Studies on the Synthesis of Compounds Related to Adenosine 3′,5′-Cyclic Phosphate. VII.[1] Synthesis and Cardiac Effects of N^6,N^6-Dialkyl Adenosine 3′,5′-Cyclic Phosphates

Shigehiro KATAOKA,*,[a] Nobuyuki YAMAJI,[a] Motohiko KATO,[a] Tomie KAWADA,[b] and Shoichi IMAI[b]

Research and Development Division, Kikkoman Corporation,[a] 399 Noda, Chiba 278, Japan and Department of Pharmacology, School of Medicine, Niigata University,[b] 757 Asahimachidori-1-bancho, Niigata 951, Japan. *Received June 12, 1990*

A series of novel N^6,N^6-dialkyl adenosine 3′,5′-cyclic phosphates (N^6,N^6-dialkyl cAMPs) was synthesized from 2′-*O*-*p*-toluenesulfonyl cAMP (2′-*O*-tosyl cAMP, 2) and tested for inotropic and chronotropic activities *in vitro*. Treatment of 2 with excess alkyl halides and sodium hydride followed by detosylation with aqueous NaOH readily gave N^6,N^6-dialkyl cAMPs (3) in good yields. Various N^6,N^6-dialkyl cAMPs having different alkyl groups at the N^6-position (9—12) were prepared by alkylation followed by detosylation of N^6-alkyl-2′-*O*-tosyl cAMPs (4) which were obtained by the reductive alkylation of 2 with aldehydes in the presence of sodium cyanoborohydride in acetic acid or tosylation of N^6-methyl cAMP. The mechanism of the detosylation is briefly discussed. Among the N^6,N^6-dialkylated derivatives, N^6,N^6-dipentyl (3f) and N^6-ethyl-N^6-heptyl (10e) derivatives were found to exhibit a potent positive inotropic effect and a weak positive chronotropic effect. The structure–activity relationships for the position and the length of alkyl residue are discussed.

Keywords alkylation; N^6,N^6-dialkyl cAMP; 2′-*O*-tosyl cAMP; detosylation, inotropic effect; chronotropic effect; phosphodiesterase inhibitor

Numerous derivatives of adenosine 3′,5′-cyclic phosphate (cAMP, **1**) have been prepared to obtain more potent biological activity than that of the original one.[2] In previous studies[1,3] we prepared various N^6- and/or 2′-*O*-alkyl cAMP derivatives which were expected to exhibit enhanced biological activities due to their high lipophilicity and found that N^6-monoalkyl cAMPs showed more potent positive inotropic effect (PIE) than 2′-*O*-alkyl, N^6,2′-*O*-dialkyl, and N^6,N^6,2′-*O*-trialkyl cAMPs. We now describe the synthesis of N^6,N^6-dialkyl cAMPs with free 2′-hydroxy group (N^6,N^6-diR cAMPs (**3**) and N^6-R_1-N^6-R_2 cAMPs having different alkyl groups (**9**—**12**)) and their cardiac activities. N^6,N^6-Dimethyl and N^6,N^6-diethyl cAMPs have been prepared from inosine 3′,5′-cyclic phosphate *via* 6-chloro-9-β-D-ribofuranosylpurine-3,5-cyclic phosphate,[4,5] but higher homologues have not been reported so far. Synthesis of N^6,N^6-diR cAMPs (**3**) from cAMP (**1**) by this method requires tedious and multistep reactions such as deamination of the 6-amino group and protection of the 2′-hydroxy group.[4] We devised a simple method for preparing N^6,N^6-diR cAMPs (**3**) from **1**. Moreover, the synthesis of N^6-R_1-N^6-R_2 derivatives (**9**—**12**) which are of interest in the structure–activity relationships between the length of alkyl residues (R_1 and R_2) and the PIE has not been reported.

Synthesis of 2′-*O*-Protected cAMP As reported in our previous paper,[1] alkylation of **1** using excess alkyl halides and NaH gave N^6,N^6,2′-*O*-trialkyl cAMPs. Therefore, it seemed that N^6,N^6-dialkyl cAMP could be obtained from **1** when the 2′-hydroxy group of **1** is properly protected. We first protected 2′-hydroxy group of **1** using *tert*-butyldimethylsilyl chloride (TBDMSCl) which was used to protect a hydroxy group of nucleosides.[6] The tri-*n*-butylammonium (TBA) salt of **1** was treated with TBDMSCl in the presence of imidazole in dimethylformamide (DMF) to give 2′-*O*-TBDMS cAMP in 97% yield. However, dibutylation of 2′-*O*-TBDMS cAMP with excess butyl bromide and NaH in dimethyl sulfoxide (DMSO) at room temperature afforded N^6,N^6,2′-*O*-tributyl cAMP[1] and N^6,N^6-dibutyl cAMP (**3d**) in 29 and 25% yields, respectively. This revealed that deprotection of the silyl group occurred during the reaction.

Protection of 2′-hydroxy group with *p*-toluenesulfonyl (tosyl) group was examined next, and 2′-*O*-tosyl cAMP (**2**) was prepared from **1** by treatment with tosyl chloride in aqueous NaOH–dioxane at room temperature.[7] Compound (**2**) was stable under the dialkylation conditions. Though detosylation of tosylates by the reductive cleavage with sodium naphthalene or lithium aluminum hydride has been known,[8] we examined a more convenient method of detosylation. When the tosylate (**2**) was treated with aqueous NaOH–methanol at 50 °C, cAMP (**1**) was obtained in excellent yield [98% on high-performance liquid chromatography (HPLC)], which was identified by comparison of its infrared (IR) and proton nuclear magnetic resonance (^{1}H-NMR) spectra and retention time on HPLC with those of an authentic sample (Chart 1). Thus, this detosylation proceeded to give the alcohol with retention of the configuration. The mechanism of this reaction seems to be a double inversion with participation of the purine ring or the S–O bond fission initiating a nucleophilic displacement on sulfonate sulfur in the compound. To ascertain the detosylation mechanism, we examined the detosylation with MeONa in O-labeled water ($H_2{}^{18}O$)–MeOH. The secondary ion mass spectrum (SIMS) of the *p*-toluenesul-

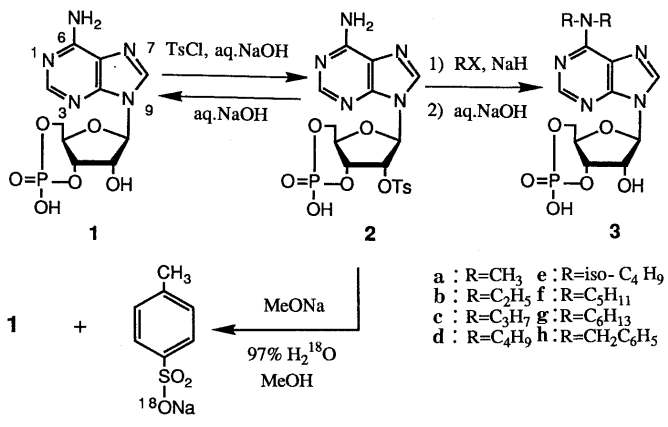


Chart 1

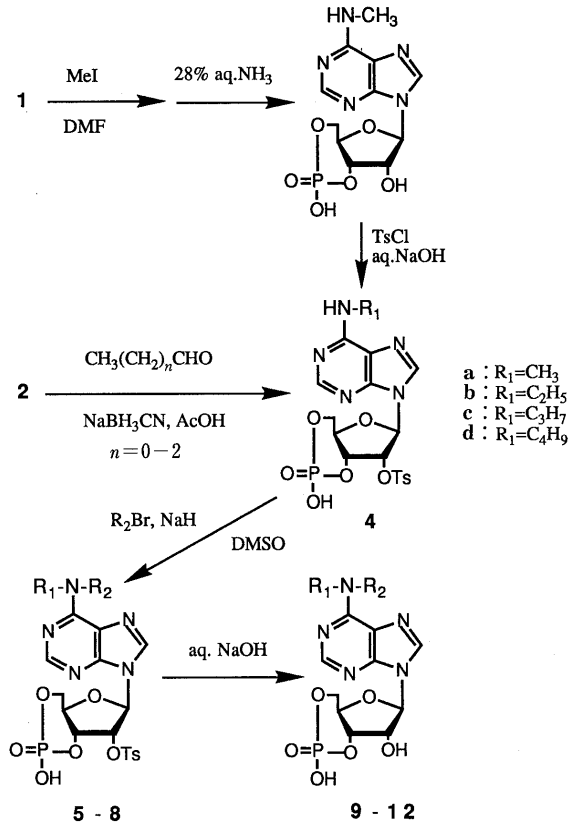
fonic acid (TsOH) isolated in 87% yield showed that the intensity of m/z 175 $(M+H+2)^+$ peak was 1.6-fold higher than that of the reference, whereas no ^{18}O was incorporated in the cAMP isolated in 81% yield. The low ^{18}O-incorporation into TsOH is probably due to ^{18}O–^{16}O exchange reaction during the isolation procedure. Therefore, we next isolated the TsONa which was formed in the reaction. The SIMS of the TsONa isolated in 75% yield showed that the intensity of m/z 219 $(M+Na+2)^+$ peak was increased up to 2.6-fold higher than that of the reference (see Table I). Although the ratio of ^{18}O-incorporation into TsOH and TsONa was not satisfactory, these results indicated that the original oxygen atom in the 2′-hydroxy group on **1** remained after detosylation of 2′-*O*-tosyl cAMP (**2**). Therefore, the hydrolysis of the tosylate was initiated by nucleophilic attack of hydroxide ion on sulfonate sulfur and resulted in the S–O bond fission to form **1** (Chart 1). Bunton and Frei reported that such S–O bond fission occurred in aryl tosylates whose geometry makes attack on carbon unfavorable.[9] Williams and Buchanan *et al.* reported the treatment of the 2,3-ditosylate of benzyl 4,6-*O*-benzylidene-α-D-glucopyranoside with methanolic sodium methoxide to afford benzyl 2,3-anhydro-4,6-*O*-benzylidene-α-D-allopyranoside,[10] and explained that the first sulfonyl group was removed by S–O bond fission, facilitated by the inductive effect on the adjacent sulfonyloxy group.[10a] Further, Ishido *et al.* reported that the 2′-hydroxy group of nucleosides was the most acidic due to the electronic effect of the heterocyclic aglycone moieties.[11] Therefore, it was considered that S–O bond fission of the tosylate (**2**) occurred due to the steric hindrance of the adenine moiety which makes attack on 2′-carbon unfavorable, and the facilitation of the reaction was due to the inductive effect of adenine moiety.

