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Purines. XXV.[1] Fission and Reclosure of the Adenine Ring by the Use of Modified Benzyloxy Groups at the 1-Position[2]

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The rates of the three reactions in the system that produces the rearranged isomers **4a**—**d** and the 5-aminoimidazole derivatives **6a**—**d** from 1-benzyloxy- and 1-(modified benzyloxy)-9-methyladenine perchlorates (**2a**—**d**·$HClO_4$) through the isolable ring-opened intermediates **3a**—**d** have been determined at various pH's and ionic strength 1.0 at 40 °C. Among four kinds of modified or unmodified benzyloxy groups in these compounds, the 4-nitrobenzyloxy group has been found to cause **2**·$HClO_4$ to open its adenine ring most rapidly, whereas it causes **3** to recyclize to **4** most slowly. The reaction of 9-methyladenine 1-oxide (**5**) with 4-nitrobenzyl bromide in $AcNMe_2$ at room temperature and subsequent treatment of the product with $NaClO_4$ produced 1-(4-nitrobenzyloxy)-9-methyladenine perchlorate (**2a**·$HClO_4$) in 91% yield. Treatment of **2a**·$HClO_4$ with boiling pyridine gave **5** in 84% yield, and reduction of **2a**·$HClO_4$ with hydrogen and Pd–C afforded 9-methyladenine (**1**) in 72% yield, demonstrating the ease with which the 4-nitrobenzyl group can be removed. These results suggest the potential utility of the 4-nitrobenzyloxy group as a control synthon for the preparation of compounds of type **3**, which are useful monocyclic intermediates for syntheses of a variety of substituted adenine derivatives.

Keywords——1-benzyloxyadenine; adenine ring-opening; *N*′-alkoxy-5-formamidoimidazole-4-carboxamidine; deformylation; Dimroth rearrangement rate study; 4-nitrobenzyloxy control synthon; catalytic hydrogenolysis; adenine 1-oxide *O*-benzylation; pyridine nucleophilic debenzylation

Introduction

The introduction of an alkyl group into the adenine ring at the 1-position usually facilitates base-catalyzed ring-opening and subsequent recyclization in the pyrimidine moiety, a process known as the Dimroth rearrangement whereby the apparent migration of the alkyl group from the endocyclic nitrogen ($N_{(1)}$) to the exocyclic nitrogen (N^6) takes place.[1,3,4] It has been found in this laboratory that replacement of the 1-alkyl group by an electron-withdrawing alkoxy group (type **2**) accelerates the ring-opening step and retards the recyclization step, rendering it possible to isolate the ring-opened intermediate (type **3**) through which the rearranged product (type **4**) and the deformylated derivative (type **6**) are obtainable (Chart 1).[3c,4a,b,5] Considering the benzyloxy group to be more strongly electron-withdrawing than a simple alkoxy group, we tried to investigate kinetically the effect of a modified benzyloxy group at the 1-position with a view to finding a suitable alkoxy group for high-yield isolation and utilization of the ring-opened intermediates (type **3**). Specific illustrations of the synthetic utility of such intermediates have been afforded by new syntheses of 1,9-dimethyladenine,[4b] 3,9-dialkyladenines,[6] 3-methyladenosine,[7] 3-methyl-2′-deoxyadenosine,[8] 7,9-dialkyladenines,[9] 5-(alkylamino)-1-β-D-ribofuranosylimidazole-4-carboxamides,[10] and 2-azaadenosine,[11] which had previously been difficult to prepare, in most cases.

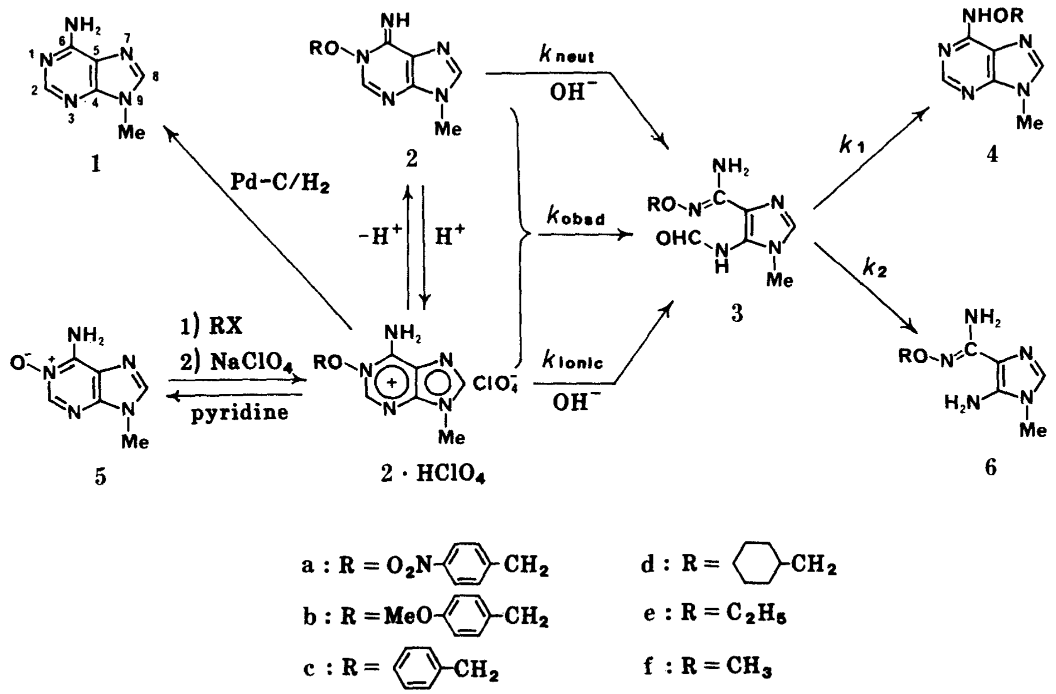

Chart 1

Results

Synthesis of Compounds

The substrates selected for the present kinetic study were the perchlorate salts of **2a**—**d**, which all possess the methyl group at the 9-position for uniformity, but carry the usual or a modified benzyloxy group at the 1-position. The perchlorate **2c**·$HClO_4$ was prepared from the *N*-oxide **5** and benzyl bromide according to the previously reported procedure[5d] (Chart 1). Similar alkylations of **5** with 4-nitrobenzyl bromide, 4-methoxybenzyl bromide, and cyclohexylmethyl iodide and subsequent treatment of the products with $NaClO_4$ furnished the perchlorates **2a**, **b**, **d**·$HClO_4$ in 91, 74, and 61% yields, respectively.

