

## Platelet Aggregation Inhibitors. 2. Inhibition of Platelet Aggregation by 5'-, 2-, 6-, and 8-Substituted Adenosines<sup>1</sup>

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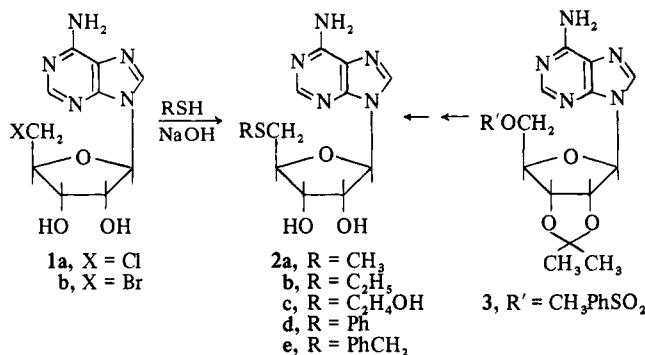
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5'-Alkyl(or aryl)thio-5'-deoxyadenosines (**2**) were synthesized by treating 5'-chloro-5'-deoxyadenosine (**1a**) with the corresponding mercaptans. 2-Amino- and/or 6-substituted adenosines (**5-7**) were synthesized by treating 6-chloroinosine (**4a**) or 6-chloroguanosine derivatives (**4c, d**) with amines and subsequent treatments. The inhibitory activities of these 5'-, 2-, and/or 6-substituted adenosines and a variety of 8-substituted adenosines (**8-11**) were tested against ADP-induced rabbit platelet aggregation. Of these nucleosides, 6-hydroxyaminopurine riboside (**5a**) and 2-amino-6-hydroxyaminopurine riboside (**5b**) showed 5-10 times the potency of adenosine. 2-Aminoadenosine (**5c**) showed the same inhibitory activity as adenosine. Incubation of **5a** and **5b** with rabbit plasma for a period of 40 min did not lead to loss of their activities. **5a** and **5b** also inhibited collagen-induced platelet aggregation and platelet adhesiveness to glass beads.

Blood platelets have been known to function in the formation of thrombi and plug,<sup>2</sup> and the key role of adenosine diphosphate (ADP) as an important initiator of platelet aggregation has been well documented.<sup>3</sup> Thus, agents that will inhibit ADP-induced platelet aggregation are of interest as potential drugs.

A variety of compounds<sup>2,4</sup> closely related to ADP, including adenosine,<sup>5,6</sup> have been known as inhibitors of ADP-induced platelet aggregation. Among the derivatives of adenosine which have been investigated as inhibitors of ADP-induced platelet aggregation,<sup>7-14</sup> 2-chloroadenosine<sup>8,9</sup> and 5'-adamantoyladenine<sup>10</sup> have been found to have 1-3 times the potency of adenosine. We now wish to report the convenient synthesis of 5'-, 2-, and/or 6-substituted adenosines and the strong inhibitory activity of hydroxylated adenosines on platelet aggregation.

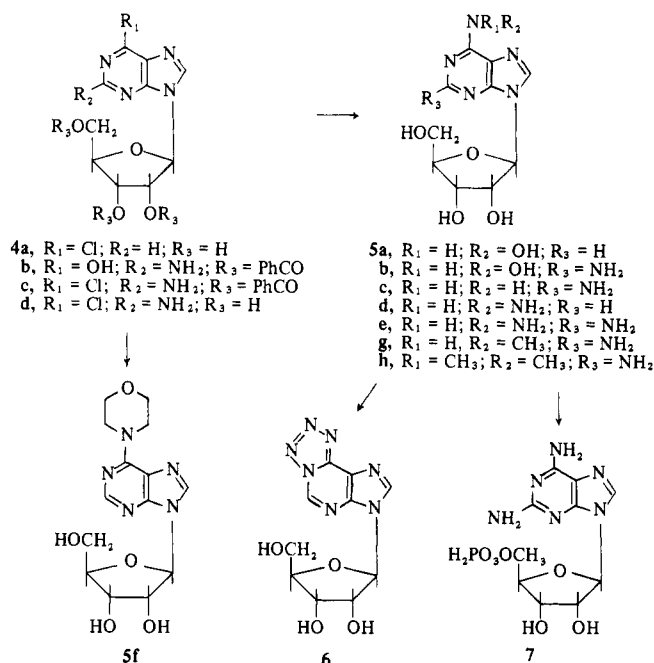
**Chemistry.** 5'-Alkyl(or aryl)thio-5'-deoxyadenosines (**2a-e**) were readily synthesized in a yield of 50-70% from 5'-chloro-5'-deoxyadenosine (**1a**)<sup>15</sup> by reaction with alkyl(or aryl)mercaptans in the presence of aqueous NaOH. The



