Purines. LXXVI.[1] Alkylation of 8-Oxoadenine Derivatives: Syntheses of 3,7-Dialkyl-, 3,9-Dialkyl-, and 3,7,9-Trialkyl-8-oxoadenines

Taisuke ITAYA,* Yasutaka TAKADA, Tae KANAI, Miki KANEKO, and Tozo FUJII

Faculty of Pharmaceutical Sciences, Kanazawa University, Takara-machi, Kanazawa 920, Japan.
Received April 7, 1997; accepted May 21, 1997

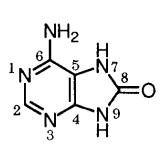
3-Alkyl-8-hydroxyadenines (5) have been shown to undergo regioselective methylation at the 7- or 9-position depending on the reaction conditions. Thus, treatment of 5a, c with dimethyl sulfate in aqueous NaOH provided 3-alkyl-7-methyl-8-oxoadenines (6d, h) in 48—60% yields, together with 3-alkyl-8-methoxyadenines (4d, h), whereas treatment of 5a—c with MeI in $AcNMe_2$ at 40 °C for 48 h and subsequent anion exchange afforded 3-alkyl-9-methyl-8-oxoadenine hydrochlorides (7d, g, h·HCl) in 50—59% yields. However, the reactions of 5a, c with EtI or $PhCH_2Br$ took place slowly, giving complex mixtures of products.

Compounds 6d,h were alternatively prepared in 51% and 31% yields, respectively, together with 3-alkyl-7,9-dimethyl-8-oxoadenine hydrochlorides (11d, h·HCl), by treatment of 3-alkyl-8-methoxyadenines (4d, h) with MeI in $AcNMe_2$ at room temperature for 6 h, followed by hydrolysis with boiling aqueous HCl. This method was applicable to ethylation with EtI, and 7-ethyl-3-methyl-8-oxoadenine (6e) was obtained in 70% yield from 8-ethoxy-3-methyladenine (4e). Compound 11h was shown to be obtainable through further methylation of 6h. Thus, 11d, h were prepared in good yields by treatment of 6d, h with MeI in $AcNMe_2$.

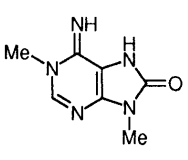
Compounds 7, to which zwitterionic structures were assigned, were stable in 0.1 N aqueous NaOH at room temperature, whereas 11d, h were no longer stable under such conditions.

Key words 8-oxoadenine alkylation; 3,7-dialkyl-8-oxoadenine; 3,9-dialkyl-8-oxoadenine; 3,7,9-trialkyl-8-oxoadenine; 8-alkoxyadenine acid hydrolysis; N^6-demethylcaissarone synthesis

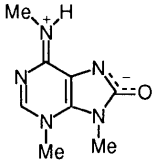
Natural occurrence of 1,9-dimethyl-8-oxoadenine (**2**) (isolated only in the form of the N^6-acetyl derivative),[2] N^6,3,9-trimethyl-8-oxoadenine (caissarone) (**3**),[3] 8-oxoadenosine derivatives,[4] and an N^6-substituted 8-oxoadenine derivative[5] and the biological activity of 8-oxoadenine-incorporating DNA[6] have directed our attention to the synthesis and chemistry of the 8-oxoadenine family.[7] Although 8-oxoadenine (**1**)[8] itself and its nucleoside 8-oxoadenosine[9] have long been known, it was not until our recent syntheses of 9-methyl-8-oxoadenine (**13**),[7a] 3-methyl-8-hydroxyadenine (**5a**),[10] 1-methyl-8-oxoadenine,[11] and 7-methyl-8-oxoadenine[11] that all six possible positional isomers of 8-oxoadenine monomethylated at a hetero atom became available. Among the 11 possible positional isomers of N^x,N^y-dimethyl-8-oxoadenines, the N^6,9- and 1,9-dimethyl isomers are known,[7a] but no N^x,3-dimethyl-8-oxoadenines have been reported. We have recently reported that the syntheses of 3-alkyl-8-hydroxyadenines (**5**) and 8-alkoxy-3-alkyladenines (**4**) from 3-alkyladenines are possible through N(7)-oxidation followed by *O*-alkylation and hydrolysis or alcoholysis, and we suggested that compounds **5** and **4** might be good precursors for syntheses of such unknown N^x,3-dialkyl-8-oxoadenines.[10] Now we report the first syntheses of 3,7-dialkyl- (**6**), 3,9-dialkyl- (**7**), and 3,7,9-trialkyl-8-oxoadenines (**11**) by utilizing **5** and **4** as starting materials.



1



2



3

* To whom correspondence should be addressed.

Results and Discussion

Methylation of 5 with Dimethyl Sulfate The first substrate selected for methylation was 3-benzyl-8-hydroxyadenine (**5c**)[10] because the methylation site may be determined by leading the products to any of six known *N*- or *O*-monomethyl-8-oxoadenines[11] by means of debenzylation. On treatment with 6 mol eq of dimethyl sulfate in 0.1 N aqueous NaOH at 30 °C for 18 h, compound **5c** provided 3-benzyl-7-methyl-8-oxoadenine (isolated as the hydrochloride **6h**·HCl in 60% yield) and 3-benzyl-8-methoxyadenine (**4h**)[10] (7%). The correctness of the structure of **6h**·HCl was corroborated by the fact that it furnished 7-methyl-8-oxoadenine[11] in 94% yield on hydrogenolysis with H_2 and Pd–C. Similar treatment of 3-methyl-8-hydroxyadenine (**5a**)[10] with dimethyl sulfate for 2.5 h afforded 3,7-dimethyl-8-oxoadenine (**6d**) (48% yield) and 3-methyl-8-methoxyadenine (**4d**)[10] (8%). The UV spectral similarity between **6d** and **6h** supported the correctness of the structure of **6d**. The IR spectrum (Nujol) of **6d**, lacking C=O absorption bands in the 1755—1680 cm^{-1} region, suggests an 8-hydroxy structure in the solid state. On the other hand, it was suggested that **6d** preferred the keto form to the enol form in $(CD_3)_2SO$ solution, because the ^{1}H-NMR spectrum measured in this solution showed a broad two-proton C(6)-NH_2 singlet at δ 6.72 but no signals assignable to imino and hydroxy protons.

Methylation of 5 with MeI Treatment of **5c**[10] with 10 mol eq of MeI in $AcNMe_2$ at 40 °C for 48 h furnished, after anion exchange, 3-benzyl-9-methyl-8-oxoadenine hydrochloride (**7h**·HCl) in 50% yield. The 9-methyl structure was assignable to **7h**·HCl on the basis of its hydrogenolysis with H_2 and Pd–C, which generated 9-methyl-8-oxoadenine (**13**)[7a] in 76% yield. Similar treatment of **5a**, **b**[10] with MeI afforded the corresponding