Conversions of **2a**, **b**, **d**·$HClO_4$ into **3a**, **b**, **d**, **4a**, **d**, and **6a**, **d** were achieved by methods similar to those employed previously[5d] for the conversions of **2c**·$HClO_4$ into **3c**, **4c**, and **6c**, respectively. Since similar direct conversions of **2b**·$HClO_4$ into **4b** and **6b** were unsatisfactory owing to competitive solvolysis to form the *N*-oxide **5**, **4b** and **6b** were separately prepared from **3b** as in the previous syntheses[5a] of the ethyl homologues of **4e** and **6e** from the 1-ethyl homologue of **3e**. Table I includes the acid dissociation constants of **2a**—**d**·$HClO_4$ and **3a**—**d** thus obtained.

Treatment of **2a**·$HClO_4$ with boiling pyridine for 1 h gave the *N*-oxide **5** in 84% yield. Reduction of **2a**·$HClO_4$ with hydrogen and Pd–C afforded 9-methyladenine (**1**) in 72% yield. These results are in general agreement with the chemical behavior observed[12] for 1-benzyloxyadenine derivatives.

Kinetic Study

The system of reactions that produces **4a**—**d** and **6a**—**d** from **2a**—**d**·$HClO_4$ through **3a**—**d** is represented in Chart 1. The rates of the three reactions (**2**·$HClO_4$→**3**; **4**←**3**→**6**) in H_2O at 40 °C and various pH's were followed by ultraviolet (UV) spectrophotometry. Since the ring-opening step was faster than the subsequent steps, it was possible to treat the former separately from the latter. In a typical experiment, ring-opening of **2a**·$HClO_4$ in six different 0.02 M buffer solutions of pH 7.00 to 11.20 at ionic strength 1.0 at 40 °C was followed by

Table I. Acid Dissociation Constants of **2** and **3**

Compound	pK_a at 40 °C and ionic strength 1.0			
	Basic	Analytical wavelength (nm)	Acidic	Analytical wavelength (nm)
2a·$HClO_4$	7.76±0.10	263	—	—
2b·$HClO_4$	8.40±0.03	300	—	—
2c·$HClO_4$	8.31±0.01[a]	300	—	—
2d·$HClO_4$	8.45±0.01	300	—	—
2e·$HClO_4$	8.50±0.04[b]	300	—	—
2f·$HClO_4$	8.48±0.06[b]	300	—	—
3a	3.24±0.06	254	10.59±0.07	271
3b	3.51±0.11	254	10.55±0.09	261
3c	3.67±0.09[b]	254	10.60±0.06[b]	260
3d	3.94±0.07	254	10.61±0.05	260
3e	3.91±0.11[b]	254	10.42±0.06[b]	256
3f	3.78±0.08[b]	255	10.36±0.04[b]	258

a) Reported previously[5d] to be 8.30±0.06.
b) Taken from ref. 5*d*.

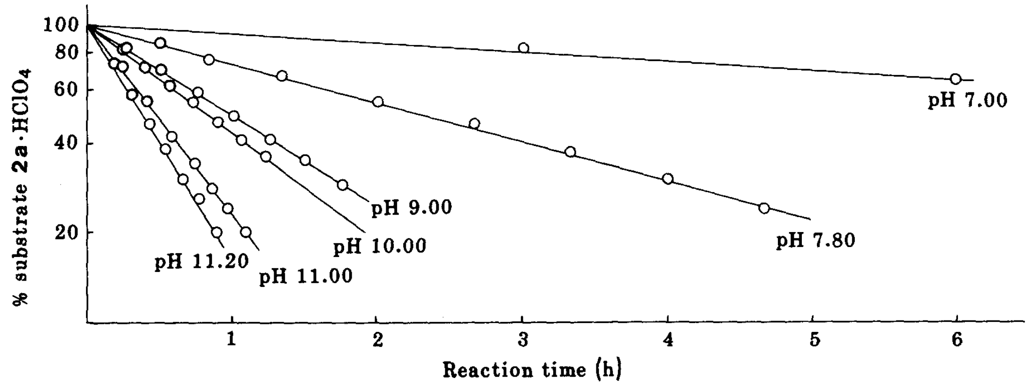

Fig. 1. First-Order Plot for Ring-Opening of **2a**·$HClO_4$ to **3a** in 0.02 M Buffer Solution at Various pH's and Ionic Strength 1.0 at 40 °C

Table II. Rates of Ring-Opening of **2a—d**·$HClO_4$ in 0.02 M Buffer Solution at 40 °C and Ionic Strength 1.0

Substrate	Pseudo-first-order rate constant, $k_{obsd} \times 10^3$ (min^{-1}) pH value						
	7.00	7.50	7.80	9.00	10.00	11.00	11.20
2a·$HClO_4$	2.4	—	5.1	12	14	25	32
2b·$HClO_4$	—	—	—	—	—	18	25
2c·$HClO_4$	0.73	—	3.4	12	15	19	24
2d·$HClO_4$	—	1.4	2.4	10	14	18	20

measuring the decrease in UV absorption at 262 nm which occurs on formation of **3a**. The semilogarithmic plots of mole fractions of the residual substrate (**2a**·$HClO_4$) against time, illustrated in Fig. 1, indicate that the reaction obeyed fairly good pseudo-first-order kinetics at all pH's. Kinetic runs with the other substrates (**2b—d**·$HClO_4$) were also treated in a similar manner, giving typical first-order plots at various pH's. Table II lists the rate constants (k_{obsd})

obtained from these plots. In the case of the 4-methoxybenzyloxy derivative **2b**·$HClO_4$, solvolysis of the 4-methoxybenzyl group occurred competitively at pH 10 and below to produce the *N*-oxide **5**, and this complexity actually hampered the kinetic study over a wide range of pH. It may be seen that in all cases the rate of ring-opening increases with increasing pH of the reaction medium. Since we have already confirmed that the catalytic coefficients of the buffer components are small in this type of reaction,[4c, 5d] the observed rate constants may be regarded, as a good approximation, as the limiting rate constants for zero buffer concentration.