TABLE I. The Ratio of ^{18}O-Incorporation into the Detosylation Products of **2**

Compd.	m/z	
	$(M+H+2)^+$ (%)[a]	$(M+Na+2)^+$ (%)[a]
TsOH[b]	14.1	
TsOH[c]	8.7	
cAMP[b]	20.4	
cAMP[c]	22.1	
TsONa[b]		20.3
TsONa[c]		7.7

a) These values are expressed as percent from the $(M+H)^+$ and $(M+Na)^+$ ion peak intensities in mass spectra. *b*) Isolated compound. *c*) Commercial sample.

Synthesis of Dialkylated cAMPs Treatment of **2** with excess alkyl halides and NaH in DMSO at room temperature, followed by detosylation of the corresponding N^6,



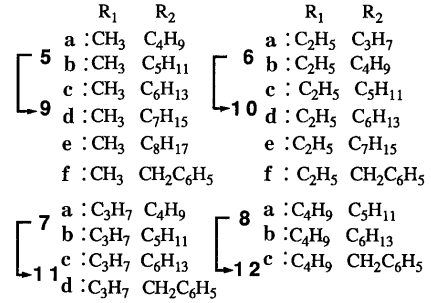


Chart 2

TABLE II. Yields and Physical Constants of N^6,N^6-DiR cAMP Derivatives (**3**)

Compd. No.	R	Yield (%)	Rf[a]	UV (nm)		Formula[d]	Analysis (%) Calcd			Found		
				pH 13[b] (ε)	pH 1[c] (ε)		C	H	N	C	H	N
3a	CH_3	49	0.18	274 (17700)	267 (17000)	$C_{12}H_{16}N_5O_6P\cdot 1/2H_2O$	39.35	4.68	19.12	39.34	4.70	19.07
3b	C_2H_5	53	0.26	276 (19600)	269 (20500)	$C_{14}H_{20}N_5O_6P\cdot 1/3H_2O$	42.97	5.32	17.90	43.03	5.34	17.91
3c	C_3H_7	67	0.33	278 (19100)	270 (20200)	$C_{16}H_{24}N_5O_6P\cdot H_2O$	44.55	6.07	16.24	44.62	5.80	16.07
3d	C_4H_9	69	0.38	278 (19300)	271 (19700)	$C_{18}H_{28}N_5O_6P\cdot 3/4H_2O$	47.52	6.53	15.39	47.59	6.40	15.29
3e	iso-C_4H_9	41	0.38	281 (19200)	273 (18700)	$C_{18}H_{28}N_5O_6P\cdot 2/3H_2O$	47.68	6.52	15.44	47.71	6.44	15.36
3f	C_5H_{11}	49	0.42	279 (19800)	271 (18100)	$C_{20}H_{32}N_5O_6P\cdot H_2O$	49.73	7.05	14.50	49.63	6.82	14.35
3g	C_6H_{13}	70	0.46	278 (19700)	271 (18400)	$C_{22}H_{36}N_5O_6P\cdot 3/4H_2O$	51.37	7.34	13.71	51.51	7.25	13.71
3h	$CH_2C_6H_5$	37	0.43	277 (22100)	276 (20000)	$C_{24}H_{24}N_5O_6P\cdot 3/2H_2O$	53.73	5.07	13.05	53.53	4.82	12.71

a) *Rf* on Kiesel gel 60 F_{254} (Merck) plate; solvent system (0.1 N NH_4Cl : MeOH : CH_3CN, 1 : 4 : 16, v/v). *b*) Solvent; 0.1 N NaOH. *c*) Solvent; 0.1 N HCl. *d*) Samples were dried over P_2O_5 at 50 °C at 3 mmHg for 4—5 h.

TABLE III. Yields and Physical Constants of N^6-R_1-N^6-R_2-2′-*O*-tosyl cAMP Derivatives (**5**—**8**)

Compd. No.	R_1	R_2	Yield (%)	Rf[a]	UV (nm)[b] (ε)	Formula[c]	Analysis (%) Calcd C	H	N	Found C	H	N
5a	CH_3	C_4H_9	70	0.27	272 (18200)	$C_{22}H_{28}N_5O_8PS$	47.74	5.10	12.65	47.60	5.19	12.44
5b	CH_3	C_5H_{11}	70	0.28	272 (18700)	$C_{23}H_{30}N_5O_8PS \cdot 5/4H_2O$	46.83	5.51	11.87	46.61	5.30	11.88
5c	CH_3	C_6H_{13}	73	0.29	273 (18200)	$C_{24}H_{32}N_5O_8PS \cdot 3/4H_2O$	48.44	5.67	11.76	48.38	5.54	11.60
5d	CH_3	C_7H_{15}	61	0.31	273 (18000)	$C_{25}H_{34}N_5O_8PS \cdot 3/2H_2O$	48.25	5.94	11.25	48.05	5.66	11.12
5e	CH_3	C_8H_{17}	69	0.32	273 (17200)	$C_{26}H_{36}N_5O_8PS \cdot 2H_2O$	48.37	6.24	10.85	48.46	5.94	10.73
5f	CH_3	$CH_2C_6H_5$	64	0.26	273 (20300)	$C_{25}H_{26}H_5O_8PS \cdot 1/4H_2O$	50.72	4.51	11.83	50.53	4.51	11.73
6a	C_2H_5	C_3H_7	70	0.26	274 (19700)	$C_{22}H_{28}N_5O_8PS \cdot 3/4H_2O$	46.60	5.21	12.36	46.52	5.11	12.31
6b	C_2H_5	C_4H_9	76	0.28	274 (19700)	$C_{23}H_{30}N_5O_8PS \cdot 1/4H_2O$	48.29	5.34	12.25	48.34	5.46	12.13
6c	C_2H_5	C_5H_{11}	82	0.29	274 (19800)	$C_{24}H_{32}N_5O_8PS$	49.57	5.55	12.04	49.75	5.67	11.93
6d	C_2H_5	C_6H_{13}	76	0.31	275 (19950)	$C_{25}H_{34}N_5O_8PS \cdot H_2O$	48.94	5.91	11.41	48.82	5.74	11.25
6e	C_2H_5	C_7H_{15}	76	0.32	275 (20500)	$C_{26}H_{36}N_5O_8PS \cdot 1/2H_2O$	50.48	5.99	11.33	50.57	6.04	11.29
6f	C_2H_5	$CH_2C_6H_5$	52	0.30	273 (19800)	$C_{26}H_{28}N_5O_8PS \cdot 4/5H_2O$	50.70	4.84	11.37	50.59	4.56	11.35
7a	C_3H_7	C_4H_9	67	0.31	273 (18500)	$C_{24}H_{32}N_5O_8PS \cdot 1/2H_2O$	48.81	4.84	11.37	48.89	5.63	11.84
7b	C_3H_7	C_5H_{11}	73	0.33	273 (19500)	$C_{25}H_{34}N_5O_8PS$	50.44	5.71	11.76	50.24	5.82	11.72
7c	C_3H_7	C_6H_{13}	70	0.34	273 (19550)	$C_{26}H_{36}N_5O_8PS \cdot 3/4H_2O$	50.14	6.02	11.24	50.01	5.90	11.16
7d	C_3H_7	$CH_2C_6H_5$	66	0.32	273 (20900)	$C_{27}H_{30}N_5O_8PS \cdot 1/2H_2O$	51.94	4.96	11.21	51.92	4.93	10.93
8a	C_4H_9	C_5H_{11}	73	0.33	275 (19950)	$C_{26}H_{36}N_5O_8PS \cdot 7/4H_2O$	48.73	6.16	10.93	48.63	5.93	10.73
8b	C_4H_9	C_6H_{13}	72	0.34	275 (19850)	$C_{27}H_{38}N_5O_8PS \cdot H_2O$	50.56	6.23	10.92	50.55	6.19	10.94
8c	C_4H_9	$CH_2C_6H_5$	68	0.32	274 (20100)	$C_{28}H_{32}N_5O_8PS \cdot H_2O$	51.87	5.24	10.80	51.81	5.06	10.63

a) *Rf* on Kiesel gel 60 F_{254} (Merck) plate; solvent system (MeOH : $CHCl_3$ = 3 : 7, v/v). b) Solvent; EtOH. c) See footnote *d*) in Table II.

N^6-diR-2′-*O*-tosyl cAMPs with 2 N NaOH (6—14 mol eq) in methanol–water solution at room temperature in a one-pot reaction afforded the desired products: **3a** (49% yield), **3b** (53%), **3c** (67%), **3d** (69%), **3e** (41%), **3f** (49%), **3g** (70%), and **3h** (37%) (Chart 1 and Table II). The ultraviolet (UV) absorption maxima of **3** occurred at 274—281 nm (pH 13) and 267—276 nm (pH 1) similar to those of N^6,N^6-diethyl cAMP previously reported.[5] The ^{1}H-NMR spectra of **3** revealed the signals due to $-CH_2NCH_2-$ at about δ 3.9 ppm (4H, br s), and a signal due to 6-NH_2 at δ 7.5 ppm (2H, br s) in **1** disappeared.

Next, we synthesized N^6-R_1-N^6-R_2 cAMPs having different alkyl groups (**9**—**12**) from **2**. The first alkylation of the 6-amino group in **2** was achieved by the reductive alkylation as previously reported.[3] Thus, **2** was treated with aldehydes in the presence of sodium cyanoborohydride in acetic acid at 50 °C to give the corresponding N^6-monoalkyl (R_1)-2′-*O*-tosyl cAMPs; **4b** (78% yield), **4c** (47%), **4d** (54%). N^6-Methyl derivative (**4a**) was prepared by the tosylation of N^6-methyl cAMP which was synthesized from 1-methyl cAMP in the usual manner using Dimroth rearrangement[12] (Chart 2).

The second alkylation at the N^6-position of the N^6-monoalkylated derivatives (**4**) was carried out by treatment of **4** with excess alkyl bromides and NaH in DMSO to affford N^6-R_1-N^6-R_2-2′-*O*-tosyl cAMPs: N^6-CH_3-N^6-R_2 derivatives (**5a**—**f**, 61—73% yields), N^6-C_2H_5-N^6-R_2 derivatives (**6a**—**f**, 52—82%), N^6-C_3H_7-N^6-R_2 derivatives (**7a**—**d**, 66—73%), and N^6-C_4H_9-N^6-R_2 derivatives (**8a**—**c**, 68—73%) (Table III). Removal of the tosyl protecting group of **5**—**8** was carried out with 2 N NaOH (5—11 mol eq) in methanol–water solution at 50—60 °C to give the desired products; **9a**—**f** (51—74% yields), **10a**—**f** (56—88%), **11a**—**d** (64—88%), and **12a**—**c** (56—72%) (Chart 2 and Table IV). The UV and ^{1}H-NMR spectra of these compounds were similar to those of **3**.

