5′-Amido Analogs of Adenosine 3′,5′-Cyclic Monophosphate[1]

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5′-Amido analogs of adenosine 3′,5′-cyclic monophosphate (**7**) are reported. Synthesis may proceed *via* the unusual cyclic diester amidates **6**. The *N*-*n*-octyl-*N*-5′-deoxyadenosyl 3′,5′-cyclic phosphoramidate (**7d**) is particularly apt for biochemical testing because of its chemical stability.

Adenosine 3′,5′-cyclic monophosphate (3′,5′-AMP, c-AMP) has been found in almost all animal tissues studied. It has also been shown to be present in bacteria, slime molds, and even higher plants. c-AMP plays a key role in regulating biochemical responses of cells, tissues or organs to external stimuli on different levels of organization.[2-4] Although its significance in mediating or moderating various hormones has stimulated a large number of investigations, the detailed mechanism of c-AMP action is not yet understood.

Derivatives or analogs of c-AMP in living systems could interfere with synthesis, degradation, or activity of the cyclic nucleotide.[5,6] Distribution and rate of cleavage by phosphodiesterases of such derivatives or analogs in tissues may differ from that of the parent compound.[7] Furthermore, they could be expected to either mimic or antagonize the actions of the cyclic nucleotides[8] and may permeate membranes more easily.

Several analogs of c-AMP have been checked for activity, *e.g.*, tubercidin 3′,5′-monophosphate,[9] the 3′-methylene cyclic phosphonate analog of cyclic AMP,[10] the 5′-methylene cyclic phosphonate analog of cyclic AMP,[10] and adenosine 3′,5′-monothionophosphate.[11] An unsuccessful attempt to prepare the 5′-amido analog of adenosine 3′,5′-cyclic monophosphate was recently reported.[12]

We prepared a series of 5′-amido analogs of adenosine 3′,5′-cyclic monophosphate (**7**)[1b] starting from 5′-tosyladenosine (**1**), which by aminolysis was converted into the toluenesulfonate salts of 5′-amino-5′-deoxyadenosine (**2**).[13] The free amines were obtained either by absorption to an acidic ion exchanger, elution of *p*-toluenesulfonic acid and desorption of **3** by 1 *N* ammonia, or alternatively by treatment with potassium *tert*-butoxide in, *e.g.*, methanol.

The amino derivatives (**3**) were phosphorylated with di(*p*-nitrophenyl) phosphochloridate[14,15] to give the diaryl phosphoramidates (**4**). Sterically hindered amines like cyclohexylamino derivative **3e** resisted phosphorylation by this method.

Treatment of **4** (except **4a**) with a pyridine–concentrated ammonia–water mixture (2:5:3) or 1 *N* NaOH in methanol yielded the 5′-*N*-alkyl- and 5′-*N*-benzylamino-5′-deoxyadenosine 3′,5′-cyclic phosphoramidates (**7**) directly. Compound **4a** is much more sensitive to alkali than **4b–d**. Treatment with the pyridine–ammonia mixture or with NaOH produced a number of decomposition products.

Synthesis of **7a** could be achieved in two steps. In the first step, one *p*-nitrophenyl group of **4a** was removed with 1 *M* triethylamine in pyridine containing 1 equiv of water. The resulting monoaryl phosphoramidate (**5a**) was cyclized with 1 *M* potassium *tert*-butoxide–*tert*-butyl alcohol in dimethyl sulfoxide following Borden and Smith's procedure.[16] Ring closure of **4** to **7** probably proceeds *via* the unusual cyclic diester amide **6**.[17] After a reaction time up to 2 hr we were able to isolate **6**.

The hydrolytic stability of the c-AMP analogs **7a–d** has been examined in view of possible biochemical activity (*cf.* Experimental Section, Table II). The *n*-octyl derivative **7d** was reasonably stable in buffer systems of pH 9–6, presumably due to shielding by the coiled *n*-octyl residue. Minimum stability in this range is shown by the *N*-methyl derivative **7a**.