Figure 2 shows a pH–rate profile obtained by plotting the rate constants for ring-opening of **2d**·$HClO_4$ as a function of pH. As in previous, similar cases,[4c,5d] a theoretical pH–rate profile was calculated from eq. (1), where v is the reaction rate;

$$v = k_{\mathrm{obsd}}[\mathbf{2d}]_{\mathrm{total}} = k_{\mathrm{ionic}}[\mathbf{2d}\cdot \mathrm{H}^+][\mathrm{OH}^-] + k_{\mathrm{neut}}[\mathbf{2d}][\mathrm{OH}^-] \qquad (1)$$

$[\mathbf{2d}]_{\mathrm{total}}$, the total concentration of **2d**; $[\mathbf{2d}\cdot\mathrm{H}^+]$, the concentration of the protonated species of **2d**; [**2d**], the concentration of the neutral species of **2d**; $[\mathrm{OH}^-]$, hydroxide ion concentration; k_{obsd}, the observed pseudo-first-order rate constant; k_{ionic} and k_{neut}, rate constants for hydroxide attack on the protonated and the neutral species; the pK_a value of **2d** is 8.45 (Table I). The theoretical pH–rate profile corresponds to the curve plotted in Fig. 2 if $k_{\mathrm{ionic}} = 1600$ and

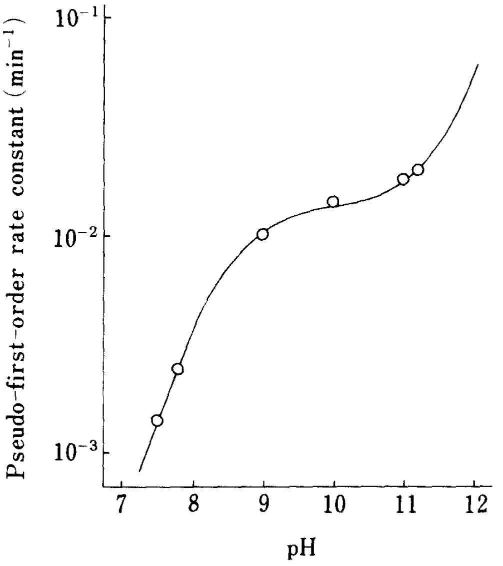

Fig. 2. pH–Rate Profile for Ring-Opening of **2d**·$HClO_4$ to **3d** at 40 °C and Ionic Strength 1.0

TABLE III. Effect of Alkoxy Groups on Ring-Opening of the Protonated and Neutral Species of 1-Alkoxy-9-methyladenines (**2**) at 40 °C and Ionic Strength 1.0

Substrate	Second-order rate constant ($M^{-1}\,min^{-1}$)	
	k_{ionic}	k_{neut}
1-(4-Nitrobenzyloxy)-9-methyladenine (**2a**)	7500	4.1
1-Benzyloxy-9-methyladenine (**2c**)	2300[a]	2.3[a]
1-(Cyclohexylmethoxy)-9-methyladenine (**2d**)	1600	1.4
1-Ethoxy-9-methyladenine (**2e**)[b]	1700	2.3
1-Methoxy-9-methyladenine (**2f**)[b]	1900	3.4

a) The value is in agreement with that reported.[5d]
b) Taken from ref. 5*d*.

Table IV. Rate Constants, k_1 and k_2, for Recyclization and Deformylation of **3a**—**d** in 0.005 M Buffer Solution at 40 °C and Ionic Strength 1.0

Substrate	Step	Pseudo-first-order rate constant (min^{-1}) pH value						
		6.20	7.00	7.50	9.00	10.00	10.70	11.20[a]
3a	$k_1 \times 10^5$	1.9	1.3[b]	1.0[b]	6.6	16	66	110
	$k_2 \times 10^5$	0.14			0.98	6.7	27	100
3b	$k_1 \times 10^5$	5.4	2.6	2.7	8.2	24	69	93
	$k_2 \times 10^5$	0.28	0.20	0.20	1.5	7.2	34	89
3c	$k_1 \times 10^5$	3.7	2.3	2.2	6.8	20	71	120
	$k_2 \times 10^5$	0.20	0.15	0.14	1.0	5.7	35	94
3d	$k_1 \times 10^5$	5.7	3.2	2.9	9.4	22	74	110
	$k_2 \times 10^5$	0.64	0.36	0.40	1.7	6.7	33	100

a) The data at this pH were obtained by extrapolation of the plot of the rate constant *vs.* buffer concentration to 0.005 M buffer concentration, using data obtained at 0.1 M, 0.05 M, and 0.02 M buffer concentration.

b) The value corresponds to $(k_1+k_2)\times 10^5$.

$k_{\text{neut}}=1.4$ (time in minutes). A similar treatment of the pseudo-first-order rate constants for **2a**, **c**·$HClO_4$ afforded the second-order rate constants listed in Table III. In the case of **2b**·$HClO_4$, however, the data in Table II were insufficient to permit the same procedure.

Next we determined the rates of recyclization (**3**→**4**) and simultaneous deformylation (**3**→**6**) of the ring-opened intermediates **3a**—**d** in H_2O at 40 °C and various pH's. The quantitative analysis of the three-component system in each kinetic run was performed in a manner similar to that reported previously.[5d] The results are summarized in Table IV.