Biological Results and Discussion The inotropic and chronotropic effects of N^6,N^6-diR cAMPs (**3**) and N^6-R_1-N^6-R_2 cAMPs (**9**—**12**) synthesized were tested using papillary muscle of the right ventricle and right atria, respectively, of guinea pig hearts. The results are shown in Tables V and VI. We reported previously that N^6-monoalkyl cAMPs (butyl-decyl, benzyl, and furfuryl) exhibited PIEs and the introduction of the substituent at 2′-hydroxy group of them led to the disappearance of their PIEs.[1,3] The resulting data of **3** indicated that their inotropic profile is similar to that of N^6-monoalkyl cAMPs.[3] Thus, the PIE was increased by increasing the length of alkyl chain (increasing lipophilicity), and the butyl derivative (**3d**) exhibited the most potent activity. The order of inotropic activity of **3** was **3d** > **3c** = **3f** > **3h** > **3e** and the active compounds (**3c**, **d**, and **f**) had 6—10 carbon atoms in the dialkyl residues at the N^6-position. The dibutyl derivative (**3d**) having 8 alkyl carbon atoms exhibited the most potent PIE among the cAMP derivatives prepared thus far. We reported the inotropic activity of N^6,N^6,2′-*O*-tributyl and N^6,N^6,2′-*O*-tripropyl cAMPs were very weak,[1] but in this study N^6,N^6-dibutyl and N^6,N^6-dipropyl cAMPs (**3d** and **3c**) exhibited potent PIE. Therefore, it was also clarified that appearance of the PIE of cAMP derivatives requires the presence of 2′-hydroxy group.[1] It was also suggested that the inotropic activity of **3** was influenced by a steric factor of 6-alkylamino chain, because the inotropic activity of the branched alkyl derivative (**3e**; isobutyl) and the aromatic derivative (**3h**; benzyl) was only about 1/9 that of **3d**. The short-chain derivatives (**3a**, **b**) did not show PIE as did N^6-mono short-chain derivatives; the reason for this seemed to be their low lipophilicities.[13] As regards the positive chronotropic effect (PCE), it was possible to determine ED_{30} values with compounds (**3a**—**d**). Among the potent PIE compounds (**3c**, **d**, **f**), dipentyl derivative (**3f**) showed the weakest chronotropic action and it was not feasible to obtain ED_{30} value in concentration up to 1 mM.

TABLE IV. Yields and Physical Constants of N^6-R_1-N^6-R_2 cAMP Derivatives (**9**—**12**)

Compd. No.	R_1	R_2	Yield (%)	*Rf*[a]	UV (nm) pH 13[b] (ε)	UV (nm) pH 1[c] (ε)	Formula[d]	Analysis (%) Calcd C	Calcd H	Calcd N	Found C	Found H	Found N
9a	CH_3	C_4H_9	71	0.40	276 (18850)	269 (19700)	$C_{15}H_{22}N_5O_6P\cdot 1/2H_2O$	44.12	5.68	17.15	44.05	5.63	17.01
9b	CH_3	C_5H_{11}	74	0.42	276 (18900)	269 (19300)	$C_{16}H_{24}N_5O_6P\cdot 1/2H_2O$	45.51	5.96	16.58	45.63	5.95	16.40
9c	CH_3	C_6H_{13}	62	0.44	276 (18800)	269 (19100)	$C_{17}H_{26}N_5O_6P\cdot 3/4H_2O$	46.31	6.29	15.88	46.33	6.21	15.81
9d	CH_3	C_7H_{15}	51	0.46	276 (18900)	269 (19900)	$C_{18}H_{28}N_5O_6P\cdot 1/2H_2O$	48.00	6.48	15.55	48.19	6.45	15.45
9e	CH_3	C_8H_{17}	60	0.47	276 (18600)	269 (19000)	$C_{19}H_{30}N_5O_6P\cdot 5/4H_2O$	47.75	6.85	14.65	47.90	6.78	14.46
9f	CH_3	$CH_2C_6H_5$	57	0.42	277 (21500)	271 (20200)	$C_{18}H_{20}N_5O_6P\cdot 3/4H_2O$	48.40	4.81	15.67	48.56	4.83	15.42
10a	C_2H_5	C_3H_7	67	0.42	277 (19650)	270 (20000)	$C_{15}H_{22}N_5O_6P\cdot 1/2H_2O$	44.12	5.68	17.15	44.21	5.59	17.15
10b	C_2H_5	C_4H_9	88	0.46	277 (19100)	270 (19200)	$C_{16}H_{24}N_5O_6P\cdot 1/2H_2O$	45.51	5.96	16.58	45.29	5.90	16.51
10c	C_2H_5	C_5H_{11}	82	0.49	277 (18500)	270 (19000)	$C_{17}H_{26}N_5O_6P\cdot 2/3H_2O$	46.47	6.27	15.94	46.35	6.24	15.72
10d	C_2H_5	C_6H_{13}	56	0.50	279 (19950)	270 (19200)	$C_{18}H_{28}N_5O_6P\cdot 5/4H_2O$	46.60	6.58	15.11	46.70	6.33	15.06
10e	C_2H_5	C_7H_{15}	79	0.51	278 (19500)	270 (19700)	$C_{19}H_{30}N_5O_6P\cdot 1/2H_2O$	49.19	6.68	15.09	49.13	6.67	15.19
10f	C_2H_5	$CH_2C_6H_5$	76	0.48	277 (21000)	271 (19500)	$C_{19}H_{22}N_5O_6P\cdot 2/3H_2O$	49.67	5.15	15.24	49.46	5.13	15.09
11a	C_3H_7	C_4H_9	75	0.52	278 (18900)	271 (19900)	$C_{17}H_{26}N_5O_6P\cdot 1/2H_2O$	46.81	6.19	16.06	46.99	6.20	15.95
11b	C_3H_7	C_5H_{11}	88	0.53	277 (19400)	271 (20500)	$C_{18}H_{28}N_5O_6P\cdot 1/2H_2O$	48.02	6.44	15.56	48.81	6.69	15.49
11c	C_3H_7	C_6H_{13}	67	0.54	278 (19100)	271 (19600)	$C_{19}H_{30}N_5O_6P\cdot 3/4H_2O$	48.69	6.72	14.49	48.81	6.69	14.74
11d	C_3H_7	$CH_2C_6H_5$	64	0.52	278 (21300)	272 (20100)	$C_{20}H_{24}N_5O_6P\cdot 3/4H_2O$	50.60	5.37	14.75	50.65	5.26	14.74
12a	C_4H_9	C_5H_{11}	67	0.56	280 (20700)	271 (20000)	$C_{19}H_{30}N_5O_6P\cdot 1/2H_2O$	49.16	6.68	15.09	49.20	6.72	14.88
12b	C_4H_9	C_6H_{13}	56	0.57	280 (20400)	271 (19400)	$C_{20}H_{32}N_5O_6P\cdot 3/4H_2O$	49.76	6.94	14.51	49.85	6.93	14.38
12c	C_4H_9	$CH_2C_6H_5$	72	0.55	278 (20700)	272 (20300)	$C_{21}H_{26}N_5O_6P\cdot 1/2H_2O$	52.09	5.58	14.46	52.05	5.49	14.20

a) *Rf* on Kiesel gel 60 F_{254} (Merck) plate; solvent system (*n*-BuOH : AcOH : H_2O, 8 : 2 : 1, v/v). *b*) Solvent; 0.1 N NaOH. *c*) Solvent; 0.1 N HCl. *d*) See footnote *d*) in Table II.

TABLE V. Inotropic and Chronotropic Effects of N^6,N^6-DiR cAMP Derivatives (**3**)

Compd. No.	R	PIE ED_{30} ($\times 10^{-4}$ M)[a]	PCE ED_{30} ($\times 10^{-4}$ M)[a]
3a	CH_3	$>10^{-3}$	6.20±1.00
3b	C_2H_5	$>10^{-3}$	4.19±0.62
3c	C_3H_7	1.77±0.27	0.80±0.16
3d	C_4H_9	0.75±0.20	1.85±0.49
3e	iso-C_4H_9	7.06±1.56	—
3f	C_5H_{11}	1.89±0.23	—
3g	C_6H_{13}	$>10^{-3}$	—
3h	$CH_2C_6H_5$	6.36±2.10	—

PIE = positive inotropic effect; PCE = positive chronotroic effect. *a*) ED_{30} = concentration required to produce 30% of the maximum response evoked by 10^{-7} M isoproterenol (ISP). Mean±S.E. (n=4—5).

TABLE VI. Inotropic and Chronotropic Effects of N^6-R_1-N^6-R_2 cAMP Derivatives (**9**—**12**)

Compd. No.	R_1	R_2	PIE ED_{30} ($\times 10^{-4}$ M)[a]	PCE ED_{30} ($\times 10^{-4}$ M)[a]
9a	CH_3	C_4H_9	5.90±0.67	3.50±0.97
9b	CH_3	C_5H_{11}	5.07±0.96	4.05±0.61
9c	CH_3	C_6H_{13}	3.72±0.85	2.50±0.64
9d	CH_3	C_7H_{15}	5.81±1.05	—
9e	CH_3	C_8H_{17}	3.93±0.99	—
9f	CH_3	$CH_2C_6H_5$	$>10^{-3}$	—
10a	C_2H_5	C_3H_7	1.54±0.76	4.40±0.90
10b	C_2H_5	C_4H_9	3.48±0.98	2.75±0.42
10c	C_2H_5	C_5H_{11}	1.91±0.41	1.38±0.21
10d	C_2H_5	C_6H_{13}	4.86±1.41	3.06±0.97
10e	C_2H_5	C_7H_{15}	1.58±0.20	—
10f	C_2H_5	$CH_2C_6H_5$	$>10^{-3}$	9.13±1.34
11a	C_3H_7	C_4H_9	2.14±0.42	1.39±0.22
11b	C_3H_7	C_5H_{11}	0.91±0.30	2.84±0.80
11c	C_3H_7	C_6H_{13}	2.47±0.57	1.15±0.25
11d	C_3H_7	$CH_2C_6H_5$	3.93±0.37	6.02±0.86
12a	C_4H_9	C_5H_{11}	0.92±0.17	2.16±0.74
12b	C_4H_9	C_6H_{13}	2.98±0.99	3.66±0.77
12c	C_4H_9	$CH_2C_6H_5$	3.39±0.65	6.40±0.81

PIE and PCE, see footnotes in Table V. *a*) See footnote *a*) in Table V.

