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N→N Alkyl and Glycosyl Migrations of Purines and Pyrimidines. I.[1] A New Migration of 3-Alkyl-N^6-acyladenine[2]

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Benzyl, allyl and 3-methyl-2-butenyl groups were found to migrate from N-3 to N-9 or N-7 position of the N^6-acyladenines. On heating in the presence of hydrogen halide, 3-benzyl-N^6-acyladenines underwent intermolecular benzyl migration giving rise to 9-benzyl derivatives and a small amounts of the 7-benzyl isomers. 3-Methyl-2-butenyl group in N^6-acyltriacanthine migrated under similar conditions to afford the 9-(3-methyl-2-butenyl) derivatives, whereas mercuric halide or cyanide caused intramolecular rearrangement to give the 9-(1,1-dimethyl-2-propenyl) isomers.

Numerous investigation on N-alkylation and N-glycosylation of purines and pyrimidines have been carried out in relation to the preparation of nucleosides and to the mode of action of biological alkylating agents, and various procedure have been devised. One procedure, involving the condensation reaction of free purine or pyrimidine bases with phosphorylated sugar halides, has been examined in our laboratory.[4]

In the course of the synthetic studies on adenine nucleotide, a significant difference was noted in the reaction product distribution between N-glycosylation and N-alkylation of N^6-acyladenines. To elucidate the cause of this remarkable but seemingly unfounded difference, benzylation and other alkylation of acyladenines were re-investigated in detail. It was thereby found that benzyl, allyl and 3-methyl-2-butenyl groups migrated from N-3 to N-9 or N-7 position of the purine ring.

N-3→N-9 Benzyl Migration

Electrophilic substitution of adenine both by alkyl and glycosyl halides has occurred mainly at the N-3 and partly at the N-9 positions as follows: The reaction of adenine with benzyl, allyl, or 3-methyl-2-butenyl halide in N,N-dimethylformamide (DMF) has given the 3-alkyladenine accompanied by the 9-alkyl isomer.[5] The glycosylation of adenine by the protected ribofuranosyl or phosphoribofuranosyl halide has afforded the 3-glycosyl and the 9-glycosyl derivative.[4,6] On the other hand, benzylation of N^6-benzoyladenine in N,N-dimethylacetamide (DMA), in the absence of a proton acceptor, has been reported to afford 3-benzyl-N^6-benzoyladenine but the formation of the 9-benzyl isomer has not been described.[7] Glycosylation reaction of N^6-acyladenines or their chloromercuri salts with various glycosyl derivatives have been employed to obtain the 9-substituted adenine nucleosides, and in all cases only the

1) This series were taken from the Doctoral Thesis of M. Miyaki, Hokkaido University, 1968.
2) A part of this work was presented as preliminary communications; S. Shimizu and M. Miyaki, *Tetrahedron Letters*, **1965**, 2059; M. Miyaki, K. Iwase, and B. Shimizu *Chem. Pharm. Bull.* (Tokyo), **14**, 87 (1966).
3) Location: *2-58, 1-chome, Hiromachi, Shinagawa-ku, Tokyo.*
4) M. Asai, M. Miyaki, and B. Shimizu, *Chem. Pharm. Bull.* (Tokyo), **15**, 1856 (1967); M. Asai, M. Miyaki, and B. Shimizu, *Agr. Biol. Chem.* (Tokyo), **31**, 319 (1967).
5) J.A. Montgomery and H. J. Thomas, *J. Am. Chem. Soc.*, **85**, 2672(1963); J. A. Montgomery and H. J. Thomas, *ibid.*, **87**, 5442 (1965); N.J. Leonard and T. Fujii, *ibid.*, **85**, 3719 (1963); N.J. Leonard and J.A. Deyrup, *ibid.*, **82**, 6202 (1960); C.J. Abshire and L. Berlinguet, *Can. J. Chem.*, **42**, 1599 (1964).
6) N.J. Leonard and R.A. Laursen, *J. Am. Chem. Soc.*, **85**, 2026 (1963); N. J. Leonard and R. A. Laursen, *Biochemistry*, **4**, 354 (1965).
7) J.A. Montgomery and H.J. Thomas, *J. Heterocyclic Chem.*, **1**, 115 (1964).