Discussion

It may be seen from Table I that the cyclohexylmethoxy and the *n*-alkyloxy analogues (**2d**—**f**) are the strongest bases among the bicyclic derivatives listed and their $\mathrm{p}K_a$'s are closely similar, reflecting the small differences in the electron-withdrawing properties of their alkoxy groups. On the other hand, the observed decrease in base strength of the 1-benzyloxy and the 1-(4-nitrobenzyloxy) derivatives (**2c**, **a**) is attributable to the electron-withdrawing nature of the benzyl and the 4-nitrobenzyl groups relative to a *n*-alkyl group. Interestingly, the 4-methoxybenzyloxy derivative **2b** shows a $\mathrm{p}K_a$ value slightly higher than that of the unsubstituted-benzyloxy derivative **2c**, reflecting the electron-donating nature of the 4-methoxy group.

The ring-opened intermediates **3a**—**f** all show both basic and acidic $\mathrm{p}K_a$'s (Table I). The basic $\mathrm{p}K_a$ is obviously due to the amidine moiety, and the lowered $\mathrm{p}K_a$'s observed for the benzyloxy-type compounds can be ascribed to the electron-withdrawing effect of the N'-substituents. In the case of the 4-methoxybenzyloxy derivative **3b**, however, the observed basic $\mathrm{p}K_a$ value might not be accurate because the compound is unstable in the acid region. On the other hand, the acidic $\mathrm{p}K_a$ is probably due to the formamido group at the 5-position,[5d] and remarkable is the strong acidity of **3a**—**f**, which is almost comparable to that ($\mathrm{p}K_a$ 9.98)[13] of trichloroacetanilide. This unusually strong acidity of the formamido group may be attributed to the electron-withdrawing character of the attached imidazole ring[14] and to stabilization of the conjugate base anion by distribution of the negative charge over the conjugated system extended by participation of the amidoxime moiety.[5d] The aralkyloxy group or the cyclohexylmethoxy group in the amidoxime moiety raises the acidic $\mathrm{p}K_a$ to a

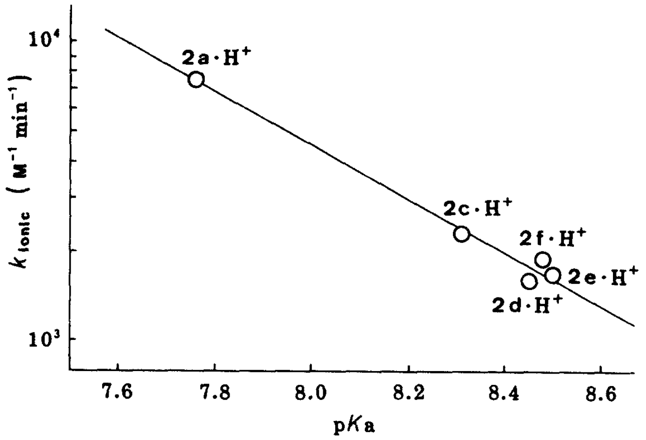

Fig. 3. pK_a-Rate Profile for Ring-Opening of the Protonated Species of **2a**, **c**—**f** to **3a**, **c**—**f** at 40 °C and Ionic Strength 1.0

small extent. This is probably caused by the bulky aralkyl or cyclohexylmethyl group, which may destabilize the conjugate base by disturbing the coplanarity of the two side-chains at the 4- and 5-positions.

The results shown in Fig. 2 and Table III indicate that compounds **2a**, **c**, **d**·$HClO_4$ undergo ring-opening to give the corresponding monocyclic intermediates **3a**, **c**, **d** according to the rate law given by eq. (1). Ring-opening of the 4-methoxybenzyloxy analogue **2b**·$HClO_4$ probably proceeds in the same manner, but it is complicated by the accompanying solvolysis of the 4-methoxybenzyloxy group to form 9-methyladenine 1-oxide (**5**) at pH 10 and below. Table III also indicates that attack of hydroxide ion on the protonated species (**2**·H^+) is much faster than that on the neutral species (**2**). The former attack seems to be influenced by an electronic factor of the 1-alkoxy group, whereas the latter attack, by both a steric and an electronic factor. Figure 3 shows the semilogarithmic plots of the second-order rate constants for the protonated species of **2a**, **c**—**f** against pK_a. It may be seen from the linear relation that the rate increases with decreasing pK_a. These results are in general agreement with our previous findings.[4a,5d] It is noteworthy that the 4-nitrobenzyloxy analogue **2a** opens its pyrimidine ring in both the protonated and neutral species most rapidly among the analogues listed in Table III.

It may be seen from Table IV that both the recyclization of **3a**—**d** to give **4a**—**d** and the simultaneous deformylation to give **6a**—**d** are slowest at near neutrality and become faster as the pH of the reaction medium is increased in all cases. The reaction around pH 6 seems to be suitable for preferential recyclization, whereas the reaction in the high pH region is favorable for the deformylation. At near neutrality, the cyclohexylmethoxy analogue **3d** cyclizes most rapidly, whereas the 4-nitrobenzyloxy analogue **3a** cyclizes most slowly among the substrates tested. The rate of the recyclization reaction seems to decrease with increasing electron-withdrawing ability of the *N′*-alkoxy group, which probably reduces the electron density of the amidoxime moiety in **3**. However, the data available in the present study are insufficient to discuss in detail the mechanisms of the above two competitive reactions.

Knowing the rate constants for all three reactions in the system that produces **4** and **6** from **2**·$HClO_4$ through **3** (Chart 1), we can calculate the maximum concentration of the ring-opened intermediate **3** in the same manner as described previously.[5d] Table V lists the theoretical maximum yields and the required times thus calculated for **3a**, **c**, **f**, which all carry a practically applicable alkoxy group. It may be seen that for isolation of the ring-opened intermediate the use of the 4-nitrobenzyloxy group is superior to the use of the other alkoxy groups, since it produces the desired product in excellent yield at near neutrality in the shortest time.