structure of 5'-methylthio-5'-deoxyadenosine (**2a**) obtained was confirmed by comparison with an authentic sample<sup>16</sup> prepared from 2',3',5'-tri-*O*-isopropylidene-5'-tosyladenosine (**3**). Several 5'-alkyl(or aryl)thio-5'-deoxyadenosines have been synthesized from **3**,<sup>16,17</sup> but the preparation of **3** involves tedious steps because of its easy transformation into an *N*<sub>3</sub>,5'-cycloadenosine derivative.<sup>18</sup> Thus, the method described above might be a readily accessible route for the preparation of 5'-alkyl(or aryl)thio-5'-deoxyadenosines, since **1a** can be easily prepared from adenosine.<sup>15</sup>

6-Hydroxyaminopurine riboside (**5a**) was obtained from 6-chloroinosine (**4a**) according to the method of Giner-Sorolla, *et al.*<sup>19</sup> 2',3',5'-Tri-*O*-benzoyl-6-chloroguanosine (**4c**) obtained by chlorination of 2',3',5'-tri-*O*-benzoyl-guanosine (**4b**) was treated with NH<sub>3</sub> at room temp and

afforded 6-chloroguanosine (**4d**) (60% from **4b**) which had been prepared previously from 2',3',5'-tri-*O*-acetylguanosine



by Gerster, *et al.*<sup>20</sup> Treatment of **4d** with hydroxylamine gave 2-amino-6-hydroxyaminopurine riboside (**5b**)<sup>21,22</sup> quantitatively. Since NH<sub>2</sub>OH has been found by us<sup>1</sup> to be a potent inhibitor of platelet aggregation, it was necessary to recrystallize **5a** and **5b** several times to remove any NH<sub>2</sub>OH remaining in products to be used for pharmacological assay. When **4c** was treated with NH<sub>3</sub> at 100°, 2-aminoadenosine (**5c**)<sup>21,23-26</sup> was obtained in a yield of 55% from **4b**. 6-Hydrazinopurine riboside (**5d**)<sup>27</sup> and 2-amino-6-hydrazinopurine riboside (**5e**)<sup>21</sup> were obtained quantitatively by reaction of **4a** and **4d** with aqueous hydrazine, respectively. Other derivatives of adenosine (**5f-h**) were obtained from **4a** or **4d** by reaction with appropriate amines. Direct phosphorylation of **5c** according to the method of Yoshikawa, *et al.*,<sup>28</sup> gave 2-aminoadenosine 5'-monophosphate (**7**).

**Biology.** The 5'-, 2-, and/or 6-, and 8-substituted adenosines were tested as inhibitors of ADP-induced rabbit platelet aggregation *in vitro* according to the method of Born and Cross.<sup>29</sup> The inhibitory activity of every compound was estimated by the extent of the decrease in the optical density after the addition of ADP, and is listed in Tables I-III.

None of the 5'-substituted adenosines (**1, 2**) and the 8-

**Table I.** Inhibition of Platelet Aggregation by 5'-Substituted Adenosines

No.	Molar concentration, <sup>b</sup> × 10 <sup>-4</sup>	Inhibition % of ADP-induced aggregation
1a <sup>a</sup>	5	0
1b <sup>a</sup>	5	0
2a	5	0
2b	5	15
2c	5	12
2d	<0.5	0
2e	5	15
Adenosine	1	45

<sup>a</sup>Kikugawa and Ichino.<sup>15</sup> <sup>b</sup>Concn given as less than (<) indicates insol compd.

