-3; **8**, 140440-36-4; **9**, 140440-37-5; **10**, 140440-38-6; **11**, 140440-39-7; **12**, 140440-40-0; **13**, 140440-41-1; **14**, 140440-42-2; **15**, 140440-43-3; **16**, 140440-44-4; **17**, 140440-45-5; **18**, 140440-46-6; TsCN, 19158-51-1; $ClCO_2Et$, 541-41-3; $(EtO)_3CH$, 122-51-0; 5-amino-4,6-dichloropyrimidine, 5413-85-4; 2-amino-4,6-dichloropyrimidine, 56-05-3; *p*-chlorophenyldiazonium chloride, 2028-74-2; cyclopentadienylthallium, 34822-90-7.