9-glycosides have been isolated.[8] The reaction of N^6-benzoyladenine with the protected ribofuranosyl, 5′-phosphoribofuranosyl or glucopyranosyl halide in acetonitrile, DMA *etc.* at 40—60° for about 40 hr gave the 9-glycosyl derivative: no 3-glycosyl isomer was obtained.[4,9] The difference in the product distribution between benzylation and glycosylation of N^6-acyladenines suggested that the effect of acyl group at N^6 on alkylation and glycosylation was differed markedly, but no reasonable explanation for this difference could be made. To detect the N-substituted derivatives other than 3-benzyl-N^6-acyladenine, were–examined the benzylation of N^6-benzoyladenine carefully.

After the reaction of N^6-benzoyladenine (I) with benzyl bromide in DMF at 100° for 3 hr, the reaction mixture was treated with NH_4OH and then separated by chromatography on silica gel. 9-Benzyl-N^6-benzoyladenine (III) (19%) was obtained besides the 3-benzyl derivative (II) (49%), and a small amount of the 3,7-dibenzyl derivative (5%) was also isolated. The reaction of I with benzyl chloride under the reported conditions[7] also gave III accompanied by II. It was noteworthy that the yield of III varied depending on the reaction conditions such as temperature and time. Treatment of I with benzyl bromide in refluxing DMF for 3 hr gave mainly III and only 3% of II. When N^6-acetyladenine was treated with benzyl bromide at 60° for 20 hr the 3- and 9-benzyl derivatives were obtained; however, at 110° for 6 days the 9-benzyl isomer alone was isolated. Similar result were obtained in the case of allyl- or 3-methyl-2-butenyl bromide. The reaction of chloromercuri salt with benzyl bromide gave the 3- and 9-benzyl derivatives. The yields of the alkylation products are listed in Table I.

Table I Yield of 3- and 9-Alkyl Derivatives in Alkylation

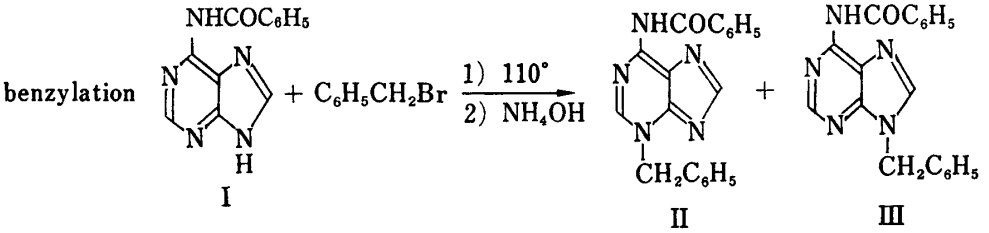
N^6-Acyladenine	Alkyl halide	Conditions	Yield (%)	
			Substituted 3-alkyl	Derivative 9-alkyl
Benzoyl	benzyl bromide	100°, 3 hr	49	19
Benzoyl	benzyl chloride	110°, 16 hr	45	41
Benzoyl	benzyl bromide	DMF, reflux, 3 hr	3	49
Acetyl	benzyl bromide	60°, 20 hr	55	29
Acetyl	benzyl bromide	110°, 140 hr	0	50
Benzoyl	allyl bromide	100°, 3 hr	48	16
Benzoyl	3-methyl-2-butenyl bromide	80°, 2 hr	16	12
Benzoyl	3-methyl-2-butenyl bromide	90°, 57 hr	0	47
Benzoyl (HgCl salt)	benzyl bromide	DMF, reflux, 3 hr	27	22

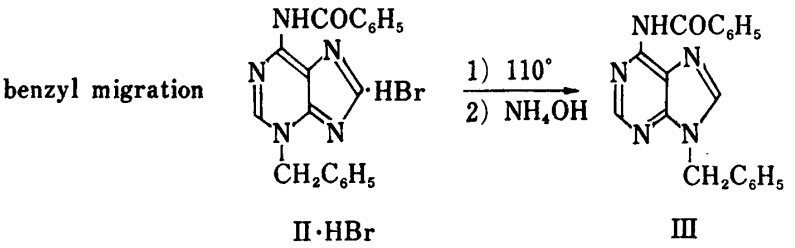
Subsequently the ratio of yields of the 3- and 9-benzyl isomers was followed by nuclear magnetic resonance (NMR) measurement of benzyl methylene protons. Benzylation at 110° in DMF initially gave more of the 3-benzyl derivative (II) than the 9-isomer (III), but prolonged heating of the reaction mixture increased the ratio of yield of III to II as indicated in Fig. 1. This change suggested that formation of III had proceeded *via* II. When II-hydrobromide was heated in DMF at the same temperature as in benzylation, conversion to III was observed as expected. After 70 hr, III was isolated in 60% yield by means of chromatography. The hydrochloride salt was also converted into III(34% yield) accompanied by a small amount of 7-benzyl-N^6-benzoyladenine (2%). A similar conversion of 3-benzyl-N^6-acetyladenine