The newly prepared 5′-amino-5′-deoxynucleosides **3a–e** act as competitive inhibitors for adenosine kinase.[18] Tests with the enzyme adenosine deaminase[19] showed that 5′-amino-5′deoxyadenosine with a small additional substituent at the amine (*e.g.*, **3b**) are still accepted as substrates, although at a rate reduced by approximately three powers of magnitude as compared with adenosine. The derivatives **3c–e** which have bulkier substituents, are not accepted as substrates and do not act as inhibitors either. Biochemical experiments with **7** in collaboration with other research groups are under way.

Experimental Section

Uv spectra were recorded on a Cary 14 spectrophotometer. For nmr measurements, a Varian HA-100 spectrometer was used, and for mass spectra an AEI-MS-9 mass spectrometer at 70 eV was used.

Melting points (uncorrected) were determined with a Kofler hot-stage microscope apparatus. Chromatographic separations

(1) (a) Part IV: Nucleotide Analogs with P–N Bonds. (b) A. Murayama, B. Jastorff, and H. Hettler, *Angew. Chem., Int. Ed. Engl.*, **9**, 640 (1970), should be considered part III.

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(15) The detailed procedure is given in the Experimental Section.

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(17) Mechanism and stereochemistry of the cyclization reaction **4** → **6** will be discussed elsewhere.

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Table I.—Physical Constants and Chromatographic Data of Compounds Prepared