**Table II.** Inhibition of Platelet Aggregation by 2- and/or 6-Substituted Adenosines

No.	Molar concentration, × 10 <sup>-4</sup>	Inhibition of aggregation					
		ADP-induced		Collagen-induced		Thrombin-induced	
		%	Rp <sup>d</sup>	%	Rp <sup>d</sup>	%	Rp <sup>d</sup>
5a <sup>a</sup>	1	87	5-10	100	10	86	0.1
	0.5	82				80	
	0.1	45		51			
5b	1	85	5-10		10	90	0.1
	0.5	73		100		76	
	0.1	58		59			
5c	1	73	1-2	100	1-2	80	0.05
	0.5	53		24		68	
	0.25	40					
5d	1	9					
5e	1	8					
5f	1	0					
5g	1	30					
5h <sup>b</sup>	1	0					
6 <sup>c</sup>	1	4					
7	1	4					
Adenosine	1	53	1	46	1	100	1
2-Chloro-adenosine	1	59	1-2				

<sup>a</sup>Giner-Sorolla, et al.<sup>19</sup> <sup>b</sup>Gerster, et al.<sup>20</sup> <sup>c</sup>Johnson, et al.<sup>27</sup> <sup>d</sup>Rp is the relative potency of the compd compared with adenosine.

substituted adenosines (8-11) showed significant inhibitory activities (Tables I, III). Among the 2- and/or 6-substituted adenosines (5-7), 6-hydroxyaminopurine riboside (5a) and 2-amino-6-hydroxyaminopurine riboside (5b) showed 5-10 times the potency of adenosine, and 2-aminoadenosine (5c) showed the same potency of adenosine. The inhibitory activities of 5a and 5b were greater than the activity of 2-chloroadenosine that has been the most powerful inhibitor among the adenosine derivatives<sup>8</sup> (Table II).

Compounds (5a-c) were incubated with plasma over the longer intervals before the addition of ADP (Figure 1). Inhibitory effects of 5a-c increased for 5 to 10 min, and then decreased as in the case of adenosine. Although 5c lost its activity completely after the interval of 60 min, as in the case of adenosine, the inhibitory activities of 5a and 5b did not greatly diminish over the incubation interval of 60 min. It has been reported that the loss of the inhibitory activity of adenosine is due to the instability of the nucleoside to plasma adenosine deaminase.<sup>8</sup> The more powerful inhibitor, 2-chloroadenosine, does not lead to loss of its activity when it is incubated with plasma for a period of up to 40 min, because it is not deaminated in plasma.<sup>8</sup> Retention of the inhibitory activities of 5a and 5b when they were incubated with plasma must be due to their stabilities to adenosine deaminase.

Our recent findings<sup>1</sup> show that hydroxylamine has about

**Table III.** Inhibition of Platelet Aggregation by 8-Substituted Adenosine Derivatives<sup>a</sup>

No.	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	X	Molar concentration, × 10 <sup>-4</sup>	Inhibition % of ADP-induced aggregation
8a	NH <sub>2</sub>	OH	H		1	6
8b	NH <sub>2</sub>	SH	H		1	0
8c	NH <sub>2</sub>	Br	H		1	8
8d	NH <sub>2</sub>	N <sub>3</sub>	H		1	9
8e	NH <sub>2</sub>	NH <sub>2</sub>	CH <sub>3</sub> CO		1	0
8f	CH <sub>3</sub> NH	Br	H		1	7
8g	(CH <sub>3</sub> ) <sub>2</sub> N	Br	H		1	0
9				S	1	18
10				S	1	27
11a				S	1	0
11b				O	1	10
Adenosine					1	40

<sup>a</sup>Compds 8-11 were gifts of Professor M. Ikehara.