In addition, the 4-nitrobenzyloxy group is readily introduced into the adenine ring at the

Table V. Theoretical Maximum Yield of the Ring-Opened Intermediate (**3**) in the Reaction of **2**·$HClO_4$ in 0.005 M Buffer Solution at 40 °C and Ionic Strength 1.0

Substrate		Reaction conditions		Ring-opened intermediate	
No.	R	pH	Time (min)	No.	Maximum yield (%)
2a·$HClO_4$	4-$O_2NC_6H_4CH_2$	7.00	2200	**3a**	97
		7.50	1400	**3a**	99
		9.00	420	**3a**	96
2c·$HClO_4$	$C_6H_5CH_2$	7.00	4800	**3c**	89
		7.50	2300	**3c**	93
		9.00	420	**3c**	96
2f·$HClO_4$	CH_3	7.00	5000	**3f**	81[a)]
		7.50	2100	**3f**	89[a)]
		9.00	350	**3f**	95[a)]

a) Calculated by adopting the previously reported rate constants.[5d)]

1-position and can be removed easily by the reactions exemplified in Chart 1. Thus, the present study provides a firm basis on which this group may be used as a convenient control synthon for chemical modification of adenine derivatives and nucleic acids under mild reaction conditions.

Experimental

General Notes——All melting points were determined by using a Yamato MP-1 capillary melting point apparatus and are corrected. See refs. 4*c* and 5*d* for details of other instrumentation and measurements. Microanalyses were performed by Mr. Y. Itatani and his associates at Kanazawa University. The following abbreviations are used: br = broad, d = doublet, m = multiplet, s = singlet, sh = shoulder.

Materials——Analytically pure samples of 1-benzyloxy-9-methyladenine perchlorate (**2c**·$HClO_4$), *N′*-benzyloxy-5-formamido-1-methylimidazole-4-carboxamidine (**3c**), N^6-benzyloxy-9-methyladenine (**4c**), and 5-amino-*N′*-benzyloxy-1-methylimidazole-4-carboxamidine (**6c**) were prepared according to the previously reported procedure.[5d)] Other compounds were obtained as described below.

9-Methyl-1-(4-nitrobenzyloxy)adenine Perchlorate (2a·$HClO_4$)——A mixture of 9-methyladenine 1-oxide (**5**)[12a,15)] (1.65 g, 10 mmol) and 4-nitrobenzyl bromide (10.8 g, 50 mmol) in $AcNMe_2$ (60 ml) was stirred at room temperature for 4.5 h. The precipitate that resulted was filtered off, washed successively with EtOH and ether, and dried to give **2a**·HBr as a solid. The solid was dissolved in hot H_2O (150 ml), and 24% (w/v) aqueous $NaClO_4$ (20 ml) was added while hot. The resulting mixture was kept in a refrigerator, and the precipitate that formed was collected by filtration, washed successively with H_2O, EtOH, and ether, and dried to furnish **2a**·$HClO_4$ (3.64 g, 91%), mp 206 °C (dec.). Recrystallization from 50% (v/v) aqueous EtOH yielded an analytical sample as colorless needles, mp 217 °C (dec.); pK_a (Table I); UV $\lambda_{max}^{95\%\ aq.\ EtOH}$ 261 nm (ε 21900); $\lambda_{max}^{H_2O}$ (pH 1) 262 (21600); $\lambda_{max}^{H_2O}$ (pH 7) 262 (21400); $\lambda_{max}^{H_2O}$ (pH 13) (unstable) 260 (*ca.* 20000), 266 (*ca.* 20000); nuclear magnetic resonance (NMR) (Me_2SO-d_6) δ: 3.85 (3H, s, NMe), 5.54 (2H, s, $ArC\underline{H}_2O$), 7.92 and 8.29 (4H, AB type d's, J = 9 Hz, aromatic protons), 8.47 and 9.00 (1H each, s, purine protons), 9.96 (br, NH). *Anal.* Calcd for $C_{13}H_{13}ClN_6O_7$: C, 38.96; H, 3.27; N, 20.97. Found: C, 38.93; H, 3.38; N, 21.22.

1-(4-Methoxybenzyloxy)-9-methyladenine Perchlorate (2b·$HClO_4$)——A mixture of **5**[12a,15)] (3.30 g, 20 mmol) and 4-methoxybenzyl bromide[16)] (20.0 g, 99 mmol) in $AcNMe_2$ (80 ml) was stirred at room temperature for 16.5 h. The precipitate that resulted was filtered off and washed with ether (40 ml) to afford a first crop of **2b**·HBr. Addition of ether (80 ml) to the above filtrate yielded a second crop of **2b**·HBr. The first and second crops of **2b**·HBr were combined and dissolved in H_2O (840 ml), and 26% (w/v) aqueous $NaClO_4$ (40 ml) was added. The crystals that deposited were filtered off, washed successively with H_2O, EtOH, and ether, and dried to give **2b**·$HClO_4$ (5.70 g, 74%), mp 150 °C (dec.). Recrystallization from EtOH gave an analytical sample as colorless needles, mp 150 °C (dec.);

pK_a (Table I); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 233 nm (sh) (ε 16400), 260 (13800); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 260 (13100); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 260 (12800); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 13) 259 (12700), 265 (sh), (11800); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.78 (3H, s, OMe), 3.84 (3H, s, NMe), 5.35 (2H, s, $\mathrm{ArC\underline{H}_2O}$), 6.94 and 7.49 (4H, AB type d's, $J=9$ Hz, aromatic protons), 8.43 and 8.76 (1H each, s, purine protons), 9.5—10.3 (br, NH). *Anal.* Calcd for $\mathrm{C_{14}H_{16}ClN_5O_6}$: C, 43.59; H, 4.18; N, 18.16. Found: C, 43.30; H, 4.16; N, 18.46.

1-(Cyclohexylmethoxy)-9-methyladenine Perchlorate (2d · $\mathrm{HClO_4}$)——A mixture of **5**[12a,15] (825 mg, 5 mmol) and cyclohexylmethyl iodide (2.85 g, 12.7 mmol) in $\mathrm{AcNMe_2}$ (30 ml) was stirred under an atmosphere of nitrogen at 78—80 °C for 15 h. The reaction mixture was worked up in a manner similar to that described above for **2b** · $\mathrm{HClO_4}$, giving crude **2d** · $\mathrm{HClO_4}$ (1.11 g, 61%), mp 239—241 °C (dec.). For analysis, it was recrystallized from EtOH to produce colorless scales, mp 246—246.5 °C (dec.); pK_a (Table I); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 261 nm (ε 12100); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 261 (11900); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 261 (12000); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 13) 258 (12500), 265 (sh) (11400); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.86 (3H, s, NMe), 4.19 (2H, d, $J=7$ Hz, $\mathrm{C_6H_{11}C\underline{H}_2O}$), 8.49 and 8.98 (1H each, s, purine protons), 9.82 (br, NH). *Anal.* Calcd for $\mathrm{C_{13}H_{20}ClN_5O_5}$: C, 43.16; H, 5.57; N, 19.36. Found: C, 43.13; H, 5.54; N, 19.61.