Compd and net formula (mol wt)	No.	Mp, °C	Elemental analysis, %[b] C	H	N	S	Uv (MeOH) max, mμ	Nmr,[a] δ, ppm	Mass spectrum m/e (rel intensity)	E^b	R_f, pc A	B	R_f, tlc[c] C	D	A
5′-Deoxyadenosylmethylammonium *p*-toluenesulfonate $C_{18}H_{24}N_6O_6S$ (453.1)	**2b**		47.58 (47.89)	5.77 (5.60)	18.49 (18.86)	7.06 (7.04)	259	2.26 (s, 3) 2.58 (s, 3) 7.11 (d, 2, J = 8 Hz) 7.46 (d, 2, J = 8 Hz)		−0.05	0.52	0.54			0.87
5′-*N*-Benzylamino-5′-deoxyadenosine $C_{17}H_{20}N_6O_3$ (356.4)	**3c**		57.29 (57.41)	5.66 (5.61)	23.58 (23.70)		259	7.28 (s, 5)	357 (<1, $M^+ + 1$) 136 (98, B + 2)[d] 135 (70, B + 1) 91 (100)	0.1	0.73	0.76		0.39	1.15
5′-*N*-*n*-Octylamino-5′-deoxyadenosine $C_{18}H_{30}N_6O_3$ (378.4)	**3d**	169	57.27 (57.36)	7.99 (8.07)	22.21 (22.33)		259	0.83 (m, 3) 1.24 (s, 12) 1.5–1.8 (m, 2)	380 (<1, $M^+ + 2$) 378 (<1, M^+) 136 (100, B + 2) 135 (40, B + 1)		0.86	0.85		0.19	1.18
5′-*N*-Cyclohexylamino-5′-deoxyadenosine $C_{16}H_{24}N_6O_3$ (348.4)	**3e**		55.15 (55.00)	6.94 (6.90)	24.11 (24.18)		259	1.0–2.0 (m, 10)	349 (<1, $M^+ + 1$) 136 (100, B + 2) 135 (50, B + 1)	0.2	0.8	0.77		0.10	1.22
Di-*O*-(*p*-nitrophenyl) *N*-(5′-deoxyadenosyl)phosphoramidate $C_{22}H_{21}N_8O_{10}P$ (588.4)	**4a**	223	44.94 (44.78)	3.60 (3.96)	18.06 (18.89)		266	7.51 (dd, 4, J = 8 Hz) 8.12 (dd, 6, J = 8 Hz)[e]				0.89	1.20	1.1	
O-(*p*-Nitrophenyl) *N*-(5′-deoxyadenosyl)-phosphoramidate $C_{16}H_{18}N_7O_8P$ (467.3)	**5a**						262 298 (shoulder) 260			0.38	0.50	0.71		0.33	0.92
Di-*O*-(*p*-nitrophenyl) *N*-methyl-*N*-(5′-deoxyadenosyl)phosphoramidate $C_{23}H_{23}N_8O_{10}P$ (602.5)	**4b**	184	45.86 (45.89)	3.85 (3.98)	18.60 (18.45)			2.83 (d, 3, J = 12 Hz) 7.41 (dd, 4, J = 8 Hz) 8.21 (dd, 4, J = 8 Hz)	139 (80, PNP)[f] 136 (65, B + 2) 135 (100, B + 1)		0.90 0.87	0.88 0.89	1.13 1.47	1.09 1.21	
Di-*O*-(*p*-nitrophenyl) *N*-benzyl-*N*-(5′-deoxyadenosyl)phosphoramidate $C_{29}H_{27}N_8O_{10}P$ (678.6)	**4c**	130	51.33 (51.19)	4.01 (4.25)	16.52 (16.36)		263	7.3 (m, 11)[g] 8.12 (m, 6)[e]	139 (100, PNP) 136 (99, B + 2) 135 (19, B + 1) 91 (78)						
Di-*O*-(*p*-nitrophenyl) *N*-*n*-octyl-*N*-(5′-deoxyadenosyl)phosphoramidate $C_{30}H_{37}N_8O_{10}P$ (700.6)	**4d**	88	51.43 (51.46)	5.33 (5.38)	16.01 (15.94)		262	0.83 (m, 3) 1.08 (s, 12) 1.5–1.8 (m, 2) 7.42 (dd, 4, J = 8 Hz) 8.11 (dd, 6, J = 8 Hz)[e]	700 (<1, M^+) 139 (80, PNP) 136 (22, B + 2) 135 (100, B + 1)		0.88	0.93	1.34	1.28	
O-(*p*-Nitrophenyl) *N*-methyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate $C_{17}H_{18}N_7O_7P$ (463.3)	**6b**	160	44.06 (43.92)	3.91 (3.99)	21.15 (21.00)		260	2.83 (d, 3, J = 12 Hz) 5.1 (q, 1, J = 5, 12 Hz) 7.52 (d, 2, J = 8 Hz) 8.11 (d, 2, J = 8 Hz)	463 (<1, M^+) 139 (40, PNP) 136 (100, B + 2) 135 (98, B + 1)		0.77	0.81	1.33	0.98	
O-(*p*-Nitrophenyl) *N*-benzyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate $C_{23}H_{22}N_7O_7P$ (539.4)	**6c**	134	51.20 (51.15)	4.11 (4.41)	18.18 (17.88)		262	5.3 (q, 1, J = 5, 12 Hz) 7.35 (m, 5) 7.5 (d, 2, J = 8 Hz) 8.11 (d, 2, J = 8 Hz)	539 (<1, M^+) 139 (99, PNP) 136 (98, B + 2) 135 (100, B + 1)		0.84	0.91	1.52	1.18	

O-(*p*-Nitrophenyl) *N*-*n*-octyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate $C_{24}H_{32}N_7O_7P$ (561.5)	**6d**	168	51.33 (51.17)	6.75 (6.89)	17.47 (17.56)	262	0.82 (m, 3) 1.22 (s, 12) 1.4–1.7 (m, 2) 5.28 (q, 1, J = 5, 12 Hz) 7.53 (d, 2, J = 8 Hz) 8.12 (d, 2, J = 8 Hz)	561 (<1 M^+) 139 (78, PNP) 136 (95, B + 2) 135 (100, B + 1)	0.90	0.92	1.26
N-(5′-Deoxyadenosyl) 3′,5′-cyclic phosphoramidate $C_{10}H_{13}N_6O_5P$ (328.2)	**7a**					260			0.46	0.31	0.43	0.06
N-Methyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate, Na salt $C_{11}H_{14}N_6O_5PNa$ (364.3)	**7b**		36.27 (36.31)	3.88 (4.00)	23.08 (23.21)	259			0.45	0.41	0.40	0.08
N-Benzyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate, Na salt $C_{17}H_{18}N_6O_5PNa$ (440.4)	**7c**					259			0.34	0.60	0.75	0.17
N-*n*-Octyl-*N*-(5′-deoxyadenosyl) 3′,5′-cyclic phosphoramidate, Na salt $C_{18}H_{28}N_6O_5PNa$ (462.5)	**7d**					259			0.37	0.85	0.85	0.26