10 times the inhibitory activity of adenosine against platelet aggregation, and the N or O substitutions of hydroxylamine greatly reduce the inhibitory activity of NH<sub>2</sub>OH. Compounds 5a and b were N-substituted derivatives of NH<sub>2</sub>OH and had the same potent activity as NH<sub>2</sub>OH. The inhibitory profiles of 5a and 5b, however, were different from the inhibitory profile of NH<sub>2</sub>OH. Thus, the inhibitory activity of NH<sub>2</sub>OH decreases the longer the interval of incubation with plasma and the loss of activity is complete after 40 min,<sup>1</sup> whereas the inhibitory activities of 5a and 5b increased for 5-10 min and then slightly decreased but were maintained over 40 min (Figure 1). The difference of activity between free NH<sub>2</sub>OH and 5a and b showed that the inhibition by 5a and 5b was not due to NH<sub>2</sub>OH contamination in 5a and 5b or NH<sub>2</sub>OH production by hydrolysis of 5a and 5b.

The inhibitory effects of 5a-c against collagen- and thrombin-induced platelet aggregation were tested. The inhibitory

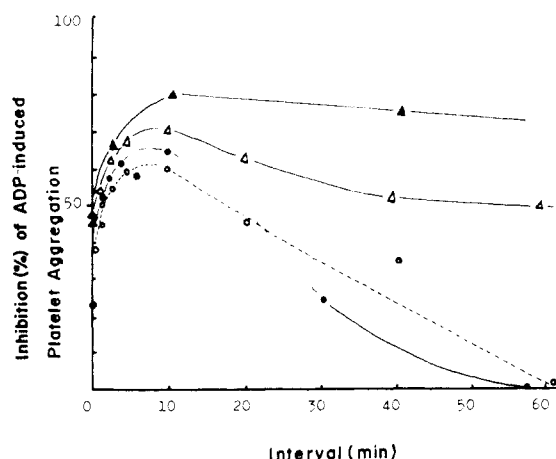


Figure 1. Inhibition of ADP-induced platelet aggregation brought about by adding 6-hydroxyaminopurine riboside (5a) ( $0.25 \times 10^{-4} M$ ),  $\Delta$ — $\Delta$ ; 2-amino-6-hydroxyaminopurine riboside (5b) ( $0.25 \times 10^{-4} M$ ),  $\circ$ — $\circ$ ; 2-aminoadenosine (5c) ( $10^{-4} M$ ),  $\Delta$ — $\Delta$ ; and adenosine ( $10^{-4} M$ ),  $\circ$ — $\circ$  at increasing intervals of time before the addition of ADP ( $10^{-5} M$ ).

**Table IV.** Inhibition of Platelet Adhesiveness by 6-Hydroxyaminopurine Riboside (**5a**), 2-Amino-6-hydroxyaminopurine Riboside (**5b**), and 2-Aminoadenosine (**5c**)

No.	PRCP		Whole blood	
	Adhesiveness,	Inhibition (%)	Adhesiveness,	Inhibition (%)
Control	0.53		0.31	
<b>5a</b>	0.04	92	0	100
<b>5b</b>	0.04	92	0	100
<b>5c</b>	0.14	74	0	100
Adenosine	0.38	28	0.06	81
2-Chloro-adenosine	0.06	89	0	100

activities of **5a** and **5b** against collagen-induced platelet aggregation were 10 times the potency of adenosine. In the case of thrombin-induced aggregation the activities of **5a** and **5b** were lower than that of adenosine (Table II).

Decrease of adhesiveness of platelets to glass beads is known with drugs that interfere with platelet function and/or coagulation mechanism.<sup>30</sup> **5a-c** were tested against rabbit platelet adhesiveness to glass beads according to the method of Hellem<sup>30</sup> (Table IV). The inhibitory activities of **5a** and **5b** were greater than those of adenosine and 2-chloroadenosine, and the activity of **5c** was greater than that of adenosine.

In conclusion, the hydroxylated adenosine derivatives (**5a, b**) were the most potent inhibitors of ADP- and collagen-induced platelet aggregation and platelet adhesiveness to glass beads among the adenosine derivatives known.<sup>7-14</sup> 2-Aminoadenosine (**5c**) was also an inhibitor of platelet aggregation and adhesiveness. Born, *et al.*,<sup>9</sup> have already tested the inhibitory activities of **5a** and **5c** against human platelet aggregation mediated by ADP, but have not recognized any significant activities. The discrepancies might be attributed to the differences of platelet sources, rabbit and human, although there have been no significant differences observed in the inhibitions of platelet aggregation by adenosine and related compounds between rabbit and human platelets.<sup>8,31</sup>

## Experimental Section†‡

**Biological Methods and Materials.** ADP(Na<sub>2</sub>), collagen (bovine achilles tendon) and thrombin were purchased from Sigma Chemical Co., Ltd., Tokyo Chemical Industry Co., Ltd., and Sankyo Co., Ltd., respectively. All glassware coming into contact with the whole blood or PRCP was siliconized using a 10% soln of Shin Etsu Silicone-KC 88 in petr ether. Glass beads (diameter, 0.5 mm) were the products of Tokyo Sugitoh Optical and Precision Glass Co., Ltd.