5-Formamido-1-methyl-*N*′-(4-nitrobenzyloxy)imidazole-4-carboxamidine (3a)——A suspension of **2a** · $\mathrm{HClO_4}$ (401 mg, 1 mmol) in 0.5 M phosphate buffer (pH 6.7) (300 ml) was stirred at 42 °C for 48 h. The crystals that resulted were filtered off, washed with $\mathrm{H_2O}$, and dried to give **3a** (280 mg, 88%), mp 198—199 °C (dec.). Recrystallization from 70% (v/v) aqueous EtOH produced a pure sample as yellowish needles, mp 198.5—199.5 °C (dec.); pK_a (Table I); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 266 nm (ε 14700); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 267 (14300); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 216 (17900), 268 (11900); $\lambda_{\max}^{\mathrm{H_2O}}$(pH 13) 271 (18900); IR $\nu_{\max}^{\mathrm{CHCl_3}}$ (0.005 M solution) $\mathrm{cm^{-1}}$: 3508 ($\mathrm{NH_2}$), 3394 ($\mathrm{NH_2}$ and CONH), 1709 (CONHAr); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.42 and 3.45 (s each, NMe), 5.00 and 5.06 (s each, $\mathrm{ArC\underline{H}_2O}$), 5.83 (2H, s, $\mathrm{NH_2}$), 7.60 (1H, s, $\mathrm{C_{(2)}}$-H), 7.63 and 8.17 (4H, AB type d's, $J=9$ Hz, aromatic protons), 7.98 (d, $J=11$ Hz, HCON, *trans*-**3a**), 8.17 (s, HCON, *cis*-**3a**), 9.34 (d, $J=11$ Hz, CONH, *trans*-**3a**), 9.53 (CONH, *cis*-**3a**).[17] *Anal.* Calcd for $\mathrm{C_{13}H_{14}N_6O_4}$: C, 49.05; H, 4.43; N, 26.41. Found: C, 49.32; H, 4.63; N, 26.59.

5-Formamido-*N*′-(4-methoxybenzyloxy)-1-methylimidazole-4-carboxamidine (3b)——A stirred suspension of **2b** · $\mathrm{HClO_4}$ (1.54 g, 40 mmol) in 0.5 M carbonate buffer (pH 9.5) (160 ml) was kept at 40 °C for 4 h. The reaction mixture was worked up as described above for **3a**, affording crude **3b** (779 mg, 64%), mp 161—163 °C. Recrystallization from EtOH provided an analytical sample as colorless pillars, mp 170.5—171 °C; pK_a (Table I); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 226 nm (ε 21900), 260 (sh) (7800); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 226 (17900), 252 (sh) (9100); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 225 (20500), 254 (sh) (7500); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 13) 262 (13200); IR $\nu_{\max}^{\mathrm{CHCl_3}}$ (0.005 M solution) $\mathrm{cm^{-1}}$: 3504 ($\mathrm{NH_2}$), 3392 ($\mathrm{NH_2}$ and CONH), 1705 (CONHAr); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.42 and 3.46 (s each, NMe), 3.73 (3H, s, OMe), 4.79 and 4.85 (s each, $\mathrm{ArC\underline{H}_2O}$), 5.58 (2H, s, $\mathrm{NH_2}$), 6.86 and 7.32 (4H, AB type d's, $J=9$ Hz, aromatic protons), 7.57 (1H, s, $\mathrm{C_{(2)}}$-H), 8.08 (d, $J=10$ Hz, HCON, *trans*-**3b**), 8.22 (s, HCON, *cis*-**3b**), 9.38 (d, $J=10$ Hz, CONH, *trans*-**3b**), 9.55 (CONH, *cis*-**3b**).[17] *Anal.* Calcd for $\mathrm{C_{14}H_{17}N_5O_3}$: C, 55.43; H, 5.65; N, 23.09. Found: C, 55.69; H, 5.66; N, 23.01.

***N*′-(Cyclohexylmethoxy)-5-formamido-1-methylimidazole-4-carboxamidine (3d)**——A suspension of **2d** · $\mathrm{HClO_4}$ (1.34 g, 3.7 mmol) in 0.5 M carbonate buffer (pH 9.5) (130 ml) was stirred at 40 °C for 6.5 h. The crystals that resulted were filtered off, washed with $\mathrm{H_2O}$, and dried to give **3d** (810 mg, 78%), mp 145.5—147.5 °C. Recrystallization from 25% (v/v) aqueous EtOH yielded a pure sample as colorless prisms, mp 148—149 °C; pK_a (Table I); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 224 nm (ε 13600), 254 (sh) (6900); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 255 (7700); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 220 (12000), 249 (sh) (6900); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 13) 257 (11000); IR $\nu_{\max}^{\mathrm{CHCl_3}}$ (0.005 M solution) $\mathrm{cm^{-1}}$: 3500 ($\mathrm{NH_2}$), 3390 ($\mathrm{NH_2}$ and CONH), 1705 (CONHAr); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.50 and 3.54 (s each, NMe), 3.70 (m, $\mathrm{C_6H_{11}C\underline{H}_2O}$), 5.54 (2H, s, $\mathrm{NH_2}$), 7.62 (1H, s, $\mathrm{C_{(2)}}$-H), 8.10 (d, $J=11$ Hz, HCON, *trans*-**3d**), 8.22 (s, HCON, *cis*-**3d**), 9.35 (d, $J=11$ Hz, CONH, *trans*-**3d**), 9.53 (CONH, *cis*-**3d**).[17] *Anal.* Calcd for $\mathrm{C_{13}H_{21}N_5O_2}$: C, 55.89; H, 7.58; N, 25.07. Found: C, 55.75; H, 7.49; N, 25.21.