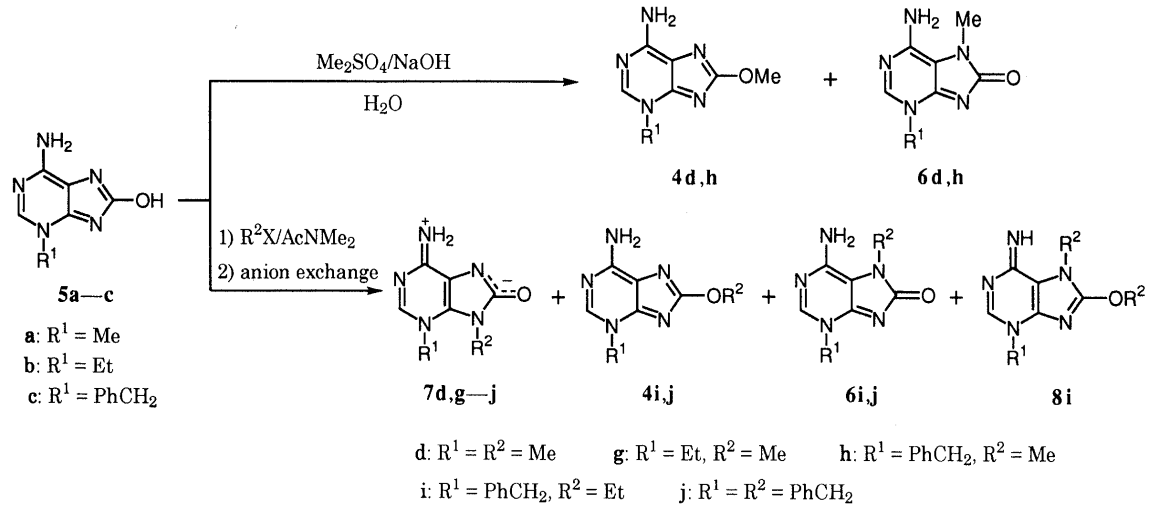


Chart 1

3-alkyl-9-methyl-8-oxoadenine hydrochlorides (**7d**, **g**·HCl) in 59% and 50% yields, respectively. The correctness of the structures of **7d**, **g**·HCl was established by their UV spectral similarity to **7h**·HCl. The free bases **7d**, **h** were prepared by neutralization of aqueous solutions of the salts **7d**, **h**·HCl. The IR spectra of **7d**, **h** lacked C=O absorption bands; the UV spectra resembled those of caissarone (**3**), with an expected hypsochromic shift of the maxima due to N^6-demethylation. Unlike 8-unsubstituted 3,9-dialkyladenines,[12] **7d**, **h** were stable in 0.1 N aqueous NaOH at room temperature as judged by UV spectroscopy, and the pK_a 6.63 for N^6-demethylcaissarone (**7d**) was similar in magnitude to those (6.78 and 6.73)[7b] of **3** and N^6,N^6,3,9-tetramethyl-8-oxoadenine. The above results indicate zwitterionic structures for **7d**, **h**, as in the case of **3**.[7b] We have already reported that **3** is capable of forming a hetero-base pair with 2′,3′,5′-tri-*O*-acetylguanosine in $(CD_3)_2SO$.[7b] The N^6-demethyl analogues **7d**, **h** were expected to behave similarly. However, their extremely poor solubility in $(CD_3)_2SO$ made it difficult to investigate such interactions. Compound **7d** was hardly soluble in either H_2O or organic solvents and was inert to further methylation with MeI in $AcNMe_2$ at 40 °C in either the absence or presence of NaH.

Ethylation and Benzylation of 5 Unlike the above methylation of **5** with MeI, ethylation of **5a**[10] with 5 mol eq of EtI in $AcNMe_2$ proceeded only slowly at 40 °C to afford a complex mixture of products after 72 h. We were able to recover **5a** in 41% yield, but failed to isolate the products. Benzylation of **5a** with $PhCH_2Br$ under similar conditions also took place slowly to give a complex mixture of products, from which we could not obtain any pure compounds.

On the other hand, the products from similar ethylation and benzylation of **5c**[10] could be separated by chromatography. Thus, treatment of **5c** with 10 mol eq of EtI in $AcNMe_2$ at 40 °C for 48 h gave, after anion exchange, 3-benzyl-8-ethoxyadenine (**4i**) in 32% yield, together with **5c** (9% recovery) and two monoethylated compounds, which were inferred to be 3-benzyl-7-ethyl-8-oxoadenine (**6i**) (2%) and 3-benzyl-9-ethyl-8-oxoadenine (**7i**) (1%) by comparison of their ^{1}H-NMR spectra with those of the 7-methyl and 9-methyl analogues (**6h** and **7h**). The structure of the major product **4i** was assigned by direct comparison with an authentic sample prepared according to the reported procedure.[10] The diethylated structure was suggested for a third minor product, based on the ^{1}H-NMR spectrum, and this compound was presumed to be 3-benzyl-8-ethoxy-7-ethyladenine hydrochloride (**8i**·HCl) on the basis of its acid hydrolysis, leading to **6i**.

Benzylation of **5c**[10] with an excess of $PhCH_2Br$ at 40 °C for 48 h produced three compounds, together with **5c** (11% recovery). The major product, obtained in 13% yield, was determined to be 3,7-dibenzyl-8-oxoadenine (**6j**) by comparison of its UV and ^{1}H-NMR spectra with those of **6h**. The structure of one of the minor products, obtained in 3% yield, was established to be 3-benzyl-8-benzyloxyadenine (**4j**) by comparison of its ^{1}H-NMR spectrum with that of **4i** and by hydrolysis with hot 1 N aqueous HCl to give **5c**. Although the other minor product was most likely 3,9-dibenzyl-8-oxoadenine (**7j**), its structure could not be determined.

Alkylation of 4 3-Benzyl-8-methoxyadenine (**4h**)[10] afforded a complex mixture of products on treatment with 5 mol eq of MeI in $AcNMe_2$ at room temperature for 6 h. The mixture was then subjected to hydrolysis with boiling 1 N aqueous HCl after treatment with Amberlite IRA-402 (Cl^-), affording **6h**·HCl in 32% yield and the dimethylated product **11h**·HCl in 22% yield. The use of an equimolar amount of MeI could not suppress the formation of the latter product. The yield of **11h**·HCl was raised to 71%, with a decreased yield (25%) of **6h**·HCl, when the methylation was conducted at 30 °C for 18 h. These results suggest that **11h** might have been formed through further methylation of **6h**. Indeed, **11h** was obtained (isolated as the hydrochloride in 98% yield) on treatment of **6h**·HCl with MeI in the presence of 1 mol eq of K_2CO_3 at 30 °C for 48 h, verifying the 7-methyl structure for **11h**. Debenzylation of **11h**·HCl with H_2 and Pd–C generated the dimethyl compound **12** in 87% yield, and this was identical with the product from methylation of 9-methyl-8-oxoadenine (**13**), establishing the correctness of the 9-methyl structure for **11h** (Chart 3). Furthermore, the correctness of the 7,9-dimethyl structure for **12** was supported by the UV

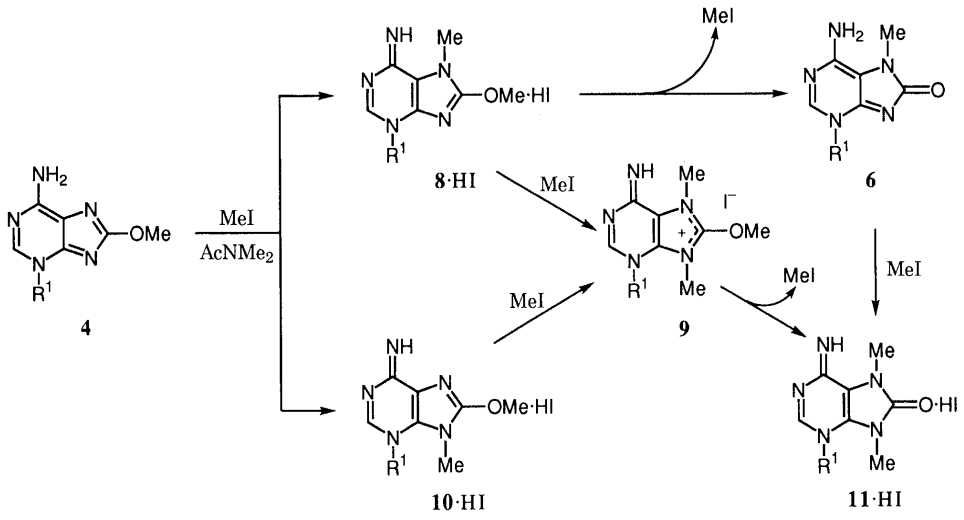


Chart 2

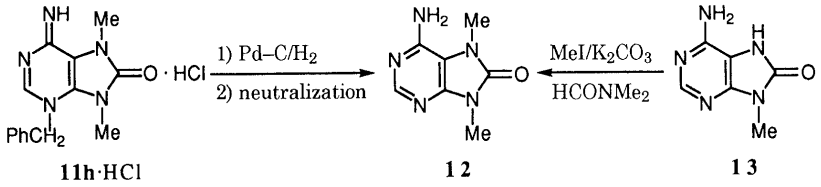


Chart 3

spectral similarity to 7-methyl-8-oxoadenosine.[11,13]