**Platelet-Rich Citrated Plasma (PRCP).** Male rabbits (weight, 2-3 kg) were anesthetized with Et<sub>2</sub>O. The carotid artery was carefully cut and cannulated with a 20-cm piece of siliconized polyethylene tubing. The first 5-ml portion of blood was discarded, and about 60-90 ml portion of blood was transferred into siliconized polyethylene centrifuge tubes which contained 0.1 vol of 3.8% Na citrate. The blood was centrifuged at 1000 rpm for 10 min. The supernatant PRCP (20-40 ml) was removed with a siliconized pipet, stored at room temp in a siliconized tube and used within 10 hr. The PRCP contained 6-8 × 10<sup>8</sup> platelets per ml.

**Platelet Aggregation.** The rate and extent of platelet aggregation were measured by the optical density method of Born and Cross<sup>29</sup> by using Evans EEL 169 platelet aggregation meter.

A cuvette contg 1.0 ml of PRCP and (for controls) 10 μl of saline or (for test compds) 10 μl of a soln of the compd in saline was placed in the aggregation meter and allowed to incubate at 37°

for 3 min. At this point, the PRCP was challenged with 10 μl of a soln of ADP, thrombin, or 100 μl of a soln of collagen in saline. The final concn of ADP, collagen, and thrombin required for aggregation was 10<sup>-5</sup> M, 0.53 mg/ml and 0.3 unit/ml, respectively. The % inhibition of aggregation by a test compd was calcd by dividing the max deflection in the optical density curve in the presence of the compd by that observed in the control, then multiplying by 100. As the sensitivity of platelets to aggregating agents varies from prepn to prepn and by storing period of PRCP, an arbitrarily chosen adenosine as a reference standard was tested in every PRCP prepn almost simultaneously with the test compds and the control. The results were listed in Tables I-III. The molar concn of adenosine was divided by the molar concn of the test compd required to exhibit the same extent of inhibition as adenosine. The results were listed in Table II as the relative potency (*Rp*) value.

**Incubation Effects with Plasma.** A cuvette contg 1.0 ml of PRCP and 10 μl of a soln of the compd in saline was placed in the aggregation meter and allowed to incubate at 37° up to 60 min and the PRCP was challenged with 10 μl of a soln of ADP (10<sup>-5</sup> M). Per cent inhibition of ADP-induced platelet aggregation was plotted vs. incubation interval (Figure 1).

**Platelet Adhesiveness to Glass Beads.** The extent of platelet adhesiveness was measured according to the method of Hellem.<sup>30</sup> About a 7-cm piece of siliconized Tygon R flexible plastic tubing (diam, 3 mm) with siliconized Saran filter cloth F 201 at one end and a siliconized glass funnel at the other end was packed with 0.72 g of glass beads. A tube contg 0.8 ml of PRCP and 0.1 ml of saline (for control) or 0.1 ml of a soln of the test compd (10<sup>-3</sup> M) in saline was kept at room temp for 10 min. After it was added with 0.1 ml of a soln of ADP (10<sup>-6</sup> M) in saline, an aliquot (0.5 ml) of the mixt was immediately passed through the glass beads column during 20 sec, and the platelets were counted as soon as possible. Adhesiveness of platelets was expressed by  $(A - B)/A$ ; *A* is the count of platelets before passing through the column, and *B* is the count of platelets after passing through the column. The % inhibition of the test compd was expressed by  $((C - D)/C) \times 100$ ; where *C* means the adhesiveness of control, and *D* means the adhesiveness with the test compd.