[a] Nmr data are only reported for groups representative of the compound. [b] Electrophoretic mobilities refer to TMP. [c] R_f values refer to thymidine. [d] B = base. [e] *p*-Nitrophenyl and base. [f] PNP = *p*-nitrophenol. [g] *p*-Nitrophenyl, benzyl and amino groups of the base. [h] Found values in parentheses.

on paper were carried out on Schleicher and Schüll 2043 b mgl in solvent system A [1-propanol–ammonia–water (7:2:1)] or solvent system B [ethanol–1 M ammonium acetate (7:3)] by the descending technique.

For analytical tlc we used commercial silica plates Merck F_{254} in solvent system C [chloroform–methanol (85:15)], D [acetone–benzene–water (8:2:1)], or A. Separations on a preparative scale were carried out on silica plates Merck PF_{254} with solvents C and D or on silica gel columns, using an LKB fraction collector.

Electrophoresis was performed on Whatman 3 MM paper; buffer system: 0.1 M triethylammonium bicarbonate, pH 7.4 (E). DEAE cellulose was purchased from Whatman. All R_f values reported are reproducible with sufficient accuracy.

Physical constants and chromatographic data of compounds prepared are given in Table I. The yields given in the individual procedures refer to materials as described in Table I.

Derivatives of 5′-Amino-5′-deoxyadenosine (3). Reaction Procedure with Volatile Amines. 5′-Deoxyadenosylmethylammonium *p*-Toluenesulfonate (2b).—Liquid methylamine (20 ml) was condensed to 5′-*O*-tosyladenosine (1) (2.1 g, 5 mmol) in a pressure flask equipped with a safety valve. The closed system was kept at room temperature for 3 days. After evaporation of the excess methylamine, **2b** was obtained in quantitative yield as a white, hygroscopic powder.

Reaction Procedure with Liquid Amines. 5-Deoxyadenosyl-*n*-octylammonium *p*-Toluenesulfonate (2d).—To compound 1 (5 g, 11.8 mmol), *n*-octylamine (40 ml) was added and the mixture was kept at room temperature for 5 days. Ether (400 ml) was added and **2d** was filtered off, yield 4.8 g (74%). In the same way the tosylates of the following compounds were prepared: 5′-*N*-benzylamino-5′-deoxyadenosine (**3c**) in quantitative yield; 5′-*N*-cyclohexylamino-5-deoxyadenosine (**3e**), yield 80%.

Preparation of Free Amino Nucleosides (3) from 2 by an Ion Exchange Process.—The ammonium salt **2** (1 mmol) in a small volume of water was applied to a column filled with 25 ml of acidic Dowex 50 ion exchanger. *p*-Toluenesulfonic acid was washed off with distilled water, and finally the free amino nucleosides (**3**) were eluted with 1 N ammonia in almost quantitative yield using a fraction collector.

Preparation of Free Amino Nucleosides (3) from 2 by Treatment with Potassium *tert*-Butoxide in Methanol.—The ammonium salt **2** (1 mmol) was dissolved with stirring in anhydrous methanol (20 ml). Potassium *tert*-butoxide (1.01 mmol) was added. After approximately 5 min, anhydrous ether (40 ml) was added, and potassium *p*-toluenesulfonate was filtered off. After evaporation of the solvent the free amino nucleosides were obtained. Yields were in the range of 90%.

The amino nucleosides with lipophilic residues R are sparingly water soluble and can therefore be prepared by dissolving the ammonium tosylates in alkaline aqueous solution and filtering **3** off. The yields are somewhat lower (about 70%).