Similar treatment of 8-methoxy-3-methyladenine (**4d**)[10] with MeI at room temperature for 6 h provided, after acid hydrolysis, **6d** in 51% yield and **11d**·HCl in 22% yield. The structure of **11d**·HCl was assignable on the basis of the UV spectral similarity to the 3-benzyl analogue **11h**·HCl and was further supported by its formation in 77% yield on methylation of **6d** with MeI in $AcNMe_2$ at 30 °C for 24 h. Unlike the zwitterions **7d, h**, compounds **11d, h** were not stable under alkaline conditions as judged from their UV spectra taken in 0.1 N aqueous NaOH at room temperature, suggesting that they underwent ring-opening at the pyrimidine moiety in analogy with 3,9-dialkyladenines.[12]

We then isolated the products of the methylation of **4d** without recourse to acid hydrolysis in order to get an insight into the reaction features. Two trimethyl compounds were isolated by means of preparative TLC, together with **6d** (28%) and **11d** (isolated as the hydrochloride in 11% yield). One of them, obtained in 29% yield, was 8-methoxy-3,7-dimethyladenine hydriodide (**8d**·HI), because it generated **6d** on treatment with boiling 1 N aqueous HCl. The 8-methoxy-3,9-dimethyl structure **10d**·HI was assignable to the other (obtained in 12% yield) on the basis of its acid hydrolysis that yielded **7d**·HCl. It is interesting to note that we could not find **7d** in the hydrolysate of the reaction mixture, as mentioned above. This apparent discrepancy may be resolved by assuming the formation of 3,7,9-trimethyl-8-methoxyadeninium iodide (**9d**) from **10d** in the course of methylation. If this compound coexists in the reaction mixture, even to a small

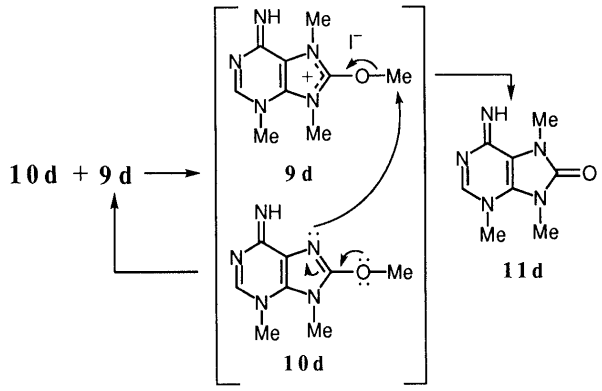
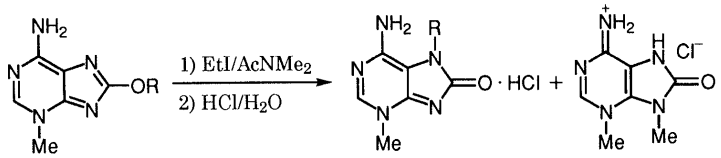


Chart 4

extent, it might methylate **10d** on treatment with hot aqueous HCl, resulting in the formation of **11d** and regeneration of **9d**: the net reaction is $O^8 \rightarrow N(7)$ methyl migration in **10d** (Chart 4). Thus, the results of the methylation of **4d, h** with an excess of MeI may be illustrated as shown in Chart 2.

Ethylation of **4d**[10] with an excess of EtI in $AcNMe_2$ at 40 °C for 4 d afforded a more complex mixture of products. We obtained two dimethyl compounds [**6d** (7%) and N^6-demethylcaissarone hydrochloride (**7d**·HCl) (16%)], together with **6e** (17%), from the acid hydrolysate of the reaction mixture (Chart 5). The formation of **6d** is explicable in terms of competitive reaction of MeI, which might have been liberated from the methoxy group in the course of the reaction, with slower reaction of the initially added EtI. Similar alkyl migrations have been thoroughly



4d,e **6d,e·HCl** **7d·HCl**

d: R = Me **e**: R = Et

Chart 5

investigated by us in connection with cross alkylations of 1-alkoxyadenines.[14] Compound **7d** was most likely produced through methylation of **5d**, which was formed by demethylation of **4d**. The reaction of the 8-ethoxy analogue **4e** with EtI under similar conditions was simple, and **6e** was obtained, after acid hydrolysis of the reaction mixture, in 70% yield as the sole isolable product. The correctness of the structure of **6e** was corroborated by comparison of its UV and ^{1}H-NMR spectra with those of **6d**.

In conclusion, we have achieved the preparation of hitherto unknown 3,7-dialkyl- (**6**), 3,9-dialkyl- (**7**), and 3,7,9-trialkyl-8-oxoadenines (**11**) by alkylations of **5** and **4**, both of which are obtainable from 3-alkyladenine 7-oxides through 7-alkoxy-3-alkyladenines.[10] Thus, the present results provide further examples of the synthetic utility of an *N*-alkoxy group as a control synthon for chemical modification of the adenine ring.[15]

Experimental

General Notes All melting points were determined by using a Yamato MP-1 or a Büchi model 530 capillary melting point apparatus, and values are corrected. Spectra reported herein were recorded on a JEOL JMS-SX102A mass spectrometer, a Hitachi model 320 UV spectrophotometer [for solutions in 95% aqueous EtOH, 0.1 N aqueous HCl (pH 1), 0.005 M phosphate buffer (pH 7), and 0.1 N aqueous NaOH (pH 13)], a Shimadzu FTIR-8100 IR spectrophotometer, or a JEOL JNM-EX-270 NMR spectrometer. Unless otherwise stated, NMR spectra were recorded at 25 °C in $(CD_3)_2SO$ with Me_4Si as an internal standard, but sodium 3-(trimethylsilyl)-1-propanesulfonate was used for CF_3CO_2D solution. Elemental analyses and MS measurements were performed by Mr. Y. Itatani, Dr. M. Takani, and their associates at Kanazawa University. Flash chromatography was performed according to the reported procedure.[16] The following abbreviations are used: br = broad, m = multiplet, q = quartet, s = singlet, sh = shoulder, t = triplet.

Methylation of 5a with Dimethyl Sulfate Dimethyl sulfate (of *ca.* 95% purity) (730 mg, 5.5 mmol) was added to a solution of **5a**[10] (302 mg, 1.83 mmol) in 0.1 N aqueous NaOH (55 ml), and the mixture was stirred at room temperature for 2.5 h. The resulting yellow solution was concentrated *in vacuo*, and the residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (20 : 7 : 1, v/v)] to afford **4d** (27 mg, 8%), mp 258—260 °C (dec.), which was identical (by comparison of the IR spectrum and TLC mobility) with an authentic specimen;[10] crude 3,7-dimethyl-8-oxoadenine (**6d**) (174 mg), mp 290—292 °C (dec.); and **5a** (25 mg, 8% recovery), mp > 300 °C. Recrystallization of crude **6d** from H_2O afforded pure **6d** (157 mg, 48%), mp > 300 °C. Further recrystallization of this sample from H_2O provided an analytical sample of **6d** as colorless plates, mp > 300 °C; MS *m/z*: 179 (M^+); UV $\lambda_{max}^{95\%\,EtOH}$ 235 nm (ε 15400), 301 (17200); $\lambda_{max}^{H_2O}$ (pH 1) 219 (21900), 290 (18300); $\lambda_{max}^{H_2O}$ (pH 7) 217 (19100), 230 (16900), 296 (18400); $\lambda_{max}^{H_2O}$ (pH 13) 230 (17000), 296 (18500); IR ν_{max}^{Nujol} cm^{-1}: 3478, 3254, 3131 (OH and NH), 1667, 1651 (C=N); ^{1}H-NMR δ: 3.38 [3H, s, N(7)-Me], 3.65 [3H, s, N(3)-Me], 6.72 (2H, br s, NH_2), 8.09 [1H, s, C(2)-H]. *Anal.* Calcd for $C_7H_9N_5O$: C, 46.92; H, 5.06; N, 39.09. Found: C, 46.66; H, 5.06; N, 38.93.