A tube contg 0.9 ml of citrated whole blood and 0.1 ml of a soln of a test compd (10<sup>-3</sup> M) in saline was kept at room temp. An aliquot (0.5 ml) was immediately passed through the column during 50 sec. Adhesiveness and % inhibition were estimated similarly as in the case of PRCP (Table IV).

**Chemical Methods and Materials.** 5'-Alkyl(or aryl)thio-5'-deoxyadenosines (**2a-e**). 5'-Chloro-5'-deoxyadenosine (**1a**)<sup>15</sup> (3.0 g) was dissolved in a mixt of 24 ml of 2 N NaOH and 4.2 ml of 20% soln of NaSCH<sub>3</sub> in H<sub>2</sub>O. The mixt was heated at 80° for 1 hr, then acidified with AcOH. The product was filtered off and dried *in vacuo*. 5'-Methylthio-5'-deoxyadenosine (**2a**) (2.1 g, 70%) was obtd as needles, mp 205-206° dec. Recrystn from H<sub>2</sub>O afforded pure **2a**, mp 211-212° dec, mmp with an authentic sample<sup>16</sup> prepd from **3** (mp 205-207° dec) showed no depression.

Other 5'-alkyl(or aryl)thio-5'-deoxyadenosines (**2b-e**) were similarly prepd by reaction of **1a** with the corresponding mercaptans.

**6-Chloroguanosine (4d).** 2',3',5'-Tri-*O*-benzoylguanosine (**4b**)<sup>32</sup> (20 g) was added with stirring to a soln of POCl<sub>3</sub> (30 ml) and *N,N'*-diethylaniline (8 ml) at room temp. The mixt was heated to reflux at 150° for 3.5 min. Excess POCl<sub>3</sub> was evapd *in vacuo*. The resulting syrup was poured over excess ice with stirring. Using two 500-ml portions of CHCl<sub>3</sub>, the aqueous mixt was extd, and the combined exts were washed with H<sub>2</sub>O until neutral to pH paper. After drying the CHCl<sub>3</sub> ext (Na<sub>2</sub>SO<sub>4</sub>), the solvent was evapd to leave a mass of 2',3',5'-tri-*O*-benzoyl-6-chloroguanosine (**4c**) (24 g). **4c** (12 g) was

**Table V.** 5'-Alkyl (or aryl)thio-5'-deoxyadenosines

5'-Deoxy-adenosine	Reagent	Yield, %	Mp, °C	Anal.
5'-Methylthio- ( <b>2a</b> )	MeSNa- 2 N NaOH	70	211-212	(C <sub>11</sub> H <sub>11</sub> O <sub>3</sub> N <sub>5</sub> S), C, H, N
5'-Ethylthio- ( <b>2b</b> )	EtSH-2 N NaOH	61	187-188	(C <sub>12</sub> H <sub>13</sub> O <sub>3</sub> N <sub>5</sub> S), C, H, N
5'-Hydroxyethylthio- ( <b>2c</b> )	HOCH <sub>2</sub> CH <sub>2</sub> SH- 2 N NaOH	51	192	(C <sub>12</sub> H <sub>13</sub> O <sub>4</sub> N <sub>5</sub> S), C, H, N
5'-Phenylthio- ( <b>2d</b> )	PhSH-2 N NaOH	64	134	(C <sub>16</sub> H <sub>11</sub> O <sub>3</sub> N <sub>5</sub> S), C, H, N
5'-Benzylthio- ( <b>2e</b> )	PhCH <sub>2</sub> SH-2 N NaOH	65	150-152	(C <sub>17</sub> H <sub>15</sub> O <sub>3</sub> N <sub>5</sub> S), C, H, N

†Melting points were determined on a Buchi-Tottoli melting point apparatus and uncorrected.

‡Where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values.