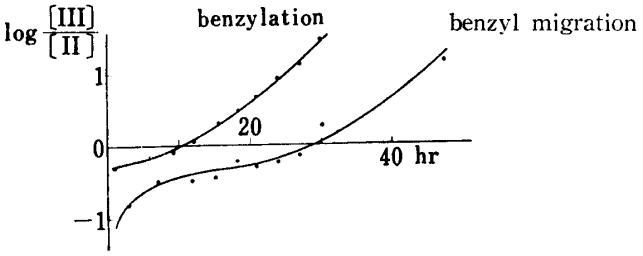
8) J. Davoll, B. Lythgoe, and A.R. Todd, *J. Chem. Soc.*, **1948**, 967, 1685; T. Ukita and H. Hayatsu, *J. Am. Chem. Soc.*, **84**, 1879 (1962); Y. Ishido and T. Sato, *Bull. Chem. Soc. Japan*, **34**, 347 (1961); J. Temple, R.L. Mckee, and J.A. Montgomery, *J. Org. Chem.* **28**, 2304 (1963); T. Nishimura, B. Shimizu, and I. Iwai, *Chem. Pharm. Bull.* (Tokyo), **12**, 352, 357, 1471 (1964); T. Nishimura and B. Shimizu, *Agr. Biol. Chem.* (Tokyo), **28**, 224 (1964); N. Yamaoka, K. Aso, and K. Matsuda, *J. Org. Chem.* **30**, 149 (1965).

9) B. Shimizu, M. Miyaki, and K. Iwase, Japanese Patent No. 536100.

hydrobromide gave 9-benzyl-N^6-acetyladenine in 32% yield. Importance of the acyl group in N^6 for the successful conversion was demonstrated by the fact that the hydrobromide of 3-benzyladenine did not undergo this migration at 110°. Later, it was found to react with difficulty at a higher temperature such as 180°, as will be described in the following paper.







〔II〕 : yield of 3-benzyl-N^6-benzoyladenine
〔III〕 : yield of 9-benzyl-N^6-benzoyladenine

Fig. 1. Ratio of the Yields of 3- and 9-Benzyl-N^6-benzoyladenines in Benzylation and Benzyl Migration

Nature of Benzyl Migration

Sample of 5×10^{-2}M, 5×10^{-3}M, and 5×10^{-4}M solutions of 3-benzyl N^6-benzoyladenine (II) hydrobromide in DMF was heated at 100—110° for 41 hr. Formation of an extremely small amount (2%) of the 9-benzyl derivative (III) in 5×10^{-4}M solution as compared with that (28%) in 5×10^{-2}M solution suggested that the N-3→N-9 migration was intermolecular in nature if the solvent effect on the migration reaction is not considered. These results however seemed to be unsatisfactory in explaining the process of the migration, because the debenzoylated product (3-benzyladenine) was also formed in the dilute solution.

A mixture of equimolar amounts of labeled 3-benzyl-N^6-benzoyladenine (II*) hydrobromide and cold 3-allyl-N^6-benzoyladenine (IV) hydrobromide was heated at 90—110° for 60 hr in DMF, and the products were isolated. The specific radioactivity of 9-benzyl-N^6-benzoyladenine (III*) was 62% of the activity of starting II*–HBr. In spite of the use of non-labeled 3-allyl derivative (IV) as starting material 9-allyl-N^6-benzoyladenine (V*) was found to have 60% specific activity of that of II*–HBr. Such a distribution of radioactivity suggested that the benzyl and allyl migration were intermolecular, however, the yield of V* was quite low (9% from IV) as compared with that of III* (42% from II*–HBr) (Chart 1).