On the basis of the results of **3** for cardiac effect, we decided to synthesize the compounds (**9**—**12**) whose total carbon number (R_1+R_2) in N^6-dialkyl chain was in the range of 5—10. Compounds (**9**—**12**) showed PIE except for **9f** and **10f**. Compounds (**10a**: $R_1, R_2 = C_2H_5, C_3H_7$), (**10c**: C_2H_5, C_5H_{11}), (**10e**: C_2H_5, C_7H_{15}), (**11b**: C_3H_7, C_5H_{11}), (**12a**: C_4H_9, C_5H_{11}) exhibited potent PIEs almost the same or twice as that of N^6-monoheptyl cAMP [ED_{30} = 1.60± 0.19 ($\times 10^{-4}$ M)], which was the most potent cardiotonic agent among N^6-monoalkyl cAMP derivatives.[3] In the chronotropic effect, all compounds exhibited a positive action and with most of them it was possible to obtain ED_{30} value of PCE. Among the potent PIE compounds (**10a, c, e**, **11b**, and **12a**), **10e** showed too weak a positive effect to obtain ED_{30} value of PCE in concentration up to 1 mM.

These results indicate that N^6,N^6-dialkyl derivatives of cAMP could exhibit a potent PIE and PCE. The compound (**3b**) was known to activate a protein kinase and to be a potent inhibitor of heart phosphodiesterase.[14] Thus, we presume that the positive effects of compounds which were synthesized in the present work might be due to activities similar to those of **3b**. In the series of novel N^6,N^6-dialkyl cAMPs prepared, **3f** and **10e** were found to have potent PIEs with relatively minor increases in heart rate, so the pharmacological evaluations of these compounds as cardiotonic agents are of interest. It is concluded that substitution of the 6-amino group in cAMP is a potentially useful approach for obtaining higher activity.

Experimental

^{1}H-NMR spectra were taken at 200 MHz on a JEOL JNM-FX200 NMR spectrometer in DMSO-d_6. All ^{1}H-NMR data are reported in ppm

downfield from tetramethylsilane as an internal standard. UV absorption spectra were recorded with a Hitachi 557 spectrophotometer. SIMS were recorded on a Hitachi M-80B spectrometer. IR spectra were taken on a JASCO A-202 spectrophotometer. HPLC was performed on a Finepak SIL C_{18} column (4.6 × 250 mm) with MeOH–10 mM acetate buffer (pH 4) containing 1 mM tetra-*n*-butylammonium chloride (5:5—8:2, v/v) as eluents. The eluate was monitored at 265 nm (compound **4**) or 275 nm (compounds **3** and **5**—**12**). Analytical and preparative thin-layer chromatographies (TLC) were performed on Kiesel gel 60 F_{254} (Merck) plates. Chromatographic separation was done on Merck Silica gel 60 or Dowex 50w-x8 (H^+) or charcoal (Shirasagi KHL 250, Takeda Chemical Industries, Ltd.) with the indicated eluents. Analytical samples were dried over P_2O_5 at 50 °C at 3 mmHg for 4—5 h.

Preparation of 2′-*O*-Tosyl cAMP (2) A solution of tosyl chloride (86 g, 450 mmol) in dioxane (600 ml) was added to a solution of **1** (32.9 g, 100 mmol) in 200 ml of aqueous NaOH (11 g, 260 mmol), and the solution was stirred at room temperature overnight. The resulting precipitate was filtered off, washed with dioxane, and dried *in vacuo* to give **2**. The mother liquor was concentrated *in vacuo* and the precipitate was filtered off, washed with dioxane, and dried *in vacuo* to afford **2**. Total yield was 38.6 g (80%). IR ν_{max}^{KBr} cm^{-1}: 3565, 3300, 3040, 2870, 2825, 1680, 1600, 1185. ^{1}H-NMR δ: 2.33 (3H, s, CH_3), 5.10—5.30 (1H, m, H-3′), 5.58 (1H, d, J=6.4 Hz, H-2′), 6.20 (1H, s, H-1′), 7.30 and 7.71 (2H each, d, J=8.4 Hz, phenyl H's), 7.86 (2H, br s, NH_2), 8.23 (2H, s, purine H's overlap). UV λ_{max}^{EtOH} nm (ε): 258 (14400). *Anal.* Calcd for $C_{17}H_{18}N_5O_6PS \cdot 2/3H_2O$: C, 41.23; H, 3.90; N, 14.14. Found: C, 41.03; H, 3.85, 14.16.

Detosylation of 2′-*O*-Tosyl cAMP (2) (i) Isolation of cAMP and TsOH: Compound (**2**, 90 mg, 0.19 mmol) was suspended in absolute MeOH (3 ml)–97% ^{18}O-labeled H_2O ($H_2{}^{18}O$, 2 ml) solution and the suspension was dissolved by 28% MeONa in MeOH (0.3 ml, 7.7 mol eq) under dry nitrogen atmosphere. The mixture was refluxed for 3.5 h with stirring and evaporated *in vacuo*. The resulting residue was subjected to chromatography on Dowex 50w-x8 (H^+) with H_2O. The earlier fractions gave TsOH (28 mg, 87%) as a colorless amorphous solid. The later fractions afforded cAMP (49.5 mg, 81%) as a colorless amorphous solid. Each structure was confirmed by respective comparison of its IR, MS and ^{1}H-NMR spectra and retention time on HPLC with those of an authentic sample.

(ii) Isolation of TsONa: Compound (**2**, 101 mg, 0.21 mmol) was dissolved in dry hot pyridine (10 ml) and the solution was evaporated to dryness *in vacuo*, taken up in dry hot pyridine (10 ml) and reevaporated to dryness. Finally, it was dried for 2 h under high vacuum. The residue was suspended in absolute MeOH (3 ml)–97% $H_2{}^{18}O$ (2.5 ml) solution and then 28% MeONa in MeOH (0.3 ml, 7 mol eq) was added under a dry nitrogen atmosphere. The mixture was refluxed for 2.5 h with stirring. The solution was evaporated *in vacuo*, taken up in absolute MeOH, reevaporated to dryness, and the whole operation was repeated once more. The residue was suspended in absolute MeOH and the resulting insoluble material was removed by filtration. The filtrate was concentrated to a small volume *in vacuo* and the residue was subjected to preparative TLC (MeOH : $CHCl_3$ = 3 : 7, v/v) to give TsONa (30 mg, 75%) as a colorless amorphous solid. The structure was confirmed by comparison of its IR, MS and ^{1}H-NMR spectra and retention time on HPLC with those of an authentic sample.

General Procedure for the Preparation of N^6,N^6-Dialkyl cAMPs (3) To a stirred solution of **2** (2.0 g, 4.1 mmol) in DMSO (24 ml) was added 60% sodium hydride (3—9 mol eq) and then an alkyl bromide (4.1—5.4 mol eq for **3b**—**d**, **3f**—**h**) or alkyl iodide (4.9 and 9.5 mol eq for **3a** and **3e**) was added dropwise to the mixture. The mixture was stirred at room temperature for 3 h—2 d, then H_2O–MeOH (60 ml : 60 ml for **3a**—**e**, 30 ml : 70 ml for **3f**—**h**) was added followed by detosylation with 2 N NaOH (6—14 mol eq) for 2—5 d at room temperature. The solution was adjusted to about pH 7 with conc. HCl and evaporated *in vacuo*. The residue was dissolved in a small amount of H_2O, adjusted to pH 2 with 2 N HCl, and applied to a charcoal column (2.2 × 18 cm). After being washed with H_2O, the column was eluted with EtOH–H_2O–28% aqueous NH_3 (10 : 10 : 1, v/v). The eluate was collected and evaporated to dryness *in vacuo*. The resulting residue was dissolved in H_2O, adjusted to pH 2 with 2 N HCl, and purified by preparative TLC (MeOH : $CHCl_3$ = 3 : 7—4 : 6, v/v) to give the desired product. An analytical sample was obtained by treatment with column chromatography on Dowex 50w-x8 (H^+) (eluent: EtOH–H_2O).

3a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 2950, 2900, 1660, 1600. ^{1}H-NMR δ: 3.45 (6H, s, 2 × CH_3), 4.67 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.02 (1H, s, H-1′), 8.24 and 8.33 (1H each, s, purine H's).

3b: A colorles amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3420, 3100, 2980, 2955, 2900, 1640, 1600, 1555. ^{1}H-NMR δ: 1.20 (6H, t, J=7.0 Hz, 2 × CH_3), 3.96 (4H, br s, $N(CH_2)_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

3c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3410, 2950, 2925, 2870, 1640, 1585, 1565. ^{1}H-NMR δ: 0.89 (6H, t, J=7.3 Hz, 2 × CH_3), 1.65 (4H, q, J=7.3 Hz, 2 × $C\underline{H}_2CH_3$), 3.84 (4H, br s, $N(CH_2)_2$), 4.66 (1H, d, J=4.9 Hz, H-2′), 4.85—5.05 (1H, m, H-3′), 6.00 (1H, s, H-1′), 8.22 and 8.31 (1H each, s, purine H's).

3d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3420, 2950, 2925, 2860, 1640, 1585, 1565. ^{1}H-NMR δ: 0.91 (6H, t, J=7.1 Hz, 2 × CH_3), 1.20—1.45 (4H, m, 2 × $C\underline{H}_2CH_3$), 1.50—1.75 (4H, m, $N(CH_2C\underline{H}_2)_2$), 3.89 (4H, br s, $N(CH_2)_2$), 4.68 (1H, d, J=4.9 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.00 (1H, s, H-1′), 8.22 and 8.31 (1H each, s, purine H's).

3e: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3420, 2950, 2925, 2855, 1635, 1585, 1565. ^{1}H-NMR δ: 0.87 (12H, t, J=6.6 Hz, 4 × CH_3), 2.00—2.25 (2H, m, 2 × CH), 3.80 (4H, br s, $N(CH_2)_2$), 4.69 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.00 (1H, s, H-1′), 8.22 and 8.33 (1H each, s, purine H's).