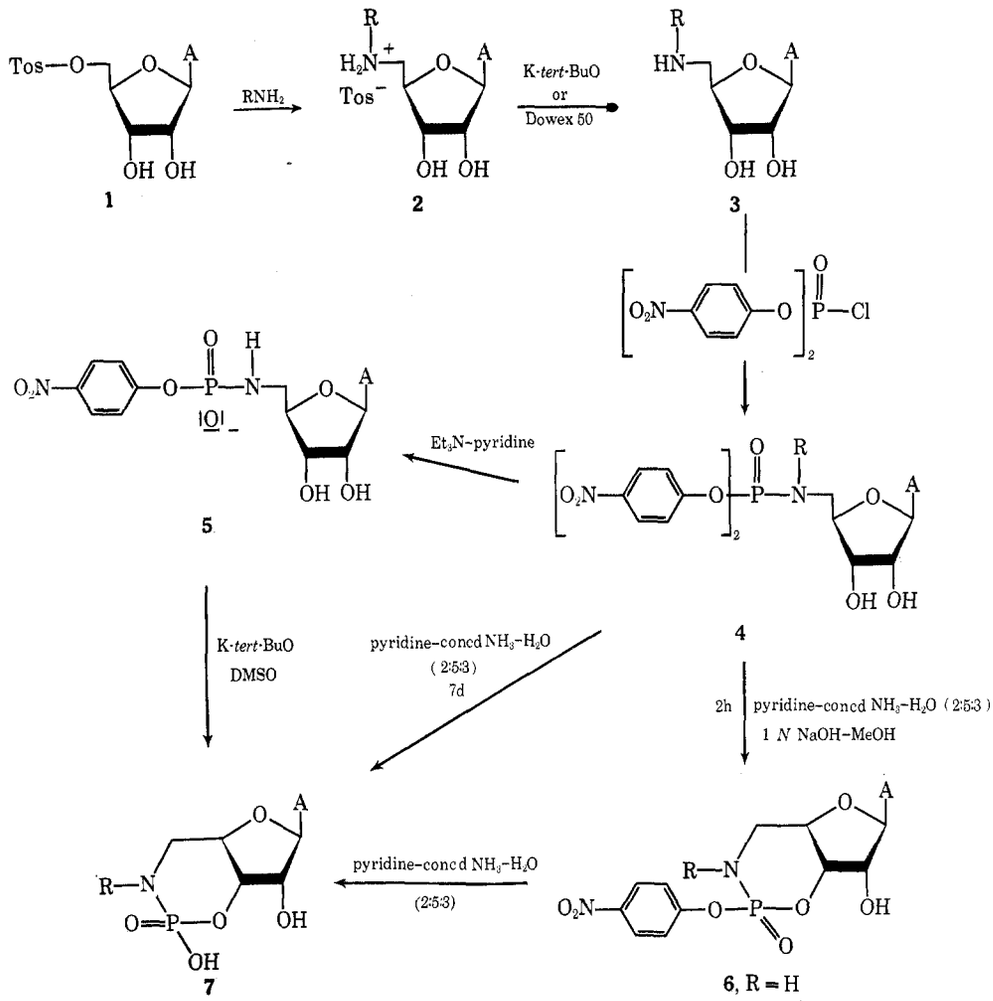
Amino nucleosides (**3**) prepared by one of the above methods were sufficiently pure for further synthetic steps. In order to obtain analytical samples, the compounds were purified on a silica gel column with solvent A.

Preparation of Hydriodides from 2.—The nucleoside ammonium tosylate (**2b**) (0.5 mmol) was dissolved in a mixture of methanol (9 ml) and acetone (20 ml), and $LiI \cdot 2H_2O$ (0.55 mmol) was added. After 20 min the solvent was evaporated and the residue was dissolved in a mixture of benzene–methanol (1:1). The first crop of crystals usually contains lithium tosylate. Indicative for tosylate is a twin peak at 260 and 254 mμ in the uv. On standing in the refrigerator **2b** crystallizes out. The hydriodide **2b** was recrystallized from benzene–methanol–petroleum ether (bp 30–60°) at 4°.