9-Methyl-*N*^{6}-(4-nitrobenzyloxy)adenine (4a)——A solution of **2a** · $\mathrm{HClO_4}$ (401 mg, 1 mmol) in a mixture of $\mathrm{HCONMe_2}$ (50 ml) and 0.5 M phosphate buffer (pH 6.7) (150 ml) was heated under reflux for 28 h. After cooling, the crystals that resulted were filtered off, washed with $\mathrm{H_2O}$ and dried to afford **4a** (178 mg, 59%), mp 242.5—243.5 °C (dec.). On recrystallization from EtOH, it produced an analytical sample as yellowish prisms, mp 242.5—243.5 °C (dec.); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 272 nm (ε 19400); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 276 (19900); $\lambda_{\max}^{\mathrm{H_2O}}$(pH 7) 272 (22600); $\lambda_{\max}^{\mathrm{H_2O}}$(pH 13) 286 (20100); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.67 (3H, s, NMe), 5.20 (2H, s, $\mathrm{ArC\underline{H}_2O}$), 7.65 and 7.70 (1H each, s, purine protons), 7.69 and 8.19 (4H, AB type d's, $J=9$ Hz, aromatic protons), 11.28 (br, NH). *Anal.* Calcd for $\mathrm{C_{13}H_{12}N_6O_3}$: C, 52.00; H, 4.03; N, 27.99. Found: C, 51.95; H, 4.08; N, 27.82.

***N*^{6}-(4-Methoxybenzyloxy)-9-methyladenine (4b)**——A solution of **3b** (455 mg, 1.5 mmol) in 0.5 M phosphate buffer (pH 6.7) (90 ml) was heated under reflux for 2 h. After cooling, the precipitate that resulted was filtered off, washed with $\mathrm{H_2O}$, and recrystallized from 70% (v/v) aqueous EtOH to yield **4b** (303 mg, 71%), mp 238 °C (dec.). Further recrystallizations in the same way gave an analytical sample as slightly brownish needles, mp 238—238.5 °C (dec.); UV $\lambda_{\max}^{95\%\ \mathrm{aq.\ EtOH}}$ 272 nm (ε 17300); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 1) 273 (16400); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 7) 272 (17800); $\lambda_{\max}^{\mathrm{H_2O}}$ (pH 13) 282 (13600); NMR ($\mathrm{Me_2SO}$-d_6) δ: 3.64 (3H, s, NMe), 3.73 (3H, s, OMe), 4.92 (2H, s, $\mathrm{ArC\underline{H}_2O}$), 6.88 and 7.36 (4H, AB type d's, $J=9$ Hz, aromatic protons), 7.70 and 7.83 (1H each, s, purine protons), 10.91 (br, NH). *Anal.* Calcd for $\mathrm{C_{14}H_{15}N_5O_2}$: C, 58.93; H, 5.30; N, 24.55. Found: C, 58.99; H, 5.34; N, 24.35.

***N*^{6}-(Cyclohexylmethoxy)-9-methyladenine (4d)**——A solution of **2d** · $\mathrm{HClO_4}$ (543 mg, 1.5 mmol) in $\mathrm{H_2O}$ (150 ml) was passed through a column packed with Amberlite IRA-410 ($\mathrm{HCO_3^-}$) (5 ml), and the column was further eluted with $\mathrm{H_2O}$. The eluate (250 ml) was concentrated *in vacuo* to a volume of *ca.* 30 ml and then heated under reflux for 5 h.

After cooling, the crystals that resulted were filtered off, washed with H_2O, and dried to give **4d** (263 mg, 67%), mp 198.5—200.5 °C (dec.). Recrystallization from EtOH gave an analytical sample as colorless prisms, mp 201—202 °C (dec.); UV $\lambda_{\max}^{95\%\ \text{aq. EtOH}}$ 270 nm (ε 14000); $\lambda_{\max}^{H_2O}$(pH 1) 271 (14100); $\lambda_{\max}^{H_2O}$ (pH 7) 270 (14800); $\lambda_{\max}^{H_2O}$ (pH 13) 285 (11200); NMR (Me_2SO-d_6) δ: 3.67 (3H, s, NMe), 3.74 (2H, d, J=6 Hz, $C_6H_{11}C\underline{H}_2O$), 7.66 and 7.78 (1H each, s, purine protons), 10.83 (br, NH). *Anal.* Calcd for $C_{13}H_{19}N_5O$: C, 59.75; H, 7.33; N, 26.80. Found: C, 59.85; H, 7.37; N, 26.83.

5-Amino-1-methyl-*N*′-(4-nitrobenzyloxy)imidazole-4-carboxamidine (6a)——A solution of **2a**·$HClO_4$ (200 mg, 0.5 mmol) and NaOH (800 mg) in H_2O (16 ml) was stirred at 48—50 °C for 2 h. The crystals that deposited were filtered off, washed with H_2O, and recrystallized from EtOH to afford **6a** (101 mg, 70%) as orange scales, mp 192—193 °C (dec.); UV $\lambda_{\max}^{95\%\ \text{aq. EtOH}}$ 269 nm (ε 19600); $\lambda_{\max}^{H_2O}$ (pH 1) 275 (18000); $\lambda_{\max}^{H_2O}$(pH 7) 270 (18200); $\lambda_{\max}^{H_2O}$ (pH 13) 270 (18200); NMR (Me_2SO-d_6) δ: 3.40 (3H, s, NMe); 5.08 (2H, s, $ArC\underline{H}_2O$), 5.18 (2H, NH_2), 5.72 (2H, NH_2), 7.14 (1H, s, $C_{(2)}$–H), 7.71 and 8.22 (4H, AB type d's, J=9 Hz, aromatic protons). *Anal.* Calcd for $C_{12}H_{14}N_6O_3$: C, 49.65; H, 4.86; N, 28.95. Found: C, 49.56; H, 5.02; N, 28.72.

Similar alkaline hydrolysis of **3a** at 50 °C for 45 min also produced **6a** in 77% yield.