3f: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3425, 2950, 2925, 2850, 1640, 1585, 1565. ^{1}H-NMR δ: 0.87 (6H, t, J=6.6 Hz, 2 × CH_3), 1.20—1.45 (8H, m, 2 × $(C\underline{H}_2)_2CH_3$), 1.55—1.75 (4H, m, $N(CH_2C\underline{H}_2)_2$), 3.89 (4H, br s, $N(CH_2)_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.21 and 8.31 (1H each, s, purine H's).

3g: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3250, 2950, 2920, 2850, 1635, 1585, 1565. ^{1}H-NMR δ: 0.75—0.95 (6H, m, 2 × CH_3), 1.30 (12H, s like, 2 × $(C\underline{H}_2)_3CH_3$), 1.50—1.75 (4H, m, $N(CH_2C\underline{H}_2)_2$), 3.88 (4H, br s, $N(CH_2)_2$), 4.65 (1H, d, J=4.4 Hz, H-2′), 4.85—5.05 (1H, m, H-3′), 5.99 (1H, s, H-1′), 8.21 and 8.30 (1H each, s, purine H's).

3h: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3010, 2910, 1630, 1585, 1575. ^{1}H-NMR δ: 4.70 (1H, d, J=5.1 Hz, H-2′), 4.85—5.05 (1H, m, H-3′), 5.24 (4H, br s, $N(CH_2)_2$), 6.04 (1H, s, H-1′), 7.20—7.45 (10H, m, phenyl H's), 8.32 and 8.37 (1H each, s, purine H's).

***N*^{6}-Methyl-2′-*O*-tosyl cAMP (4a)** Methyl iodide (137 g, 0.96 mol) was added to a solution of triethylammonium salt of **1** (100 g, 0.3 mol) in DMF (1.4 l), and the mixture was stirred at 40 °C overnight. The resulting precipitate was filtered off, washed with acetone, and dried *in vacuo* to afford the iodide salt of 1-methyl cAMP (104 g, 73%) as a white powder.[12a] UV $\lambda_{max}^{0.1\,N\,HCl}$ nm: 259; $\lambda_{max}^{0.1\,N\,NaOH}$ nm: 261, 267sh, 300sh.

The iodide salt of 1-methyl cAMP (99 g, 0.21 mmol) was dissolved in a mixture of H_2O (200 ml) and 28% aqueous NH_3 (35 ml). The solution was stirred at 60 °C for 11 h and adjusted to pH 7 with 2 N HCl followed by evaporation *in vacuo*. The residue was dissolved in EtOH–H_2O (200 ml each) and adjusted to pH 2 with 2 N HCl to give N^6-methyl cAMP (54 g, 75%) as a white powder. UV $\lambda_{max}^{H_2O}$ nm: 264; $\lambda_{max}^{0.1\,N\,NaOH}$ nm: 262; $\lambda_{max}^{0.1\,N\,NaOH}$ nm: 266.

A solution of tosyl chloride (46 g, 24 mmol) in dioxane (300 ml) was added to a solution of N^6-methyl cAMP (16.5 g, 4.8 mmol) in 110 ml of aqueous NaOH (4.65 g, 11 mmol). The mixture was stirred at room temperature for 2 h and H_2O (400 ml) was added to the solution. The resulting precipitate was removed by filtration and the filtrate was concentrated to about 250 ml *in vacuo*. After extraction of TsOH with $CHCl_3$ (100 ml), the precipitate that had been deposited was filtered off, washed with H_2O and dried *in vacuo* to give **4a** as a white powder. The liquor was treated once more with $CHCl_3$ (100 ml) to give **4a** as a white powder. Total yield was 84% (20.1 g). IR ν_{max}^{KBr} cm^{-1}: 3640, 3430, 3250, 3020, 2930, 2820, 1680, 1620, 1600, 1180. ^{1}H-NMR δ: 2.32 (3H, s, $PhC\underline{H}_3$), 3.02 (3H, br s, NCH_3), 5.05—5.18 (1H, m, H-3′), 5.59 (1H, d, J=5.6 Hz, H-2′), 6.20 (1H, s, H-1′), 7.29 and 7.70 (2H each, d, J=8.4 Hz, phenyl H's), 8.01 (1H, br s, NH), 8.16 and 8.24 (1H each, s, purine H's). UV ν_{max}^{EtOH} nm (ε): 263 (16200). *Anal.* Calcd for $C_{18}H_{20}N_5O_8PS \cdot 1/4H_2O$: C, 43.09; H, 4.09; N, 13.95. Found: C, 43.11; H, 4.00; N, 13.93.

General Procedure for the Preparation of N^6-Alkyl-2′-*O*-tosyl cAMPs (4b—d) An aldehyde (13—14 mol eq) was added to a suspension of **2** (7.24 g, 15 mmol) in AcOH (130 ml) with stirring, and after 0.5 h sodium cyanoborohydride (5.8—8.0 mol eq) was added to the mixture. The solution was heated at 50 °C for 1—2 d. A small amount of H_2O was added and the reaction solution was evaporated *in vacuo*. The residue was purified by the methods described below.

4b: The residue was dissolved in H_2O–MeOH, adjusted to pH 2 with 2 N HCl, and applied to chromatography on charcoal (3.2 × 28 cm). After being washed with H_2O, the column was eluted with EtOH–H_2O–28% aqueous NH_3 (10 : 10 : 1, v/v). The eluate was evaporated *in vacuo* and the resulting residue was chromatographed on silica gel (120 g, MeOH : $CHCl_3$ = 3 : 17, v/v). The resulting ammonium salt of **4b** was dissolved in H_2O–MeOH followed by adjustment to pH 2 with 2 N HCl to afford **4b**

(78%) as a white powder. IR ν_{max}^{KBr} cm^{-1}: 3600, 3445, 3250, 3020, 2980, 2900, 2875, 1680, 1620, 1600, 1180. ^{1}H-NMR δ: 1.20 (3H, t, J=7.2 Hz, CH_3), 2.33 (3H, s, PhC$\underline{H}_3$), 3.59 (2H, br s, NCH_2), 5.05—5.18 (1H, m, H-3′), 5.60 (1H, d, J=6.6 Hz, H-2′), 6.19 (1H, s, H-1′), 7.29 and 7.70 (2H each, d, J=7.8 Hz, Phenyl H's), 7.97 (1H, br s, NH), 8.16 and 8.21 (1H each, s, purine H's). UV λ_{max}^{EtOH} nm (ε): 264 (16200). *Anal.* Calcd for $C_{19}H_{22}N_5O_8PS \cdot 3/2H_2O$: C, 42.38; H, 4.68; N, 13.00. Found: C, 42.53; H, 4.42; N, 12.99.

4c: The residue was worked up in a similar manner to that of **4b** to give the desired product in 50% yield as a white powder. IR ν_{max}^{KBr} cm^{-1}: 3440, 3070, 3020, 2955, 2930, 2875, 1675, 1615, 1600, 1180. ^{1}H-NMR δ: 0.92 (3H, t, J=7.2 Hz, CH_3), 1.63 (2H, q, J=7.2 Hz, C$\underline{H}_2$CH$_3$), 2.32 (3H, s, PhC$\underline{H}_3$), 3.57 (2H, br s, NCH_2), 5.03—5.17 (1H, m, H-3′), 5.60 (1H, d, J=6.3 Hz, H-2′), 6.19 (1H, s, H-1′), 7.29 and 7.70 (2H each, d, J=8.2 Hz, phenyl H's), 8.03 (1H, br s, NH), 8.16 and 8.21 (1H each, s, purine H's). UV λ_{max}^{EtOH} nm (ε): 264 (16200). *Anal.* Calcd for $C_{20}H_{24}N_5O_8PS \cdot 3/2H_2O$: C, 43.48; H, 4.92; N, 12.67. Found: C, 43.65; H, 4.76; N, 12.64.

4d: The residue was dissolved in H_2O (500 ml) and extracted with $CHCl_3$ (2 × 80 ml). The combined extracts were dried over Na_2SO_4 and evaporated *in vacuo*. The residue was chromatographed on silica gel (125 g, MeOH : $CHCl_3$ = 1 : 7—7 : 13, v/v). The resulting sodium salt of **4d** was dissolved in H_2O–MeOH and adjusted to pH 2 with 2 N HCl to afford the desired product in 54% yield as a white powder. IR ν_{max}^{KBr} cm^{-1}: 3430, 3100, 3040, 2950, 2930, 2865, 1675, 1620, 1600sh, 1180. ^{1}H-NMR δ: 0.91 (3H, t, J=7.2 Hz, CH_3), 1.25—1.45 (2H, m, C$\underline{H}_2$CH$_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.32 (3H, s, PhC$\underline{H}_3$), 3.57 (2H, br s, NCH_2), 5.03—5.17 (1H, m, H-3′), 5.60 (1H, d, J=5.6 Hz, H-2′), 6.19 (1H, s, H-1′), 7.29 and 7.70 (2H each, d, J=8.2 Hz, phenyl H's), 8.02 (1H, br s, NH), 8.16 and 8.21 (1H each, s, purine H's). UV λ_{max}^{EtOH} nm (ε): 264 (17300). *Anal.* Calcd for $C_{21}H_{26}N_5O_8PS \cdot 1/3H_2O$: C, 46.25; H, 4.89; N, 12.84. Found: C, 46.14; H, 4.90; N, 12.69.

General Procedure for the Preparation of N^6-R_1-N^6-R_2-2′-*O*-Tosyl cAMPs (5—8) To a stirred solution of **4** (3 mmol) in DMSO (40 ml) was added 60% sodium hydride (3—4 mol eq) and then an alkyl bromide (3—4 mol eq) was added dropwise to the mixture. The solution was stirred at room temperature for 1—3 h and adjusted to pH 7 with 0.2 N HCl followed by evaporation *in vacuo*. The resulting residue was dissolved in H_2O and adjusted to pH 2 with 2 N HCl followed by extraction with $CHCl_3$. The extracts were dried over Na_2SO_4 and evaporated *in vacuo*. The residue was subjected to chromatography on silica gel (MeOH–$CHCl_3$) or preparative TLC (silica gel, MeOH–$CHCl_3$) to give the desired product. An analytical sample was obtained by treatment with column chromatography on Dowex 50w-x8 (H^+) (eluent: EtOH–H_2O) or recrystallization.