When II–HBr was heated at 110° for 70 hr in DMF together with an equimolar amount of N^6-benzoyladenine-8-^{14}C (I*), the isolated 9-benzyl-N^6-benzoyladenine (III*) had 52% specific activity of that of the initial I*. A small amount of 7-benzyl-N^6-benzoyladenine was isolated

and found to have 52% activity (Chart 1). When the migration reaction in the presence of an equimolar amount of N^6-benzoyladenine-8-^{14}C was stopped after heating at 120° for only 4 hr, the recovered 3-benzyl derivative was found to have activity similar to 9-benzyl isomer, the migration product. Such labeling of the 3-benzyl derivative indicated that the benzoyl group released from N-3 of II by hydrogen bromide attacked N-3 of N^6-benzoyladenine to reform the 3-benzyl derivative.

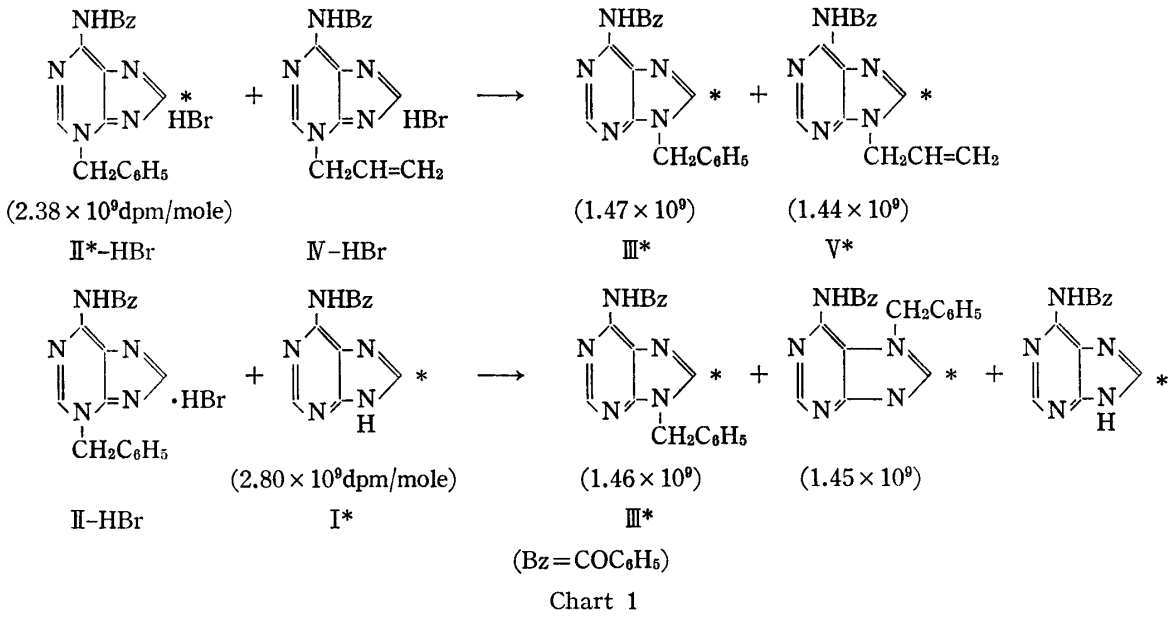


Chart 1

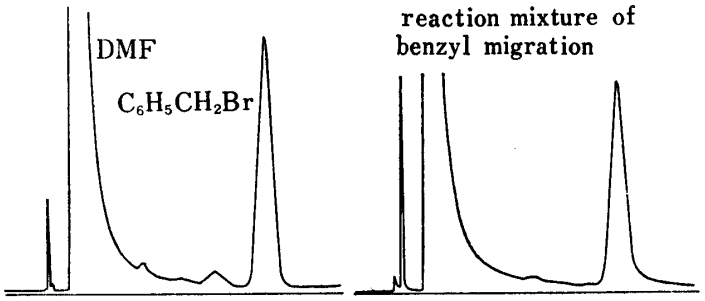


Fig. 2. Gas Chromatogram of Benzyl Migration Reaction Mixture

sample: 8.5 μl
column: 20% Silicon high vacuum grease-Chromosorb (40/60), 120°
carrier: N_2 100 ml/min, H_2 115 ml/min
detector: F.I.D.