3,7-Dimethyl-8-oxoadenine hydrochloride (**6d**·HCl) was prepared by dissolving **6d** (55 mg, 0.31 mmol) in hot H_2O (1 ml) and adjusting the pH of the solution to 1 with 10% aqueous HCl. The resulting solution was concentrated *in vacuo*, and the residue was recrystallized from 90% (v/v) aqueous MeOH to give **6d**·HCl (34 mg, 52%), mp > 300 °C. Further recrystallization of this sample from 90% (v/v) aqueous MeOH afforded an analytical sample of **6d**·HCl as colorless plates, mp > 300 °C; UV $\lambda_{max}^{95\%\,EtOH}$ 235 nm (sh) (ε 11600), 298 (17100); $\lambda_{max}^{H_2O}$ (pH 1) 219 (21600), 290 (17900); $\lambda_{max}^{H_2O}$ (pH 7) 217 (19000), 230 (16800), 296 (18400); $\lambda_{max}^{H_2O}$ (pH 13) 230 (17000), 296 (18400); IR ν_{max}^{Nujol} cm^{-1}: 3301, 3156 (NH), 1709 (C=O), 1655 (C=N); ^{1}H-NMR δ: 3.50 [3H, s, N(7)-Me], 3.81 [3H, s, N(3)-Me], 8.12 (2H, br s, NH_2), 8.49 [1H, s, C(2)-H]. *Anal.* Calcd for $C_7H_9N_5O$·HCl: C, 38.99; H, 4.67; N, 32.48. Found: C, 38.82; H, 4.65; N, 32.37.

Methylation of 5c with Dimethyl Sulfate Dimethyl sulfate (of *ca.* 95% purity) (400 mg, 3.01 mmol) was added to a solution of **5c**[10] (241 mg, 1 mmol) in 0.1 N aqueous NaOH (30 ml), and the mixture was stirred at 30 °C for 18 h. The reaction mixture was concentrated *in vacuo*, and the residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (20 : 7 : 1, v/v)] to afford **6h**, mp 75—83 °C (dec.); ^{1}H-NMR δ: 3.38 [3H, s, N(7)-Me], 5.30 (2H, s, PhC$\underline{H}_2$), 6.84 (2H, br s, NH_2), 7.26—7.46 (5H, m, $\underline{Ph}CH_2$), 8.33 [1H, s, C(2)-H], and crude **4h**. Crude **4h** was recrystallized from a mixture of 5% aqueous NH_3 (1 ml) and MeOH (1 ml) to give **4h** (19 mg, 7%), mp 207—208 °C. This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **4h**.[10] Compound **6h** was triturated with one drop of 10% aqueous HCl. The mixture was concentrated *in vacuo*, and the residue was dried to give 3-benzyl-7-methyl-8-oxoadenine hydrochloride (**6h**·HCl) (174 mg, 60%), mp 278—282 °C (dec.). Recrystallization of this sample from 5% aqueous HCl afforded an analytical sample of **6h**·HCl as colorless plates, mp 281—288 °C (dec.); UV $\lambda_{max}^{95\%\,EtOH}$ 239 nm (ε 16000), 302 (15800); $\lambda_{max}^{H_2O}$ (pH 1) 222 (26800), 293 (17700); $\lambda_{max}^{H_2O}$ (pH 7) 234 (18400), 298 (16900); $\lambda_{max}^{H_2O}$ (pH 13) 234 (18400), 298 (16900); IR ν_{max}^{Nujol} cm^{-1}: 3294, 3115 (NH), 1707 (C=O), 1653 (C=N); ^{1}H-NMR δ: 3.48 [3H, s, N(7)-Me], 5.47 (2H, s, PhC$\underline{H}_2$), 7.33—7.47 (5H, m, $\underline{Ph}CH_2$), 8.21 (2H, br, NH_2), 8.70 [1H, s, C(2)-H]. *Anal.* Calcd for $C_{13}H_{13}N_5O$·HCl: C, 53.52; H, 4.84; N, 24.01. Found: C, 53.63; H, 4.83; N, 23.73.

Methylation of 5 with MeI The procedure employed for the preparation of **7d**·HCl will be described below in detail. The other methylations were accomplished similarly.

3,9-Dimethyl-8-oxoadenine Hydrochloride (*N*^{6}-Demethylcaissarone Hydrochloride) (7d·HCl) A mixture of **5a**[10] (330 mg, 2 mmol), MeI (2.84 g, 20 mmol), and $AcNMe_2$ (40 ml) was stirred at 40 °C for 48 h and then concentrated *in vacuo*. The residue was dissolved in H_2O (10 ml), and the solution was passed through a column packed with Amberlite IRA-402 (Cl^-) (4 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo*, and the residue was recrystallized from 5% aqueous HCl to afford **7d**·HCl (253 mg, 59%), mp 263—265 °C (dec.). Further recrystallization of this sample from 5% aqueous HCl afforded an analytical sample of **7d**·HCl as colorless plates, mp 274—276 °C (dec.); pK_a (at 30 °C and ionic strength 1.0) 6.63 ± 0.03;[17] UV $\lambda_{max}^{95\%\,EtOH}$ 224 nm (ε 20900), 294 (19000); $\lambda_{max}^{H_2O}$ (pH 1) 222 (21400), 291 (19800); $\lambda_{max}^{H_2O}$ (pH 7) 299 (15400); $\lambda_{max}^{H_2O}$ (pH 13) 305 (16600); IR ν_{max}^{Nujol} cm^{-1}: 3312, 3146 (NH), 1740 (C=O), 1668 (C=N); ^{1}H-NMR δ: 3.60 [3H, s, N(9)-Me], 4.11 [3H, s, N(3)-Me], 8.47 [1H, s, C(2)-H], 8.50 (2H, br, NH_2), 11.96 [1H, br s, N(7)-H]. *Anal.* Calcd for $C_7H_9N_5O$·HCl: C, 38.99; H, 4.67; N, 32.48. Found: C, 38.88; H, 4.71; N, 32.45.

3-Ethyl-9-methyl-8-oxoadenine Hydrochloride (7g·HCl) Compound **7g**·HCl (110 mg, 50%), mp 263—265 °C (dec.), obtained from **5b**·1/2H_2O[10] (179 mg, 0.951 mmol) was further recrystallized from 5% aqueous HCl to afford an analytical sample as colorless plates, mp 267—268 °C (dec.); UV $\lambda_{max}^{95\%\,EtOH}$ 224 nm (ε 21500), 295 (19100); $\lambda_{max}^{H_2O}$ (pH 1) 222 (21800), 291 (19300); $\lambda_{max}^{H_2O}$ (pH 7) 301 (15000); $\lambda_{max}^{H_2O}$ (pH 13) 305 (16000); IR ν_{max}^{Nujol} cm^{-1}: 3333, 3117 (NH), 1725 (C=O), 1674 (C=N);

^{1}H-NMR δ: 1.46 (3H, t, J=7 Hz, MeCH$_2$), 3.57 [3H, s, N(9)-Me], 4.49 (2H, q, J=7 Hz, MeCH$_2$), 8.55 [1H, s, C(2)-H], 8.56 (2H, br, NH$_2$), 12.09 [1H, br s, N(7)-H]. *Anal.* Calcd for $C_8H_{11}N_5O \cdot HCl$: C, 41.84; H, 5.27; N, 30.49. Found: C, 41.55; H, 5.28; N, 30.20.

3-Benzyl-9-methyl-8-oxoadenine Hydrochloride (7h·HCl) The hydrochloride **7h**·HCl (146 mg, 50%), mp 226—227 °C (dec.), obtained from **5c**[10] (241 mg, 1 mmol) was further recrystallized from 5% aqueous HCl to afford an analytical sample as colorless prisms, mp 229—231 °C (dec.); UV $\lambda_{max}^{95\%\,EtOH}$ 226 nm (ε 24000), 297 (18600); $\lambda_{max}^{H_2O}$ (pH 1) 224 (25300), 293 (19600); $\lambda_{max}^{H_2O}$ (pH 7) 304 (15100); $\lambda_{max}^{H_2O}$ (pH 13) 308 (15500); IR ν_{max}^{Nujol} cm^{-1}: 3262, 3212, 3061 (NH), 1730 (C=O), 1688, 1661 (C=N); ^{1}H-NMR δ: 3.26 [3H, s, N(9)-Me], 5.81 (2H, s, PhCH$_2$), 7.20—7.28 (3H) and 7.34—7.48 (2H) (m each, PhCH$_2$), 8.46 and 8.82 (1H each, br, NH$_2$), 8.64 [1H, s, C(2)-H], 12.02 [1H, br s, N(7)-H]. *Anal.* Calcd for $C_{13}H_{13}N_5O \cdot HCl$: C, 53.52; H, 4.84; N, 24.01. Found: C, 53.43; H, 4.80; N, 23.96.