5a: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3440, 3050, 2950, 2920, 2860, 1640, 1590, 1170. ^{1}H-NMR δ: 0.93 (3H, t, J=7.1 Hz, CH_3), 1.20—1.45 (2H, m, C$\underline{H}_2$CH$_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.31 (3H, s, PhC$\underline{H}_3$), 3.38 (3H, br s, NCH_3), 4.03 (2H, br s, NCH_2), 5.05—5.15 (1H, m, H-3′), 5.62 (1H, d, J=5.5 Hz, H-2′), 6.17 (1H, s, H-1′), 7.27 and 7.69 (2H each, d, J=8.3 Hz, phenyl H's), 8.13 and 8.19 (1H each, s, purine H's).

5b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3425, 3050, 2950, 2920, 2850, 1640, 1590, 1170. ^{1}H-NMR δ: 0.88 (3H, t, J=6.0 Hz, CH_3), 1.20—1.45 (4H, m, $(C\underline{H}_2)_2CH_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.30 (3H, s, PhC$\underline{H}_3$), 3.38 (3H, br s, NCH_3), 4.00 (2H, br s, NCH_2), 4.90—5.08 (1H, m, H-3′), 5.61 (1H, d, J=4.9 Hz, H-2′), 6.11 (1H, s, H-1′), 7.25 and 7.67 (2H each, d, J=7.4 Hz, phenyl H's), 8.11 and 8.19 (1H each, s, purine H's).

5c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3425, 3070, 2950sh, 2920, 2850, 1640, 1590, 1175. ^{1}H-NMR δ: 0.88 (3H, t, J=6.0 Hz, CH_3), 1.30 (6H, s like, $(C\underline{H}_2)_3CH_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.23 (3H, s, PhC$\underline{H}_3$), 3.37 (3H, br s, NCH_3), 3.80—4.25 (5H, m, H-4′, 5′, NCH_2), 4.80—4.95 (1H, m, H-3′), 5.60 (1H, d, J=5.4 Hz, H-2′), 6.01 (1H, s, H-1′), 7.21 and 7.63 (2H each, d, J=8.1 Hz, phenyl H's), 8.11 and 8.18 (1H each, s, purine H's).

5d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3425, 3030, 2920, 2850, 1640, 1590, 1175. ^{1}H-NMR δ: 0.75—0.90 (3H, m, CH_3), 1.15—1.40 (8H, m, $(C\underline{H}_2)_4CH_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.27 (3H, s, PhC$\underline{H}_3$), 3.37 (3H, br s, NCH_3), 3.98 (2H, br s, NCH_2), 4.80—4.95 (1H, m, H-3′), 5.60 (1H, d, J=6.3 Hz, H-2′), 6.01 (1H, s, H-1′), 7.21 and 7.63 (2H each, d, J=8.4 Hz, phenyl H's), 8.10 and 8.18 (1H each, s, purine H's).

5e: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3410, 3025, 2910, 2850, 1640, 1590, 1175. ^{1}H-NMR δ: 0.84 (3H, s like, CH_3), 1.24 (10H, s like, $(C\underline{H}_2)_5CH_3$), 1.50—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.62 (3H, s, PhC$\underline{H}_3$), 3.37 (3H, br s, NCH_3), 3.90 (2H, br s, NCH_2), 4.83—4.97 (1H, m, H-3′), 5.61 (1H, d, J=5.9 Hz, H-2′), 6.02 (1H, s, H-1′), 7.20 and 7.63 (2H each, d, J=7.1 Hz, phenyl H's), 8.11 and 8.18 (1H each, s, purine H's).

5f: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3430, 3080, 3020, 2980, 2910, 1640, 1590, 1175. ^{1}H-NMR δ: 2.28 (3H, s, PhC$\underline{H}_3$), 3.37 (3H, br s, NCH_3), 5.00—5.15 (1H, m, H-3′), 5.29 (2H, br s, NCH_2), 5.63 (1H, d, J=6.6 Hz, H-2′), 6.17 (1H, s, H-1′), 7.20—7.45 (7H, m, phenyl H's), 7.79 (2H, d, J=8.1 Hz, phenyl H's), 8.15 and 8.25 (1H each, s, purine H's).

6a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3075, 2950, 2925, 2860, 1640, 1590, 1170. ^{1}H-NMR δ: 0.92 (3H, t, J=7.2 Hz, CH_3), 1.21 (3H, t, J=7.0 Hz, CH_3), 1.55—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.30 (3H, s, PhC$\underline{H}_3$), 3.86 (4H, br s, $N(CH_2)_2$), 5.00—5.15 (1H, m, H-3′), 5.62 (1H, d, J=5.9 Hz, H-2′), 6.14 (1H, s, H-1′), 7.26 and 7.69 (2H each, d, J=8.1 Hz, phenyl H's), 8.12 and 8.18 (1H each, s, purine H's).

6b: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3420, 3055, 2950, 2910, 2860, 1635, 1585, 1170. ^{1}H-NMR δ: 0.93 (3H, t, J=7.3 Hz, CH_3), 1.21 (3H, t, J=6.8 Hz, CH_3), 1.25—1.45 (2H, m, C$\underline{H}_2$CH$_3$), 1.55—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.30 (3H, s, PhC$\underline{H}_3$), 3.91 (4H, br s, $N(CH_2)_2$), 5.05—5.17 (1H, m, H-3′), 5.63 (1H, d, J=5.6 Hz, H-2′), 6.16 (1H, s, H-1′), 7.27 and 7.70 (2H each, d, J=8.3 Hz, phenyl H's), 8.12 and 8.19 (1H each, s, purine H's).

6c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3050, 2950, 2925, 2850, 1640, 1590, 1170. ^{1}H-NMR δ: 0.89 (3H, t, J=6.8 Hz, CH_3), 1.20 (3H, t, J=6.8 Hz, CH_3), 1.25—1.45 (4H, m, $(C\underline{H}_2)_2CH_3$), 1.55—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.30 (3H, s, PhC$\underline{H}_3$), 3.89 (4H, br s, $N(CH_2)_2$), 5.05—5.20 (1H, m, H-3′), 5.63 (1H, d, J=6.1 Hz, H-2′), 6.16 (1H, s, H-1′), 7.27 and 7.69 (2H each, d, J=8.3 Hz, phenyl H's), 8.13 and 8.19 (1H each, s, purine H's).

6d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3070, 2950, 2925, 2850, 1640, 1590, 1170. ^{1}H-NMR δ: 0.80—0.95 (3H, m, CH_3), 1.20 (3H, t, J=6.8 Hz, CH_3), 1.32 (6H, s like, $(C\underline{H}_2)_3CH_3$), 1.55—1.78 (2H, m, $NCH_2C\underline{H}_2$), 2.28 (3H, s, PhC$\underline{H}_3$), 3.90 (4H, br s, $N(CH_2)_2$), 4.90—5.05 (1H, m, H-3′), 5.61 (1H, d, J=5.4 Hz, H-2′), 6.06 (1H, s, H-1′), 7.23 and 7.67 (2H each, d, J=7.8 Hz, phenyl H's), 8.12 and 8.18 (1H each, s, purine H's).

6e: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3080, 2950, 2925, 2850, 1640, 1590, 1170. ^{1}H-NMR δ: 0.80—0.95 (3H, m, CH_3), 1.20 (3H, t, J=6.8 Hz, CH_3), 1.10—1.40 (8H, m, $(C\underline{H}_2)_4CH_3$), 1.55—1.75 (2H, m, $NCH_2C\underline{H}_2$), 2.28 (3H, s, PhC$\underline{H}_3$), 3.70—4.20 (7H, m, H-4′, 5′, $N(CH_2)_2$), 4.75—4.88 (1H, m, H-3′), 5.55 (1H, d, J=6.6 Hz, H-2′), 6.00 (1H, s, H-1′), 7.23 and 7.65 (2H each, d, J=8.3 Hz, phenyl H's), 8.10 and 8.18 (1H each, s, purine H's).

6f: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3445, 3050, 3025, 2975, 2930, 2900, 1640, 1590, 1175. ^{1}H-NMR δ: 1.17 (3H, t, J=6.8 Hz, CH_3), 2.28 (3H, s, PhC$\underline{H}_3$), 3.92 (2H, br s, NCH_2), 5.05—5.20 (1H, m, H-3′), 5.26 (2H, br s, $NC\underline{H}_2$Ph), 5.66 (1H, d, J=5.9 Hz, H-2′), 6.19 (1H, s, H-1′), 7.15—7.40 (7H, m, phenyl H's) 7.71 (2H, d, J=8.3 Hz, phenyl H's), 8.17 and 8.24 (1H each, s, purine H's).

7a: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3445, 3055, 2950, 2900, 2870, 1640, 1590, 1175. ^{1}H-NMR δ: 0.89 (3H, t, J=7.3 Hz, CH_3), 0.91 (3H, t, J=7.2 Hz, CH_3), 1.20—1.45 (2H, m, C$\underline{H}_2$CH$_3$), 1.50—1.80 (4H, m, $N(CH_2C\underline{H}_2)_2$), 2.31 (3H, s, PhC$\underline{H}_3$), 3.86 (4H, br s, $N(CH_2)_2$), 5.00—5.17 (1H, m, H-3′), 5.63 (1H, d, J=5.6 Hz, H-2′), 6.16 (1H, s, H-1′), 7.27 and 7.70 (2H each, d, J=8.5 Hz, phenyl H's), 8.11 and 8.19 (1H each, s, purine H's).

7b: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3430, 3055, 3030, 2950, 2925, 2860, 1640, 1590, 1175. ^{1}H-NMR δ: 0.75—1.00 (6H, m, 2 × CH_3), 1.20—1.45 (4H, m, $(C\underline{H}_2)_2CH_3$), 1.50—1.80 (4H, m, $N(CH_2C\underline{H}_2)_2$), 2.30 (3H, s, PhC$\underline{H}_3$), 3.86 (4H, br s, $N(CH_2)_2$), 5.00—5.17 (1H, m, H-3′), 5.62 (1H, d, J=5.6 Hz, H-2′), 6.16 (1H, s, H-1′), 7.27 and 7.70 (2H each, d, J=8.3 Hz, phenyl H's), 8.12 and 8.19 (1H each, s, purine H's).