Anal. Calcd for $C_{11}H_{17}N_6O_3I$ (hydriodide of 5′-deoxymethylamino-5′-deoxadenosine[20]): C, 32.31; H, 4.29; I, 31.00. Found: C, 32.55; H, 4.39; I, 29.92.

Di(*p*-nitrophenyl) Phosphorochloridate.[15]—Diphenyl phosphorochloridate (33 g, 0.125 ml) in a three-neck flask equipped with stirrer, thermometer, dropping funnel, and drying tubes was dissolved in dry carbon tetrachloride (50 ml). A mixture consisting of 30% anhydrous nitric acid plus 70% sulfuric acid monohydrate (29 ml) was added dropwise at a rate which permits keeping the reaction mixture at 10–12°. After 4 hr stirring at 10–15° the reaction mixture was extracted with a total of 500 ml of methylene chloride with exclusion of moisture. The methylene

(20) A complete X-ray analysis of the compound was carried out by W. Saenger, *FEBS (Fed. Eur. Biochem. Soc.) Lett.*, **10**, 81 (1970).



2, 3	R
a	H
b	methyl
c	benzyl
d	*n*-octyl
e	cyclohexyl

4, 6, 7	R
a	H
b	methyl
c	benzyl
d	*n*-octyl

chloride solution was made neutral by stirring with anhydrous calcium carbonate. The solvent was removed *in vacuo* and the residue was dissolved in a little chloroform. It can be precipitated with dry petroleum ether (bp 40–60°). After the solution was refluxed shortly the compound crystallized out as greenish prisms (85%), mp 93–95° (lit.[15] mp 97–97.5°).

Di-*O*-(*p*-nitrophenyl) *N*-(5′-Deoxyadenosyl)phosphoramidate (4a).—5′-Amino-5′-deoxyadenosine (**3a**) (1 mmol)[19] in dry pyridine (10 ml) was evaporated to dryness three times. The substance was dissolved in dry pyridine (20 ml) and triethylamine (840 μl, 6 mmol). With rapid stirring at −40°, di(*p*-nitrophenyl) phosphochloridate (0.394 g, 1.1 mmol)[15] was added in portions during 1 hr. The reaction mixture was kept overnight at −20°, and after filtration the solvent was evaporated off. The residue was dissolved in dioxane–methanol (250 ml, 6:4). The solvent was reduced to 20 ml and **4a** was separated on preparative silica plates with solvent D, yield 0.265 g (45%).

***O*-(*p*-Nitrophenyl) *N*-(5′-Deoxyadenosyl)phosphoramidate (5a).**—Compound **4a** (0.294 g, 0.5 mmol) was dissolved in 1 *M* triethylamine (50 ml) in undried pyridine and kept overnight at room temperature. The reaction mixture was evaporated to dryness and **5a** separated on a DEAE cellulose column using the following conditions: DE 52-cellulose (HCO_3^- form), volume 260 ml, linear gradient water → 0.1 *M* triethylammonium bicarbonate, 3 l. each, **5a** between 0.024 and 0.031 *M*, yield 220 mg (95%).

5′-Amino-5′-deoxyadenosine 3′,5′-Cyclic Phosphoramidate (7a).—The triethylammonium salt of the *p*-nitrophenyl ester amidate (**5a**) (135 mg, 0.235 mmol) in anhydrous pyridine (10 ml) was evaporated to dryness three times and dissolved in anhydrous dimethyl sulfoxide (23 ml). After addition of 1 *M* potassium *tert*-butoxide in *tert*-butyl alcohol (2.7 ml) the reaction mixture was kept at 16° for 45 min. Then it was poured into anhydrous ether (5 l.) and kept overnight at −18°. After filtration, the residue was dissolved in a small amount of water and the cyclophosphoric ester amidate (**7a**) isolated on a DEAE cellulose column in two steps. The residue was first applied to DEAE cellulose (HCO_3^- form) (200 ml) and all inorganic substances were washed off with water, since ion exchanger Dowex 50 even as ammonium salt catalyzes decomposition of **7a**. Nucleotide material was eluted with 0.03 *M* triethylammonium bicarbonate, evaporated to dryness, and made free of triethylammonium bicarbonate by repeated evaporation of dry ethanol.