5-Amino-*N*′-(4-methoxybenzyloxy)-1-methylimidazole-4-carboxamidine (6b)——A solution of **3b** (182 mg, 0.6 mmol) and NaOH (900 mg) in H_2O (18 ml) was heated under reflux for 15 min. The crystals that resulted were filtered off, washed with H_2O, and dried to give **6b** (138 mg, 84%), mp 162—165 °C. Recrystallization from EtOH yielded an analytical sample as slightly pinkish prisms, mp 165—167 °C; UV $\lambda_{\max}^{95\%\ \text{aq. EtOH}}$ 268 nm (ε 13500); $\lambda_{\max}^{H_2O}$ (pH 1) 228 (16600), 278 (10300); $\lambda_{\max}^{H_2O}$ (pH 7) 225 (20100), 266 (11200); $\lambda_{\max}^{H_2O}$ (pH 13) 266 (11200); NMR (Me_2SO-d_6) δ: 3.41 (3H, s, NMe), 3.77 (3H, s, OMe), 4.82 (2H, s, $ArC\underline{H}_2O$), 5.22 (2H, NH_2), 5.45 (2H, NH_2), 6.85 and 7.26 (4H, AB type d's, J=9 Hz, aromatic protons), 7.03 (1H, s, $C_{(2)}$–H). *Anal.* Calcd for $C_{13}H_{17}N_5O_2$: C, 56.71; H, 6.22; N, 25.44. Found: C, 56.47; H, 6.33; N, 25.57.

5-Amino-*N*′-(cyclohexylmethoxy)-1-methylimidazole-4-carboxamidine (6d)——A solution of **2d**·$HClO_4$ (544 mg, 1.5 mmol) and NaOH (402 mg) in H_2O (15 ml) was heated under reflux for 1 h. The reaction mixture was then neutralized with 10% aqueous HCl and kept in a refrigerator. The crystals that formed were filtered off, washed with H_2O, and recrystallized from 50% (v/v) aqueous EtOH to furnish **6d** (176 mg, 47%), mp 148.5—150 °C (dec.). Repeated recrystallization in the same way gave a pure sample as pinkish prisms, mp 150.5—151 °C (dec.); UV $\lambda_{\max}^{95\%\ \text{aq. EtOH}}$ 220 nm (ε 10700), 263 (11400); $\lambda_{\max}^{H_2O}$ (pH 1) 225 (7600), 280 (9500); $\lambda_{\max}^{H_2O}$ (pH 7) 221 (10700), 263 (9800); $\lambda_{\max}^{H_2O}$ (pH 13) 263 (9800); NMR (Me_2SO-d_6) δ: 3.39 (3H, s, NMe), 3.64 (2H, d, J=6 Hz, $C_6H_{11}C\underline{H}_2O$), 5.22 (2H, NH_2), 5.35 (2H, NH_2), 7.05 (1H, s, $C_{(2)}$–H). *Anal.* Calcd for $C_{12}H_{21}N_5O$: C, 57.34; H, 8.42; N, 27.87. Found: C, 57.52; H, 8.28; N, 27.64.

Reaction of 2a·$HClO_4$ with Pyridine——A mixture of **2a**·$HClO_4$ (1.20 g, 3 mmol) and pyridine (30 ml) was heated under reflux for 1 h. The crystals that resulted were filtered off, washed successively with H_2O, EtOH, and ether, and dried over P_2O_5 at 110 °C and 2 mmHg for 5.5 h to yield **5** (277 mg, 57%) as a colorless solid, mp 293.5—294 °C (dec.), which was identical with an authentic sample.[12a,15] The filtrate and washings were combined and concentrated to dryness *in vacuo*. The residue was triturated with hot EtOH (100 ml), an insoluble solid [1-(4-nitrobenzyl)pyridinium perchlorate, mp 183—185 °C (lit.[18] mp 181—182 °C)] (329 mg, 35%) removed by filtration, and the ethanolic filtrate concentrated *in vacuo* to dryness. The residue was dissolved in H_2O (110 ml), the aqueous solution passed through a column of Amberlite CG-120 (Na^+) (20 ml), and the column eluted with H_2O. Concentration of the eluate (360 ml) under reduced pressure left a semisolid, which was triturated with EtOH (10 ml) to produce an insoluble solid. The solid was collected by filtration, washed with H_2O, and dried over P_2O_5 as described above to yield a second crop of **5** (140 mg, 28%). The total yield of **5** was 417 mg (84%).

Hydrogenolysis of 2a·$HClO_4$——A solution of **2a**·$HClO_4$ (601 mg, 1.5 mmol) in 50% (v/v) aqueous EtOH (200 ml) was hydrogenated over 10% Pd–C (600 mg) at atmospheric pressure and room temperature for 8 h. The catalyst was removed by filtration and washed successively with H_2O and EtOH. The filtrate and washings were combined and concentrated *in vacuo* after addition of 1 N aqueous HCl (50 ml). The residue was triturated with EtOH (5 ml) and an insoluble solid was filtered off, washed with a little EtOH, and dissolved in H_2O (10 ml). The resulting aqueous solution was passed through a column of Amberlite IRA-410 (HCO_3^-) (3 ml), and the column was eluted with H_2O. Concentration of the eluate (100 ml) under reduced pressure left a colorless solid (162 mg, 72%), mp 305 °C, which was identical with authentic 9-methyladenine (**1**).[12b,19]

The presence of *p*-toluidine in the above reaction mixture was confirmed by gas-liquid chromatographic analysis.

Determination of Acid Dissociation Constants——The pK_a's of **2a**—**d**·$HClO_4$ and **3a**, **b**, **d** at 40 °C and ionic strength 1.0 were determined in a manner similar to that described previously.[4c,5d] The results are included in Table I.

Kinetic Procedure——The rates of ring-opening of **2a**—**d**·$HClO_4$ to **3a**—**d** in 0.02 M buffer solution and those of recyclization of **3a**—**d** to **4a**—**d** and deformylation of **3a**—**d** to **6a**—**d** in 0.005 M buffer solution were measured at 40 °C and ionic strength 1.0. The procedure was similar to that employed previously.[5d] The results are summarized in the text.

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