7c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3100, 3030, 2955, 2930, 2860, 1590, 1180. ^{1}H-NMR δ: 0.78—1.00 (6H, m, 2 × CH_3), 1.32 (6H, s like, $(C\underline{H}_2)_3CH_3$), 1.52—1.78 (4H, m, $N(CH_2C\underline{H}_2)_2$), 2.28 (3H, s, PhC$\underline{H}_3$), 3.92 (4H, br s, $N(CH_2)_2$), 4.75—4.90 (1H, m, H-3′), 5.56 (1H, d, J=5.9 Hz, H-2′), 6.00 (1H, s, H-1′), 7.22 and 7.65 (2H each, d, J=8.1 Hz, phenyl H's), 8.08 and 8.18 (1H each, s, purine H's).

7d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3450, 3055, 3030, 2960, 2925, 2875, 1640, 1590, 1175. ^{1}H-NMR δ: 0.87 (3H, t, J=7.3 Hz, CH_3), 1.64 (2H, q, J=7.3 Hz, C$\underline{H}_2$CH$_3$), 2.28 (3H, s, PhC$\underline{H}_3$), 3.84 (2H, br s, NCH_2), 5.00—5.18 (1H, m, H-3′), 5.27 (2H, br s, $NC\underline{H}_2$Ph), 5.66 (1H, d, J=6.1 Hz, H-2′), 6.18 (1H, s, H-1′), 7.15—7.40 (7H, m, phenyl H's), 7.71 (2H, d, J=8.3 Hz, phenyl H's), 8.15 and 8.24 (1H each, s, purine H's).

8a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3550, 3430, 3100, 3030, 2955, 2930, 2860, 1590, 1180. ^{1}H-NMR δ: 0.80—1.00 (6H, m, 2 × CH_3), 1.20—1.45 (6H, m, C$\underline{H}_2$CH$_3$, $(C\underline{H}_2)_2H_3$), 1.50—1.75 (4H, m, $N(CH_2C\underline{H}_2)_2$), 2.27 (3H, s, PhC$\underline{H}_3$), 3.88 (4H, br s, $N(CH_2)_2$), 4.80—4.95 (1H, m, H-3′),

5.61 (1H, d, J=6.4 Hz, H-2′), 6.00 (1H, s, H-1′), 7.20 and 7.65 (2H each, d, J=8.1 Hz, phenyl H's), 8.07 and 8.17 (1H each, s, purine H's).

8b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3445, 3060, 2950, 2930, 2860, 1640, 1590, 1180. ^{1}H-NMR δ: 0.75—1.00 (6H, m, 2×CH$_3$), 1.15—1.45 (8H, m, CH$_2$CH$_3$, (CH$_2$)$_3$CH$_3$), 1.50—1.75 (4H, m, N(CH$_2$CH$_2$)$_2$), 2.30 (3H, s, PhCH$_3$), 3.87 (4H, brs, N(CH$_2$)$_2$), 4.95—5.11 (1H, m, H-3′), 5.62 (1H, d, J=6.1 Hz, H-2′), 6.11 (1H, s, H-1′), 7.25 and 7.69 (2H each, d, J=7.8 Hz, phenyl H's), 8.12 and 8.18 (1H each, s, purine H's).

8c: A white powder. IR ν_{max}^{KBr} cm^{-1}: 3430, 3055, 3025, 2950, 2930, 2870, 1640, 1590, 1180. ^{1}H-NMR δ: 0.88 (3H, t, J=7.1 Hz, CH$_3$), 1.20—1.40 (2H, m, CH$_2$CH$_3$), 1.50—1.70 (2H, m, NCH$_2$CH$_2$), 2.26 (3H, s, PhCH$_3$), 3.84 (2H, brs, NCH$_2$), 4.95—5.10 (1H, m, H-3′), 5.26 (2H, brs, NCH$_2$Ph), 5.64 (1H, d, J=5.6 Hz, H-2′), 6.10 (1H, s, H-1′), 7.15—7.40 (7H, m, phenyl H's), 7.69 (2H, d, J=8.3 Hz, phenyl H's), 8.12 and 8.23 (1H each, s, purine H's).

General Procedure for the Preparation of N^6-R_1-N^6-R_2 cAMPs (9—11) A solution of 2 N NaOH (5—11 mol eq) was added to a suspension of **5—8** (0.8—2.3 mmol) in MeOH–H_2O (1:1—2:1, v/v, total volume: 40—100 ml) with stirring and the solution was heated at 50—60 °C for 0.5—2 d. The solution was evaporated *in vacuo* and the residue was dissolved in a small amount of H_2O, adjusted to pH 2 with 2 N HCl, followed by washing with $CHCl_3$.

Compound (**9**) was purified by the method described below. The aqueous layer was concentrated to a small volume and subjected to column chromatography on Dowex 50w-8x (H^+) with EtOH–H_2O to give the desired product.

Compounds (**10—12**) were purified by the method described below. The aqueous layer was concentrated to a small volume and subjected to column chromatography on charcoal. After being washed with H_2O, the column was eluted with EtOH–H_2O–28% aqueous NH_3 (10:10:1, v/v) and the eluate was evaporated *in vacuo*. The resulting residue was dissolved in a small amount of MeOH, adjusted to pH 2 with 2 N HCl, and purified by preparative TLC with MeOH–$CHCl_3$ or column chromatography on silica gel (eluent: MeOH–$CHCl_3$) to give the desired product. An analytical sample was obtained by treatment with column chromatography on Dowex 50w-x8 (H^+) (eluent: EtOH–H_2O).

9a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3250, 3080, 2950, 2925, 2860, 1640, 1590. ^{1}H-NMR δ: 0.91 (3H, t, J=7.3 Hz, CH$_3$), 1.20—1.45 (2H, m, CH$_2$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.38 (3H, brs, NCH$_3$), 4.00 (2H, brs, NCH$_2$), 4.67 (1H, d, J=5.1 Hz, H-2′), 4.95—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.33 (1H each, s, purine H's).

9b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3090, 2950, 2925, 2850, 1645, 1590. ^{1}H-NMR δ: 0.87 (3H, t, J=6.5 Hz, CH$_3$), 1.20—1.45 (4H, m, (CH$_2$)$_2$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.38 (3H, brs, NCH$_3$), 4.00 (2H, brs, NCH$_2$), 4.67 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

9c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3250, 3100, 2950, 2925, 2850, 1645, 1590. ^{1}H-NMR δ: 0.85 (3H, t, J=6.1 Hz, CH$_3$), 1.18—1.45 (6H, m, (CH$_2$)$_3$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.39 (3H, brs, NCH$_3$), 4.01 (2H, brs, NCH$_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.95—5.05 (1H, m, H-3′), 6.02 (1H, s, H-1′), 8.23 and 8.33 (1H each, s, purine H's).

9d: A Colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3080, 2950, 2925, 2850, 1640, 1590. ^{1}H-NMR δ: 0.84 (3H, t, J=6.2 Hz, CH$_3$), 1.15—1.40 (8H, m, (CH$_2$)$_4$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.38 (3H, brs, NCH$_3$), 4.01 (2H, brs, NCH$_2$), 4.68 (1H, d, J=5.4 Hz, H-2′), 4.93—5.08 (1H, m, H-3′), 6.02 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

9e: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3375, 3100, 2920, 2850, 1645, 1590. ^{1}H-NMR δ: 0.84 (3H, t, J=6.3 Hz, CH$_3$), 1.10—1.40 (10H, m, (CH$_2$)$_5$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.39 (3H, brs, NCH$_3$), 4.00 (2H, brs, NCH$_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.08 (1H, m, H-3′), 6.02 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

9f: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 3025, 2925, 2850, 1640, 1590. ^{1}H-NMR δ: 3.36 (3H, brs, NCH$_3$), 4.69 (1H, d, J=4.9 Hz, H-2′), 4.92—5.07 (1H, m, H-3′), 5.31 (2H, brs, NCH$_2$), 6.04 (1H, s, H-1′), 7.15—7.40 (5H, m, phenyl H's), 8.29 and 8.35 (1H each, s, purine H's).

10a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3430, 3100, 2950, 2900, 1635, 1600. ^{1}H-NMR δ: 0.91 (3H, t, J=7.3 Hz, CH$_3$), 1.19 (3H, t, J=7.0 Hz, CH$_3$), 1.66 (2H, brs, q, J=7.0 Hz, CH$_2$CH$_3$), 3.91 (4H, brs, N(CH$_2$)$_2$), 4.64 (1H, d, J=4.7 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.31 (1H each, s, purine H's).

10b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 2950, 2925, 2855, 1640, 1585. ^{1}H-NMR δ: 0.92 (3H, t, J=7.3 Hz, CH$_3$), 1.19 (3H, t, J=6.7 Hz, CH$_3$), 1.25—1.45 (2H, m, CH$_2$CH$_3$), 1.55—1.75 (2H, m, NCH$_2$CH$_2$), 3.93 (4H, brs, N(CH$_2$)$_2$), 4.62 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

10c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 2950, 2920, 2855, 1640, 1585. ^{1}H-NMR δ: 0.88 (3H, t, J=4.6 Hz, CH$_3$), 1.19 (3H, t, J=6.5 Hz, CH$_3$), 1.25—1.45 (4H, m, (CH$_2$)$_2$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.92 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=5.4 Hz, H-2′), 4.90—5.08 (1H, m, H—3′), 6.01 (1H, s, H-1′), 8.22 and 8.32 (1H each, s, purine H's).

10d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 2950, 2920, 2850, 1640, 1585. ^{1}H-NMR δ: 0.86 (3H, t, J=6.5 Hz, CH$_3$), 1.19 (3H, t, J=6.8 Hz, CH$_3$), 1.31 (6H, slike, (CH$_2$)$_3$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.90 (4H, brs, N(CH$_2$)$_2$), 4.66 (1H, d, J=4.6 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.31 (1H each, s, purine H's).

10e: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3250, 3100, 2950, 2915, 2850, 1635, 1585. ^{1}H-NMR δ: 0.70—0.95 (3H, m, CH$_3$), 1.19 (3H, t, J=6.8 Hz, CH$_3$), 1.05—1.45 (8H, m, (CH$_2$)$_4$CH$_3$), 1.50—1.75 (2H, m, NCH$_2$CH$_2$), 3.92 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

10f: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3440, 3005, 2955, 2905, 1635, 1585. ^{1}H-NMR δ: 1.16 (3H, t, J=6.8 Hz, CH$_3$), 3.91 (2H, brs, NCH$_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 5.24 (2H, brs, NCH$_2$Ph), 6.03 (1H, s, H-1′), 7.28 (5H, s, phenyl H's), 8.28 and 8.33 (1H each, s, purine H's).