The material was fractionated in a second step on DEAE cellulose using the following conditions: DE 52-cellulose (HCO_3^- form), volume 200 ml, linear gradient water → 0.1 *M* triethylammonium bicarbonate, 2 l. each, **7a** between 0.018 and 0.023 *M*, yield 52 mg (52%).

Di-*O*-(*p*-Nitrophenyl) *N*-(5′-Deoxyadenosyl)phosphoramidates (4b–d).—The respective aminonucleosides **3b–d** (1 mmol) were made anhydrous by repeated azeotropic distillation with dry pyridine, then dissolved or suspended in a mixture of dry pyridine (10 ml) and triethylamine (840 μl, 6 mmol). With rapid stirring at −35° di-*p*-nitrophenyl phosphochloridate (0.394 g, 1.1 mmol)[15] was added in portions. The reaction mixture was kept at 4° overnight. Finally a 0.5 *M* sodium bicarbonate solution (1 ml)

was added. The mixture was evaporated to dryness. After water was added (25 ml) the residue was extracted several times with ethyl acetate. The extract was dried with anhydrous sodium sulfate and the solvent was evaporated. The substances can be purified by column chromatography or preferentially by preparative tlc on silica plates using the following solvent systems: compound **4b** in chloroform–methanol (85:15), yield 35%; **4c** and **4d** in chloroform–methanol (90:10), yield 34 and 70%, respectively.

O-(*p*-Nitrophenyl) *N*-(5′-Deoxyadenosyl) 3′,5′-Cyclic Phosphoramidates (6b–d). With Pyridine–Ammonia.—The respective diester amidate **4b–d** (1 mmol) in a mixture of pyridine–concentrated ammonia–water (2:5:3) (300 ml) was kept at 45° for 2 hr. The solvent was removed *in vacuo*, and the residue was washed with weakly alkaline (pH 9) water.

With Aqueous NaOH–Methanol.—The respective diester amidate **4b–d** (1 mmol) was dissolved in 100 ml of methanol, and after addition of 1 *N* NaOH (20 ml) the mixture was kept at room temperature for 2 hr. After neutralization with dilute acetic acid, the mixture was evaporated to dryness and some weakly alkaline water (pH 9) was added. Further work-up of the two procedures is identical; the yields are approximately the same.

Crude compounds of type **6** were purified by column chromatography or preferentially by preparative tlc on silica plates with the following solvent systems: compound **6b** in chloroform–methanol (85:15), yield 91%; **6c** and **6d** in chloroform–methanol (90:10); yield 91% each. Compound **6b** can be crystallized from methanol by adding ethyl acetate; **6c** and **6d** were obtained as a colorless powder.

Direct Synthesis of *N*-(5′-Deoxyadenosyl) 3′,5′-Cyclic Phosphoramidates (7b–d) from 4.—The respective diester amidate **4b–d** (1 mmol) was dissolved in a pyridine–concentrated ammonia–water mixture (100 ml, 3:5:2) and kept at 40° for 7 days; 1 *N* NaOH (3 ml) was added and the solvent was removed *in vacuo*. The residue was dissolved in methanol (10 ml) and filtered. After addition of acetone (300 ml), the desired cyclophosphoramidates **7b–d** precipitated. The precipitate was washed with acetone.

Further purification was carried out on a DEAE cellulose column using the following conditions for the individual compounds.

7b: DE 52 (HCO_3^- form); volume 75 ml for 3000 OD, 0.2 mmol; linear gradient water → 0.1 *M* triethylammonium bicarbonate, 2 l. each; fraction 15 ml; between fraction 58 and 70, 0.022–0.26 *M*; yield 50%.

7c: DE 52 (HCO_3^- form); volume 40 ml for 400 OD, 0.026 mmol; linear gradient water → 0.1 *M* triethylammonium bicarbonate, 1 l. each; fraction 9 ml; between fraction 21 and 39, 0.01 and 0.02 *M*; yield 55%.