3,9-Dimethyl-8-oxoadenine (N^6-Demethylcaissarone) (7d) Compound **7d** was prepared by adjusting the pH of a hot solution of **7d**·HCl (306 mg, 1.42 mmol) in H_2O (2 ml) to 8 with 10% aqueous Na_2CO_3. The precipitate that deposited was collected by filtration after cooling the mixture in an ice bath, washed with H_2O (2×1 ml), and dried to afford **7d** (231 mg, 91%), mp >300 °C. This was dissolved in 60% (v/v) aqueous MeOH and the solution was concentrated to half the initial volume to effect recrystallization, affording an analytical sample of **7d** as colorless prisms, mp >300 °C; MS *m/z*: 179 (M$^+$); UV $\lambda_{max}^{95\%\,EtOH}$ 313 nm;[18] $\lambda_{max}^{H_2O}$ (pH 1) 222 (ε 21300), 291 (20000); $\lambda_{max}^{H_2O}$ (pH 7) 300 (15400); $\lambda_{max}^{H_2O}$ (pH 13) 305 (16600); IR ν_{max}^{Nujol} cm^{-1}: 3214 (NH), 1669, 1632 (C=N); ^{1}H-NMR (CF_3CO_2D) δ: 3.91 [3H, s, N(9)-Me], 4.35 [3H, s, N(3)-Me], 8.42 [1H, s, C(2)-H]. *Anal.* Calcd for $C_7H_9N_5O$: C, 46.92; H, 5.06; N, 39.09. Found: C, 46.81; H, 5.05; N, 39.00.

3-Benzyl-9-methyl-8-oxoadenine (7h) The free base **7h** (67.2 mg, 86%) was prepared from **7h**·HCl (89.0 mg, 0.305 mmol) in a manner similar to that described above for the preparation of **7d**. Crude **7h** was then dissolved in 50% (v/v) aqueous MeOH and the solution was concentrated to half the initial volume to provide an analytical sample as colorless prisms, mp 295—297 °C; MS *m/z*: 255 (M$^+$); UV $\lambda_{max}^{95\%\,EtOH}$ 317 nm (ε 15000); $\lambda_{max}^{H_2O}$ (pH 1) 224 (25500), 293 (19800); $\lambda_{max}^{H_2O}$ (pH 7) 304 (15000); $\lambda_{max}^{H_2O}$ (pH 13) 307 (15100); IR ν_{max}^{Nujol} cm^{-1}: 3204 (NH), 1688, 1647 (C=N); ^{1}H-NMR δ: 3.22 [3H, s, N(9)-Me], 5.76 (2H, s, PhCH$_2$), 7.10—7.20 (2H) and 7.33—7.47 (3H) (m each, PhCH$_2$), 7.70 (2H, br, NH$_2$), 8.37±0.04[19] [1H, s, C(2)-H]. *Anal.* Calcd for $C_{13}H_{13}N_5O$: C, 61.17; H, 5.13; N, 27.43. Found: C, 61.00; H, 5.04; N, 27.30.

Ethylation of 5c A mixture of **5c**[10] (181 mg, 0.75 mmol), EtI (1.17 g, 7.5 mmol), and AcNMe$_2$ (10 ml) was stirred at 40 °C for 48 h and concentrated *in vacuo*. The residue was washed with Et$_2$O (5 ml) and dissolved in H_2O (40 ml). The aqueous solution was passed through a column of Amberlite IRA-402 (Cl$^-$) (2 ml), and the column was eluted with H_2O (30 ml). The eluate was concentrated *in vacuo*, and the residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH (4:1, v/v)]. Crude **4i** (88 mg) obtained from earlier fractions was recrystallized from $CHCl_3$ to provide pure **4i** (64 mg, 32%), mp 194—195.5 °C (dec.). This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **4i** described below.

A mixture (22 mg) obtained from the successive eluate of the above chromatography was purified by preparative TLC [silica gel, $CHCl_3$–MeOH–concentrated aqueous NH_3 (20:7:1, v/v)] to afford a colorless oil (4 mg, 2%). This compound was inferred to be 3-benzyl-7-ethyl-8-oxoadenine (**6i**) by comparison of its NMR spectrum [^{1}H-NMR δ: 1.06 (3H, t, J=7 Hz, MeCH$_2$), 3.90 (2H, q, J=7 Hz, MeCH$_2$), 5.30 (2H, s, PhCH$_2$), 6.78 (2H, s, NH$_2$), 7.30—7.42 (5H, m, PhCH$_2$), 8.34 [1H, s, C(2)-H]] with that of **6h**.

Further elution of the column afforded crude **5c**, which was recrystallized from H_2O to give pure **5c** (17 mg, 9% recovery). The mother liquor of this recrystallization was concentrated *in vacuo* to afford a colorless solid mixture (20 mg). The main component of this solid was presumed to be 3-benzyl-8-ethoxy-7-ethyladenine hydrochloride (**8i**·HCl), ^{1}H-NMR δ: 1.24 [3H, t, J=7 Hz, N(7)-CH$_2$Me], 1.42 (3H, t, J=7 Hz, OCH$_2$Me), 4.23 [2H, q, J=7 Hz, N(7)-CH$_2$Me], 4.65 (2H, q, J=7 Hz, OCH$_2$Me), 5.51 (2H, s, PhCH$_2$), 7.25—7.55 (5H, m, PhCH$_2$), 8.15—8.85 (2H, br, NH$_2$), 8.93 [1H, s, C(2)-H]. A small amount of this solid was heated in boiling 1 N aqueous HCl for 2 h, and the solution was concentrated after treatment with Amberlite IRA-402 (HCO_3^-). The residue was purified by preparative TLC [silica gel, $CHCl_3$–MeOH–concentrated aqueous NH_3 (20:7:1, v/v)] to afford **6i**, which was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with a sample of **6i** described above.

A colorless solid obtained from the last fraction of the above chromatography was recrystallized from H_2O to give a colorless solid (2 mg, 1%). This compound was inferred to be 3-benzyl-9-ethyl-8-oxoadenine (**7i**) by comparison of its NMR spectrum [^{1}H-NMR δ: 0.86 (3H, t, J=7 Hz, MeCH$_2$), 3.68 (2H, q, J=7 Hz, MeCH$_2$), 5.67 (2H, s, PhCH$_2$), 7.10—7.20 (2H) and 7.31—7.47 (3H) (m each, PhCH$_2$), 8.32 [1H, s, C(2)-H]] with that of **7h**.

Benzylation of 5c A mixture of **5c**[10] (171 mg, 0.71 mmol), PhCH$_2$Br (0.42 ml, 3.5 mmol), and AcNMe$_2$ (10 ml) was stirred at 40 °C for 48 h and concentrated *in vacuo*. The residue was washed with Et$_2$O (3×5 ml) and subjected to flash chromatography [$CHCl_3$–MeOH (8:1, v/v)]. A colorless oil (18 mg) obtained from earlier fractions was purified by preparative TLC [silica gel, AcOEt–EtOH (8:1, v/v)] to afford 3-benzyl-8-benzyloxyadenine (**4j**) (7 mg, 3%) as a colorless solid, mp 160—170 °C (dec.); ^{1}H-NMR δ: 5.37 and 5.42 (2H each, s, two PhCH$_2$'s), 7.23—7.48 (12H, m, NH$_2$ and two PhCH$_2$'s), 8.43 [1H, s, C(2)-H]. A portion (2 mg) of this compound was heated in 1 N aqueous HCl (2 ml) under reflux for 4 h. The resulting solution was treated with Amberlite IRA-402 (HCO_3^-) (5 ml) and concentrated *in vacuo* to give **5c**, which was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with an authentic specimen.[10]