11a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3250, 3100, 2955, 2935, 2875, 1640, 1590. ^{1}H-NMR δ: 0.89 (3H, t, J=7.3 Hz, CH$_3$), 0.91 (3H, t, J=7.2 Hz, CH$_3$), 1.20—1.45 (2H, m, CH$_2$CH$_3$), 1.50—1.80 (4H, m, N(CH$_2$CH$_2$)$_2$), 3.90 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=5.1 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.23 and 8.32 (1H each, s, purine H's).

11b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3250, 3100, 2955, 2935, 2875, 1635, 1585. ^{1}H-NMR δ: 0.80—0.95 (6H, m, 2×CH$_3$), 1.20—1.45 (4H, m, (CH$_2$)$_2$CH$_3$), 1.50—1.75 (4H, m, N(CH$_2$CH$_2$)$_2$), 3.88 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=4.9 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.22 and 8.32 (1H each, s, purine H's).

11c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 2960, 2935, 2870, 1640, 1590. ^{1}H-NMR δ: 0.89 (6H, t, J=7.2 Hz, 2×CH$_3$), 1.29 (6H, slike, (CH$_2$)$_3$CH$_3$), 1.50—1.75 (4H, m, N(CH$_2$CH$_2$)$_2$), 3.89 (4H, brs, N(CH$_2$)$_2$), 4.66 (1H, d, J=4.6 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.00 (1H, s, H-1′), 8.22 and 8.32 (1H each, s, purine H's).

11d: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3375, 3100, 3005, 2955, 2925, 2850, 1640, 1590. ^{1}H-NMR δ: 0.86 (3H, t, J=7.3 Hz, CH$_3$), 1.50—1.75 (2H, m, CH$_2$CH$_3$), 3.86 (2H, brs, NCH$_2$), 4.70 (1H, d, J=5.4 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 5.27 (2H, brs, NCH$_2$Ph), 6.03 (1H, s, H-1′), 7.27 (5H, s, phenyl H's), 8.27 and 8.35 (1H each, s, purine H's).

12a: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3410, 2950, 2930, 2860, 1590. ^{1}H-NMR δ: 0.91 (6H, t, J=7.3 Hz, 2×CH$_3$), 1.32 (6H, slike, CH$_2$CH$_3$, (CH$_2$)$_2$H$_3$), 1.50—1.75 (4H, m, N(CH$_2$CH$_2$)$_2$, 3.90 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=4.4 Hz, H-2′), 4.88—5.05 (1H, m, H-3′), 6.01 (1H, s, H-1′), 8.32 and 8.35 (1H each, s, purine H's).

12b: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3425, 2950, 2930, 2855, 1590. ^{1}H-NMR δ: 0.75—1.00 (3H, m, CH$_3$), 0.91 (3H, t, J=7.0 Hz, CH$_3$), 1.30 (8H, slike, CH$_2$CH$_3$, (CH$_2$)$_3$CH$_3$), 1.50—1.75 (4H, m, N(CH$_2$CH$_2$)$_2$), 3.89 (4H, brs, N(CH$_2$)$_2$), 4.68 (1H, d, J=5.4 Hz, H-2′), 4.90—5.05 (1H, m, H-3′), 6.00 (1H, s, H-1′), 8.21 and 8.31 (1H each, s, purine H's).

12c: A colorless amorphous solid. IR ν_{max}^{KBr} cm^{-1}: 3400, 3100, 3005, 2950, 2920, 2850, 1635, 1585. ^{1}H-NMR δ: 0.87 (3H, t, J=7.1 Hz, CH$_3$), 1.17—1.42 (2H, m, CH$_2$CH$_3$), 1.48—1.72 (2H, m, NCH$_2$CHC$_2$), 3.90 (2H, brs, NCH$_2$), 4.70 (1H, d, J=5.1 Hz, H-2′), 4.90—5.03 (1H, m, H-3′), 5.27 (2H, brs, NCH$_2$Ph), 6.03 (1H, s, H-1′), 7.26 (5H, s, phenyl H's), 8.26 and 8.34 (1H each, s, purine H's).

Biological Activity Male albino guinea pigs weighing 320—680 g were stunned by a blow on the head. The hearts were rapidly removed and the right atria and the papillary muscle of the right ventricle dissected out in cold bathing solution, and were supended individually in 8 ml organ baths for recording isometric contractions. The bathing solutions was Krebs–Henseleit's solution (32±0.1 °C) containing, NaCl 118 (mM); KCl 4.7; $CaCl_2$ 2.5; $NaHCO_3$ 25; $MgSO_4$ 1.2; KH_2PO_4 1.2: glucose 11 and was continuously bubbled with 95% O_2+5% CO_2. The initial tensions of 0.5 and 0.25 g were applied to the atria and papillary muscle preparations, respectively. After 30 min, the optimal resting tension was determined and maintained thereafter. The right atrium was allowed to beat spontaneously

and the papillary muscle was stimulated by square wave pulses of 1 ms duration at a frequency of 1 Hz, and at voltages of about 50% above the threshold supplied by a square-wave pulse stimulator (Nihon Kohden MSE-3) *via* a pair of silver-plated electrodes between which the preparations were placed. The isometric contraction was measured by a force-displacement transducer (Tokyo Baldwin T7-30-240) connected to a carrier-amplifier (Nihon Kohden RP-5) and the heart rate was counted by a cardiotachometer (Nihon Kohden RT-5). All the measurements were recorded on a thermostylus recorder (Watanabe Sokki Linear Corder Mark V). An equilibration period of 60 min was allowed before starting the experiments. Because of the low solubility of cAMP derivatives, they were dissolved in Krebs–Henseleit's solution and applied to the preparation by replacing less than 1.2 ml of the bathing solution. The inotropic and chronotropic effects of cAMP derivatives were expressed as percent change from the maximum response evoked by 10^{-7} M isoproterenol in each preparation.

Acknowledgments The authors are grateful to Professor T. Hino of Chiba University for his helpful advice and to Associate Professor M. Nakagawa of Chiba University for her encouragement. The authors also thank Mr. T. Matsudo for MS measurements.

References and Notes

1) Part VI: S, Kataoka, J. Imai, N. Yamaji, M. Kato, T. Kawada, and S. Imai, *Chem. Pharm. Bull.*, **38**, 1596 (1990).
2) W. R. Kukovetz and G. Poech, *Arch. Pharmakol. Exp. Pathol.*, **266**, 236 (1970); S. Imai, T. Otorii, K. Takeda, Y. Katano, and D. Horii, *Jpn. J. Pharmacol.*, **24**, 499 (1974); N. Yamaji, M. Kato, I. Matsubara, N. Shimamoto, K. Miura, and S. Imai, *Chem. Pharm. Bull.*, **28**, 1683 (1980); S, Kataoka, J. Isono, N. Yamaji, and M. Kato, *Chem. Lett.*, **1986**, 1221.
3) S. Kataoka, J. Isono, N. Yamaji, M. Kato, T. Kawada, and S. Imai, *Chem. Pharm. Bull.*, **36**, 2212 (1988).
4) R. B. Meyer, D. A. Shuman, R. K. Robins, R. J. Baver, M. K. Dimmitt, and L. N. Simon, *Biochemistry*, **11**, 2704 (1972).
5) S. O. Doskeland, D. Ogreid, R. Ekanger, P. A. Starm, J. P. Miller, and R. K. Suva, *Biochemistry*, **22**, 1094 (1983).
6) K. K. Ogilivie, K. L. Sadana, E. A. Thompson, M. A. Quilliam, and J. B. Westmore, *Tetrahedron Lett.*, **1974**, 2861; Y. Hayakawa, S. Wakabayashi, H. Kato, and R. Noyori, *J. Am. Chem. Soc.*, **112**, 1691 (1990); Y. S. Sanghri, B. K. Bhattacharya, G. D. Kini, S. S. Matsumoto, S. B. Larson, W. B. Jolley, R. K. Robins, and G. R. Renankar, *J. Med. Chem.*, **33**, 336 (1990).
7) A. M. Mian, R. Harris, R. W. Sidwell, R. K. Robins, and J. A. Khwaja, *J. Med. Chem.*, **17**, 259 (1974).
8) W. D. Closson, P. Wriede, and S. Bank, *J. Am. Chem. Soc.*, **88**, 1581 (1966); L. J. Dolby, and D. R. Rosencrantz, *J. Org. Chem.*, **28**, 1888 (1963).
9) C. A. Bunton and Y. F. Frei, *J. Chem. Soc.*, **1951**, 1872.
10) *a*) N. R. Williams, *Adv. Carbohyd. Chem. Biochem.*, **25**, 109 (1970); *b*) J. G. Buchanan, D. M. Clode, and N. Vethaviyasar, *J. Chem. Soc., Perkin Trans. 1*, **1976**, 1449.
11) Y. Ishido, N. Nakazaki, and N. Sakairi, *J. Chem. Soc., Perkin Trans. 1*, **1979**, 2088; I. Ekiel, E. Durzynkiewicz, L. Dudycz, and D. Shugar, *Biochemistry*, **17**, 1538 (1978).
12) *a*) S. Shibuya, A. Tanaka, and H. Yoshino, Japan. Kokai Patent 49-92094 (1974) [*Chem. Abstr.*, **82**, 140457*x* (1975)]; *b*) K. H. Boswell, L. F. Christensen, D. H. Shuman, and R. K. Robins, *J. Heterocycl. Chem.*, **12**, 1 (1975); *c*) J. P. Miller, K. H. Boswell, R. B. Meyer, L. F. Christensen, and R. K. Robins, *J. Med. Chem.*, **23**, 242 (1980).
13) Partition coefficients of N^6,N^6-dimethyl (**3a**), diethyl (**3b**), dibutyl (**3d**) derivatives (0.1 mmol) between octanol and 0.01 M phosphate buffer (pH 7.0) were measured at 37 °C. The amount of dialkyl derivatives in the aqueous phase at equilibrium after agitating overnight was measured by UV spectroscopy (274 nm for **3a** and **3b**, 278 nm for **3d**). The concentration of dialkyl derivatives in the octanol phase was determined by the difference. Partition coefficients (concentration in octanol/concentration in buffer) of **3a** and **3b** were 0 and 0.01, respectively; that of **3d** was 0.59.
14) J. P. Miller, C. C. Sigman, H. L. Johnson, T. Novinson, R. N. Springen, K. Senga, D. E. O'Brien, and R. K. Robins, "Advances in Cyclic Nucleotide and Protein Phosphorylation Research," Vol. 16, Raven Press, New York, 1984, pp. 277—290.