7d: DE 52 (HCO_3^- form); volume 40 ml for 400 OD, 0.026 mmol; linear gradient water → 0.1 *M* triethylammonium bicarbonate, 1 l. each; fraction 9 ml; between fraction 45 and 62, 0.02–0.028 *M*; yield 53%.

Conversion of 6 into 7.—Identical conditions as described for conversion of **4** into **7** were used for converting **6** into **7**.

Stability of Compounds 7a–d in Various Buffer Solutions.—Compounds **7a–d** (20 OD each) were incubated in buffer solution (pH 5, pH 7, and pH 9, 200 μl) at 37° for 5 hr (Table II). Then the mixture was separated on paper chromatography in solvent A. The extent of cleavage was determined spectroscopically.

TABLE II
PERCENTAGE CLEAVAGE OF 7 IN BUFFER

Compd	pH 5, %	pH 7, %	pH 9, %
7a	98	15	0
7b	100	40	0
7c	90	9	0
7d	45	4	0

Registry No.—**2b**, 30765-10-7; **2b** HI, 30461-85-9; **3c**, 30765-12-9; **3d**, 30765-13-0; **3e**, 30765-14-1; **4a**, 29845-63-4; **4b**, 29845-64-5; **4c**, 30765-17-4; **4d**, 30826-38-1; **5a**, 30765-18-5; **6b**, 29845-65-6; **6c**, 30765-20-9; **6d**, 30765-21-0; **7a**, 29845-61-2; **7b** Na salt, 30765-23-2; **7c** Na salt, 30765-24-3; **7d** Na salt, 30765-25-4.

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Mobile Keto Allyl Systems. X.[1a] The Thermal Decomposition of 2-(*o*-Methylbenzal)-3-amino-4,4-dimethyl-1-tetralones[1b]

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Several 2-(*o*-methylbenzal)-3-amino-4,4-dimethyl-1-tetralones (**4**) have been prepared and those possessing a hydrogen atom α to the nitrogen in the amino moiety were found to decompose thermally to yield 2-(*o*-methylbenzyl)-1,4-dihydro-4,4-dimethyl-1-ketonaphthalene (**8**). By the use of deuterium-labeling experiments, it has been shown that this α hydrogen atom is transferred to the benzylic position. Possible mechanisms are discussed.

In connection with other work, it was necessary to prepare several 2-(*o*-methylbenzal)-3-amino-4,4-dimethyl-1-tetralones and to study their thermal stability. Condensation of *o*-tolualdehyde with 4,4-dimethyl-1-tetralone yielded *trans*-2-(*o*-methylbenzal)-4,4-dimethyl-1-tetralone[2] (**1**) in high yield. Bromination[3] with *N*-bromosuccinimide gave 2-(α-bromo-*o*-methylbenzyl)-1,4-dihydro-4,4-dimethyl-1-ketonaphthalene (**2**). When **2** was allowed to react with cyclohexylamine, isopropylamine, and *tert*-butylamine in solvent benzene at room temperature, two products were obtained as had been observed in a similar case.[3] Besides the corresponding 2-[α-(amino)-*o*-methylbenzyl]-1,4-dihydro-4,4-dimethyl-1-ketonaphthalenes (**3**), the desired 2-(*o*-methylbenzal)-3-amino-4,4-dimethyl 1-tetralones (**4**) were obtained. It is the thermal decomposition of these compounds **4** with which this paper is concerned.

Results

While compounds **3a–c** and **4c** were stable to column chromatography, compounds **4a** and **4b** decomposed.

(1) (a) For paper IX in this series, see N. H. Cromwell, K. Matsumoto, and A. D. George, *J. Org. Chem.*, **36**, 272 (1971). (b) Presented at the 1970 Midwest Regional Meeting of the American Chemical Society, Lincoln, Nebr., Oct 1970, Abstract No. 502.

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(3) N. H. Cromwell and E. M. Wu, *J. Org. Chem.*, **33**, 1895 (1968).