Table VI. 6- and 2,6-Substituted Purine Ribosides

Starting material	Amount, g	Reagent	Reaction temp, °C	Reaction time, hr	Product	Yield, %	Mp, °C	Anal.
6-Chloroguanosine (4d)	0.5	NH <sub>2</sub> OH-EtOH	reflux	6	2-Amino-6-hydroxy-aminopurine riboside (5b)	100	228	(C <sub>10</sub> H <sub>14</sub> O <sub>5</sub> N <sub>7</sub> ·H <sub>2</sub> O), C, H, N
2',3',5'-Tri-O-benzoyl-6-chloroguanosine (4c)	12	NH <sub>3</sub> -MeOH	100	5	2-Aminoadenosine (5c)	55	247	(C <sub>10</sub> H <sub>14</sub> O <sub>5</sub> N <sub>6</sub> ·H <sub>2</sub> O), C, H, N
6-Chloroinosine (4a)	1.0	40% NH <sub>2</sub> NH <sub>2</sub>	100	4	6-Hydrazinopurine riboside (5d)	98	218-220	(C <sub>10</sub> H <sub>14</sub> O <sub>4</sub> N <sub>6</sub> ), C, H, N
6-Chloroguanosine (4d)	2.0	40% NH <sub>2</sub> NH <sub>2</sub>	100	1	2-Amino-6-hydrazinopurine riboside (5e)	100	214-216	(C <sub>10</sub> H <sub>14</sub> O <sub>4</sub> N <sub>7</sub> ·0.5H <sub>2</sub> O), C, H, N
6-Chloroinosine (4a)	0.5	morpholine-EtOH	reflux	17	6-Morpholinopurine riboside (5f)	84	160-163	(C <sub>14</sub> H <sub>19</sub> O <sub>5</sub> N <sub>5</sub> ·H <sub>2</sub> O), C, H, N
6-Chloroguanosine (4d)	0.6	MeNH-EtOH	100	5	2-Amino-6-methylaminopurine riboside (5g)	27	165	(C <sub>16</sub> H <sub>16</sub> O <sub>4</sub> N <sub>6</sub> ), C, H, N

dissolved in 150 ml of NH<sub>3</sub>-MeOH and the soln was kept at room temp overnight. It was then evapd *in vacuo*, and coevapd repeatedly with EtOH. A cryst powder of 4d (3.55 g, 60%) was obtd, mp 175° dec (lit.<sup>20</sup> mp 171-172° dec).

**2- and/or 6-Substituted Purine Ribosides (5b-g).** The compds (5b-g) were prepd by reaction of 4a, c, d with appropriate amines.

**2-Aminoadenosine 5'-Monophosphate (7).** 2-Aminoadenosine (5c) (140 mg, 0.5 mmole) was dissolved with stirring in an ice-cold soln of POCl<sub>3</sub> (1.53 g, 10 mmoles), H<sub>2</sub>O (0.018 ml, 1 mmole), and 1.25 ml of PO(OEt)<sub>3</sub>. The mixt was stirred at 0-5°. After 24 hr it was poured over 100 ml of ice water with stirring. It was then adsorbed to a column of 5 g of activated C and was washed well with H<sub>2</sub>O. The column was eluted with 500 ml of 10% NH<sub>4</sub>OH-EtOH (1:1) and the eluate was evapd to dryness. Paper chromatography (*i*-PrOH-NH<sub>4</sub>OH-H<sub>2</sub>O, 7:1:2) of the residue showed 2 spots having *R*<sub>f</sub>'s of 0.31 and 0.05, the former corresponding to that of 5c. The residue was dissolved in 10 ml of H<sub>2</sub>O and was adjusted to pH 9 with NH<sub>4</sub>OH. It was applied to a Dowex 1-X4 (HCOO<sup>-</sup>) column (1.7 × 40 cm). The column washed with 100 ml of H<sub>2</sub>O was eluted with 0.1 *M* HCOOH. Fractions at 100-200 ml were evapd to dryness and cryst powder of 7 (50 mg, 23%) was obtd. Paper chromatog showed a single spot having *R*<sub>f</sub> of 0.05 and paper electrophoresis (0.01 *M* phosphate buffer, pH 7.5) showed the same mobility as 5'-AMP. Uv spectrum was same as that of the starting material, 5c. *Anal.* (C<sub>10</sub>H<sub>13</sub>O<sub>8</sub>N<sub>6</sub>P·H<sub>2</sub>O), C, H, N.

**8-Substituted Adenosine Derivatives (8-11).** These compds were the gifts of Professor M. Ikehara.

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