A colorless glass (36 mg) obtained from the successive eluate of the above chromatography was recrystallized from MeOH to give 3,7-dibenzyl-8-oxoadenine hemihydrate (**6j**·1/2H$_2$O) (15 mg), mp 210—212 °C (dec.). The mother liquor of recrystallization was concentrated, and the residue was purified by preparative TLC [silica gel, $CHCl_3$–MeOH (8:1, v/v)] to provide a second crop of **6j**·1/2H$_2$O (16 mg; the total yield was 31 mg, 13%), mp 220—224 °C (dec.). Recrystallization of these samples from MeOH and drying over P_2O_5 at 2 mmHg and 100 °C for 3 h, followed by exposure to air at room temperature until a constant weight was reached afforded an analytical sample of **6j**·1/2H$_2$O as colorless prisms, mp 230—231.5 °C (dec.); MS *m/z*: 331 (M$^+$); UV $\lambda_{max}^{95\%\,EtOH}$ 240 nm (ε 18000), 302 (15400); $\lambda_{max}^{H_2O}$ (pH 1) 221 (29100), 292 (17100); $\lambda_{max}^{H_2O}$ (pH 7) 235 (19900), 298 (16700); $\lambda_{max}^{H_2O}$ (pH 13) 235 (19900), 298 (16700); IR ν_{max}^{Nujol} cm^{-1}: 3335, 3179 (OH and NH), 1647 (C=N); ^{1}H-NMR δ: 5.13 [2H, s, N(7)-CH$_2$Ph], 5.32 (2H, s, OCH$_2$Ph), 6.72 (2H, br s, NH$_2$), 7.11—7.45 (10H, m, two PhCH$_2$'s), 8.35 [1H, s, C(2)-H]. *Anal.* Calcd for $C_{19}H_{17}N_5O \cdot 1/2H_2O$: C, 67.04; H, 5.33; N, 20.57. Found: C, 66.95; H, 5.07; N, 20.65.

Further elution of the column in the above chromatography afforded a dibenzyl compound (20 mg), ^{1}H-NMR δ: 4.86 and 5.31 (2H each, s, two PhCH$_2$'s), 6.95—7.44 (10H, m, two PhCH$_2$'s), 7.72 (2H, br, NH$_2$), 8.20 [1H, s, C(2)-H]. The starting material **5c** (19 mg, 11%) was recovered from the last fraction of this chromatography.

Methylation of 4d A mixture of **4d**[10] (36.5 mg, 0.204 mmol), MeI (140 mg, 0.986 mmol), and AcNMe$_2$ (5 ml) was stirred at 30 °C for 6 h and concentrated *in vacuo*. The residue was washed with Et$_2$O (3×5 ml) and dried to afford a yellow solid (60 mg). This was subjected to preparative TLC [silica gel, $CHCl_3$–MeOH (4:1, v/v)]. The compound obtained from the highest zone as an orange solid was presumed to be 8-methoxy-3,7-dimethyladenine hydriodide (**8d**·HI) (19 mg, 29%) on the basis of its NMR spectrum [^{1}H-NMR δ: 3.75 [3H, s, N(7)-Me], 3.89 [3H, s, N(3)-Me],[20] 4.22 (3H, s, OMe), 8.37 (2H, br, NH$_2$), 8.63 [1H, s, C(2)-H]]. This sample (18 mg) was heated in 1 N aqueous HCl (2 ml) under reflux for 2 h. The solution was passed through a column packed with Amberlite IRA-402 (HCO_3^-) (5 ml), and the column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo* to afford **6d** (10.1 mg) as a colorless solid, mp 265—267 °C. This sample was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with authentic **6d** described above.

The next zone of the above preparative TLC gave a yellow solid (7.8 mg, 12%), which was inferred to be 8-methoxy-3,9-dimethyladenine hydriodide (**10d**·HI) on the basis of its NMR spectrum [^{1}H-NMR δ: 3.82 [3H, s, N(9)-Me], 4.15 [3H, s, N(3)-Me],[20] 4.18 (3H, s, OMe), 8.50 [1H, s, C(2)-H], 8.68 and 8.73 (1H, each, br, NH$_2$)]. This compound (6.8 mg) was heated in 1 N aqueous HCl (2 ml) under reflux for 2 h, and the solution was passed through a column packed with Amberlite IRA-402 (Cl$^-$) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo* to afford **7d**·HCl (3.7 mg) as a yellow solid, mp 265—267 °C (dec.). This sample was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with authentic **7d**·HCl described above.

The compound obtained from the third zone as a slightly yellow solid (10.3 mg, 28%), mp >300 °C, was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with authentic **6d** described above.

A slightly yellow solid (8.5 mg) obtained from the lowest zone was dissolved in H_2O (2 ml), and the solution was passed through a column of Amberlite IRA-402 (Cl^-) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo* to afford **11d**·HCl·H_2O (5.8 mg, 11%) as a yellow solid, mp 232—235 °C (dec.). This sample was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with authentic **11d**·HCl·H_2O described below.

Methylation of 4d Followed by Acid Hydrolysis A mixture of **4d**[10] (179 mg, 1 mmol), MeI (710 mg, 5 mmol), and $AcNMe_2$ (17 ml) was stirred at room temperature for 6 h and concentrated *in vacuo*. The oily residue was dissolved in H_2O (15 ml), and the solution was passed through a column of Amberlite IRA-402 (Cl^-) (2 ml). The column was eluted with H_2O (20 ml), and the eluate was concentrated *in vacuo*. The resulting solid residue was heated in 1 N aqueous HCl (15 ml) under reflux for 2 h. The resulting solution was neutralized with 10% aqueous Na_2CO_3 and concentrated *in vacuo*. The residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (20 : 7 : 1, v/v)] to provide crude **6d** (105 mg), mp >300 °C, and crude **11d**·HCl (150 mg), mp 248—254 °C (dec.). Crude **6d** was recrystallized from H_2O to afford pure **6d** (92 mg, 51%), mp >300 °C. This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **6d** described above.

Crude **11d**·HCl was triturated with a few drops of 5% aqueous HCl and recrystallized from 90% (v/v) aqueous EtOH to provide 3,7,9-trimethyl-8-oxoadenine hydrochloride monohydrate (**11d**·HCl·H_2O) (54 mg, 22%), mp 250—253 °C (dec.). Further recrystallization of this sample from 90% (v/v) aqueous EtOH, drying over P_2O_5 at 2 mmHg and 75 °C for 7 h, and exposure to air at room temperature until a constant weight was reached afforded an analytical sample of **11d**·HCl·H_2O as colorless plates, mp 253—254 °C (dec.); UV $\lambda_{max}^{95\%\,EtOH}$ 226 nm (ε 19300), 298 (18600); $\lambda_{max}^{H_2O}$ (pH 1) 224 (20100), 294 (19300); $\lambda_{max}^{H_2O}$ (pH 7) 224 (20300), 294 (19300); $\lambda_{max}^{H_2O}$ (pH 13) unstable; IR ν_{max}^{Nujol} cm^{-1}: 3339, 3106 (NH), 1725 (C=O), 1669 (C=N); ^{1}H-NMR δ: 3.57 [3H, s, N(7)-Me], 3.66 [3H, s, N(9)-Me], 4.13 [3H, s, N(3)-Me], 8.27 (2H, br, NH_2), 8.50 [1H, s, C(2)-H]. *Anal.* Calcd for $C_8H_{11}N_5O$·HCl·H_2O: C, 38.79; H, 5.70; N, 28.28. Found: C, 38.63; H, 5.62; N, 28.32.

Methylation of 4h Followed by Acid Hydrolysis A mixture of **4h**[10] (128 mg, 0.501 mmol), MeI (355 mg, 2.5 mmol), and $AcNMe_2$ (7 ml) was kept at 30 °C for 18 h and then concentrated *in vacuo*. The residue was dissolved in H_2O (10 ml), and the solution was passed through a column packed with Amberlite IRA-402 (Cl^-) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo*, and the residue was heated in 1 N aqueous HCl (10 ml) under reflux for 2 h. The resulting solution was neutralized with 10% aqueous Na_2CO_3 and concentrated *in vacuo*. The residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH (4 : 1 and then 2 : 1, v/v)]. A crude product obtained from earlier fractions was triturated with 5% aqueous HCl and then dried to afford **6h**·HCl (36.3 mg, 25%), mp 267—271 °C (dec.). This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **6h**·HCl described above.

3-Benzyl-7,9-dimethyl-8-oxoadenine hydrochloride (**11h**·HCl·1/5H_2O) (110 mg, 71%), mp 238—242 °C (dec.), obtained from later fractions was recrystallized three times from MeOH and dried over P_2O_5 at 2 mmHg and 75 °C for 10 h to afford an analytical sample of **11h**·HCl·1/5H_2O as colorless plates, mp 245—246 °C (dec.); UV $\lambda_{max}^{95\%\,EtOH}$ 228 nm (ε 22000), 301 (18900); $\lambda_{max}^{H_2O}$ (pH 1) 226 (22700), 297 (18500); $\lambda_{max}^{H_2O}$ (pH 7) 226 (22500), 297 (18300); $\lambda_{max}^{H_2O}$ (pH 13) unstable; IR ν_{max}^{Nujol} cm^{-1}: 3266, 3077 (NH), 1717 (C=O), 1651 (C=N); ^{1}H-NMR δ: 3.31 [3H, s, N(9)-Me], 3.57 [3H, s, N(7)-Me], 5.84 (2H, s, PhC$\underline{H}_2$), 7.19—7.25 (2H) and 7.35—7.48 (3H) (m each, $\underline{Ph}CH_2$), 8.45 (2H, br, NH_2), 8.67 [1H, s, C(2)-H]. *Anal.* Calcd for $C_{14}H_{15}N_5O$·HCl·1/5H_2O: C, 54.35; H, 5.34; N, 22.64. Found: C, 54.25; H, 5.19; N, 22.44. The water of crystallization of this sample was difficult to remove even by further drying at 2 mmHg and 100 °C for 6 h.

In a separate run, methylation of **4h** (155 mg, 0.607 mmol) with MeI (431 mg, 3.04 mmol) in $AcNMe_2$ (10 ml) was carried out at room temperature for 6 h. The reaction mixture was worked up in a manner similar to that described above, giving compounds **6h**·HCl (56 mg, 32%) and **11h**·HCl·1/5H_2O (42 mg, 22%).

Ethylation of 4d Followed by Acid Hydrolysis A mixture of **4d**[10] (179 mg, 1 mmol), EtI (780 mg, 5 mmol), and $AcNMe_2$ (20 ml) was stirred at 40 °C for 4 d. The resulting yellow solution was concentrated *in vacuo*, and the residue was washed with Et_2O (10 ml) and then heated in concentrated aqueous HCl (10 ml) under reflux for 2 h. The aqueous solution was concentrated *in vacuo*. The residue was dissolved in H_2O (10 ml), and this solution was passed through a column of Amberlite IRA-402 (Cl^-) (4 ml). The column was eluted with H_2O (30 ml). The eluate was concentrated *in vacuo*, and the residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (20 : 7 : 1, v/v)] to afford **6e** (33 mg, 17%), mp 240—248 °C (dec.); **6d** (13 mg, 7%), mp 250—270 °C (dec.); and **7d**·HCl (34 mg, 16%), mp 265—273 °C (dec.). Recrystallization of crude **6e** from MeOH afforded colorless plates, mp 295—298 °C. This product was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with an authentic specimen described below. Recrystallization of crude **6d** from H_2O provided colorless plates, mp >300 °C; this material was identical (by comparison of the ^{1}H-NMR spectrum and TLC mobility) with an authentic sample described above. Crude **7d**·HCl was recrystallized from 5% aqueous HCl to give colorless plates, mp 272—273 °C (dec.), and the product was confirmed to be identical (by comparison of the IR spectrum and TLC mobility) with an authentic sample described above.

Ethylation of 4e Followed by Acid Hydrolysis A mixture of **4e**·H_2O[10] (211 mg, 1 mmol), EtI (780 mg, 5 mmol), and $AcNMe_2$ (22 ml) was stirred at 40 °C for 4 d. The resulting yellow solution was concentrated *in vacuo*, and the residue was washed with Et_2O (10 ml) and then heated in 1 N aqueous HCl (20 ml) under reflux for 4 h. The aqueous solution was passed through a column of Amberlite IRA-402 (Cl^-) (4 ml), and the column was eluted with H_2O (30 ml). The eluate was concentrated *in vacuo*, and the solid residue was dried and subjected to flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (20 : 7 : 1, v/v)] to afford 7-ethyl-3-methyl-8-oxoadenine (**6e**) (136 mg, 70%), mp 288—295 °C (dec.). Recrystallization of this sample from MeOH afforded an analytical sample of **6e** as colorless plates, mp 295—298 °C (dec.); MS *m/z*: 193 (M^+); UV $\lambda_{max}^{95\%\,EtOH}$ 235 nm (ε 15400), 301 (17600); $\lambda_{max}^{H_2O}$ (pH 1) 219 (21800), 290 (18100); $\lambda_{max}^{H_2O}$ (pH 7) 217 (18800), 230 (16800), 296 (18300); $\lambda_{max}^{H_2O}$ (pH 13) 230 (16800), 296 (18400); IR ν_{max}^{Nujol} cm^{-1}: 3374, 3291, 3183 (NH), 1636 (C=N); ^{1}H-NMR δ: 1.06 (3H, t, J=7 Hz, $\underline{Me}CH_2$), 3.65 [3H, s, N(3)-Me], 3.91 (2H, q, J=7 Hz, MeC$\underline{H}_2$), 6.69 (2H, br s, NH_2), 8.10 [1H, s, C(2)-H]. *Anal.* Calcd for $C_8H_{11}N_5O$: C, 49.73; H, 5.74; N, 36.25. Found: C, 49.59; H, 5.66; N, 35.95.

Methylation of 6d Leading to 11d·HCl A mixture of **6d** (179 mg, 1 mmol), MeI (710 mg, 5 mmol), and $AcNMe_2$ (20 ml) was stirred at 30 °C for 24 h and concentrated *in vacuo*. The solid residue was dissolved in H_2O (12 ml), and the solution was passed through a column packed with Amberlite IRA-402 (Cl^-) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo* to leave a colorless solid (233 mg), mp 243—245 °C (dec.). The solid was recrystallized from 90% (v/v) aqueous EtOH to provide **11d**·HCl·H_2O (191 mg, 77%), mp 251—252.5 °C (dec.). This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **11d**·HCl·H_2O described above.

Methylation of 6h Leading to 11h·HCl A mixture of **6h**·HCl (60.0 mg, 0.206 mmol), MeI (143 mg, 1 mmol), anhydrous K_2CO_3 (14.2 mg, 0.103 mmol), and $AcNMe_2$ (6 ml) was stirred at 40 °C for 48 h. The resulting yellow solution was concentrated *in vacuo*, and the residue was washed with Et_2O (3 ml) and dissolved in a mixture of 10% aqueous HCl (1 ml) and H_2O (8 ml). This solution was passed through a column of Amberlite IRA-402 (Cl^-) (2 ml). The column was eluted with H_2O (30 ml), and the eluate was concentrated *in vacuo*. The residue was triturated with MeOH (1 ml), and the precipitate that resulted was collected by filtration, washed with MeOH (0.5 ml), and dried to afford **11h**·HCl·1/5H_2O (62.4 mg, 98%), mp 237—240 °C (dec.). This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **11h**·HCl·1/5H_2O described above.

Hydrogenolysis of 6h·HCl Leading to 7-Methyl-8-oxoadenine A solution of **6h**·HCl (30.9 mg, 0.106 mmol) in H_2O (6 ml) was shaken with 10% Pd–C (30 mg) under H_2 at atmospheric pressure and 20 °C for 2.5 h. The catalyst was filtered off and washed successively with hot H_2O (50 ml) and hot MeOH (10 ml). The filtrate and washings were combined and concentrated to a volume of *ca.* 20 ml. The residual solution was passed through a column of Amberlite IRA-402 (HCO_3^-) (2 ml), and the column was eluted with H_2O (50 ml). The eluate was concentrated *in vacuo* to leave 7-methyl-8-oxoadenine (16.4 mg, 94%), mp >300 °C, which was identical (by comparison of the IR spectrum and TLC mobility) with

an authentic specimen.[11]

Hydrogenolysis of 7h·HCl Leading to 13 A solution of **7h**·HCl (50.8 mg, 0.174 mmol) in a mixture of MeOH (3 ml) and H_2O (13 ml) was shaken under H_2 in the presence of 10% Pd–C (51 mg) at atmospheric pressure and room temperature for 2.5 h. The catalyst was filtered off and washed with hot H_2O (30 ml). The filtrate and washings were combined and passed through a column of Amberlite IRA-402 (HCO_3^-) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo* to leave **13** (21.9 mg, 76%) as a colorless solid, mp > 300 °C. This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **13**.[7a]

Hydrogenolysis of 11h·HCl Leading to 12 A solution of **11h**·HCl·1/5H_2O (40.9 mg, 0.132 mmol) in H_2O (10 ml) was shaken with 10% Pd–C (41 mg) under H_2 at atmospheric pressure and room temperature for 2.5 h. The catalyst was filtered off and washed with hot H_2O (20 ml). The filtrate and washings were combined and passed through a column of Amberlite IRA-402 (HCO_3^-) (2 ml). The column was eluted with H_2O (20 ml). The eluate was concentrated *in vacuo*, and the solid residue was recrystallized from H_2O to afford **12** (20.6 mg, 87%), mp 254—257 °C (dec.). This sample was identical (by comparison of the IR spectrum and TLC mobility) with authentic **12** described below.

3-Benzyl-8-ethoxyadenine (4i) A mixture of 3-benzyl-7-methoxyadenine perchlorate[10] (178 mg, 0.5 mmol) and a 0.1 M NaOEt solution in EtOH (10 ml) was stirred at 20 °C for 4 h. The resulting solution was concentrated *in vacuo*, and the residue was subjected to flash chromatography [$CHCl_3$–MeOH (8 : 1, v/v)] after neutralization with 10% aqueous HCl, giving **4i** (115 mg, 85%), mp 189—191 °C (dec.). Crude **4i** was recrystallized from MeOH to afford an analytical sample as colorless pillars, mp 194—195.5 °C (dec.); MS *m/z*: 269 (M^+); UV $\lambda_{max}^{95\%\,EtOH}$ 231 nm (sh) (ε 14300), 290 (12500); $\lambda_{max}^{H_2O}$ (pH 1) 226 (21100), 281 (19600); $\lambda_{max}^{H_2O}$ (pH 7) 227 (15300), 286 (13100); $\lambda_{max}^{H_2O}$ (pH 13) 227 (15000), 286 (12900); IR ν_{max}^{Nujol} cm^{-1}: 3316, 3175 (NH), 1651 (C=N); ^{1}H-NMR δ: 1.31 (3H, t, *J* = 7 Hz, MeCH$_2$), 4.32 (2H, q, *J* = 7 Hz, MeCH$_2$), 5.40 (2H, s, PhCH$_2$), 7.26—7.46 (7H, m, PhCH$_2$ and NH_2), 8.41 [1H, s, C(2)-H]. *Anal.* Calcd for $C_{14}H_{15}N_5O$: C, 62.44; H, 5.61; N, 26.01. Found: C, 62.31; H, 5.54; N, 25.92.

7,9-Dimethyl-8-oxoadenine (12) A suspension of **13**[7a] (330 mg, 2 mmol) and anhydrous K_2CO_3 (276 mg, 2 mmol) in $HCONMe_2$ (6 ml) was stirred at 100—110 °C for 1 h and cooled to room temperature. A solution of MeI (570 mg, 4.02 mmol) in $HCONMe_2$ (4 ml) was added to the mixture, and the whole was stirred at room temperature for 2 h. The reaction mixture was concentrated *in vacuo*, and the residue was neutralized with 10% aqueous HCl after addition of a small volume of H_2O. The insoluble solid that resulted was collected by filtration, washed with EtOH (1 ml), dried, and purified by flash chromatography [$CHCl_3$–MeOH–concentrated aqueous NH_3 (40 : 7 : 1, v/v)] to afford **12** (111 mg, 31%), mp 267—269 °C (dec.), and **13** (47 mg, 14% recovery). Recrystallization of crude **12** from H_2O afforded an analytical sample as colorless pillars, mp 269—272 °C (dec.); MS *m/z*: 179 (M^+); UV $\lambda_{max}^{95\%\,EtOH}$ 273 nm (ε 12000); $\lambda_{max}^{H_2O}$ (pH 1) 220 (25500), 279 (10600); $\lambda_{max}^{H_2O}$ (pH 7) 213 (35400), 273 (13200); $\lambda_{max}^{H_2O}$ (pH 13) 273 (13300); IR ν_{max}^{Nujol} cm^{-1}: 3460, 3298 (NH), 1693 (C=O), 1647 (C=N); ^{1}H-NMR δ: 3.24 [3H, s, N(9)-Me], 3.48 [3H, s, N(7)-Me], 6.57 (2H, br s, NH_2), 8.02 [1H, s, C(2)-H]. *Anal.* Calcd for $C_7H_9N_5O$: C, 46.92; H, 5.06; N, 39.09. Found: C, 46.80; H, 5.02; N, 39.13.

References and Notes

1) Paper LXXV in this series, Itaya T., Ito N., Kanai T., Fujii T., *Chem. Pharm. Bull.*, **45**, 832—841 (1997).
2) Cimino G., De Giulio A., De Rosa S., De Stefano S., Puliti R., Mattia C. A., Mazzarella L., *J. Nat. Prod.*, **48**, 523—528 (1985).
3) Zelnik R., Haraguchi M., Matida A. K., Lavie D., Frolow F., Weis A. L., *J. Chem. Soc., Perkin Trans. 1*, **1986**, 2051—2053.
4) *a*) Uramoto M., Kim C.-J., Shin-ya K., Kusakabe H., Isono K., Phillips D. R., McCloskey J. A., *J. Antibiot.*, **44**, 375—381 (1991); *b*) Matsuura N., Onose R., Osada H., *ibid.*, **49**, 361—365 (1996).
5) Kang H., Fenical W., *Tetrahedron Lett.*, **38**, 941—944 (1997).
6) Kamiya H., Miura H., Murata-Kamiya N., Ishikawa H., Sakaguchi T., Inoue H., Sasaki T., Masutani C., Hanaoka F., Nishimura S., Ohtsuka E., *Nucleic Acids Res.*, **23**, 2893—2899 (1995).
7) *a*) Fujii T., Saito T., Mori S., *Chem. Pharm. Bull.*, **38**, 2146—2150 (1990); *b*) Saito T., Mori S., Chikazawa J., Kanai T., Fujii T., *ibid.*, **41**, 1746—1752 (1993); *c*) Fujii T., Saito T., Mori S., *ibid.*, **38**, 2591—2594 (1990); *d*) Saito T., Kanai T., Fujii T., *ibid.*, **41**, 1850—1852 (1993).
8) *a*) Robins R. K., *J. Am. Chem. Soc.*, **80**, 6671—6679 (1958); *b*) Fujii T., Saito T., *Chem. Pharm. Bull.*, **21**, 1954—1959 (1973).
9) Holmes R. E., Robins R. K., *J. Am. Chem. Soc.*, **87**, 1772—1776 (1965).
10) Itaya T., Takada Y., Fujii T., *Chem. Pharm. Bull.*, **44**, 2025—2032 (1996).
11) Itaya T., Takada Y., Kanai T., Fujii T., *Chem. Pharm. Bull.*, **44**, 2318—2321 (1996).
12) Fujii T., Itaya T., Saito T., Mohri K., Kawanishi M., Nakasaka T., *Chem. Pharm. Bull.*, **37**, 1504—1513 (1989).
13) Rizkalla B. H., Robins R. K., Broom A. D., *Biochim. Biophys. Acta*, **195**, 285—293 (1969).
14) Fujii T., Itaya T., *Chem. Pharm. Bull.*, **19**, 1611—1617 (1971).
15) *a*) Fujii T., *Yakugaku Zasshi*, **116**, 355—373 (1996); *b*) Fujii T., Itaya T., *Rev. Heteroat. Chem.*, **16**, 257—285 (1997).
16) Still W. C., Kahn M., Mitra A., *J. Org. Chem.*, **43**, 2923—2925 (1978).
17) Determined in a manner similar to that described previously: Fujii T., Itaya T., Saito T., *Chem. Pharm. Bull.*, **23**, 54—61 (1975).
18) The molecular absorption coefficient was not determined because of the poor solubility of this compound.
19) The chemical shift of the C(2)-H proton was variable. We could not identify the factor creating such variability.
20) Assigned by comparison of the peak height of this signal with those of other methyl signals: Itaya T., Ito N., Fujii T., *Chem. Pharm. Bull.*, **44**, 594—598 (1996).