A 0.013 M solution of II–HBr in DMF was analyzed by gas chromatography immediately after being heated at 160° for 20 min. The peak of benzyl bromide existed on the chromatogram of the reaction mixture after the peak of DMF, as shown in Fig. 2, whereas benzyl bromide was not detected before heating. Formation of benzyl bromide in the migration reaction mixture proved that the reaction involved an intermolecular migration via formation of benzyl bromide.

9-Benzyl-N^6-benzoyladenine (III) was heated at 110° for 70 hr with an equimolar amount of N^6-benzoyladenine-8-^{14}C (6.35×10^9 dpm/mole) (I*) in the presence of hydrogen bromide. The isolated III was found to have only 0.95% (5.96×10^7 dpm/mole) of the specific activity of I*, and no 3-benzyl derivative was isolated. This fact indicated that the migration was practically irreversible.

Allyl and 3-Methyl-2-butenyl Migration

On heating at 100—110° for 30 hr in DMF, 3-allyl-N^6-acyladenine hydrobromide underwent allyl migration, giving rise to 9-allyl-N^6-acyladenine in 19% yield. Mercuric halides were found to be more effective in inducing this migration. Heating of a mixture of 3-allyl-N^6-benzoyladenine (IV) and mercuric bromide or chloride at 110° for 24 hr resulted in allyl migration affording the 9-allyl derivative (V) in 31—42% yield.

Triacanthine[10] is the only 3-substituted adenine isolated from biological sources, and migration of 3-methyl-2-butenyl group was expected ot occur with the N^6-acyl derivative. When N^6-benzoyltriacanthine (VI) hydrobromide was heated at 95—100° for 40 hr in DMF N-3→N-9 migration occurred yielding 9-(3-methyl-2-butenyl)-N^6-benzoyladenine (VII) (20%). Although heating of VI with mercuric bromide at 110° for 30 hr caused the migration, only a small amount (3%) of VII was obtained. The major product (40%), similar to the 9-substituted N^6-benzoyladenine in ultraviolet (UV) spectra, differed from VII in melting point and R_f value by thin-layer chromatography. This isomer (VIII) was found to have a 1,1-dimethyl-2-propenyl group by comparison of its NMR spectrum with that of the 3-methyl-2-butenyl derivative (Fig. 3). In the spectra of two debenzoylated isomers, the peaks due to allylic systems were almost the same as those of the corresponding N^6-benzoyl derivatives. Formation of different products in the 3-methyl-2-butenyl migration suggested the migration proceeded by different mechanisms according to the catalyst employed.

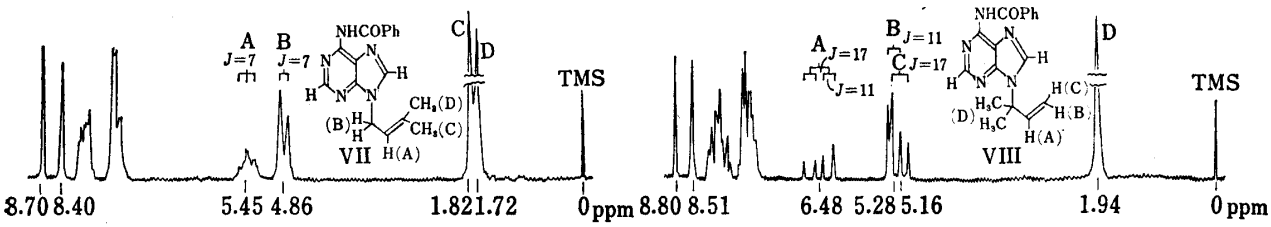


Fig. 3. NMR Spectra of VII and VIII in d_6-DMSO at 60 Mc

Nature of 3-Methyl-2-butenyl Migration

(1) Intermolecular Nature of Migration by Hydrogen Bromide: When 3-(3-methyl-2-butenyl)-N^6-benzoyladenine (VI) hydrobromide was heated with an equimolar amount of N^6-benzoyladenine-8-^{14}C (6.19×10^9 dpm/mole) (I*) at 110° for 24 hr in DMF, 9-(3-methyl-2-butenyl)-N^6-benzoyladenine (VII*) thereby formed had 51% (3.11×10^9 dpm/mole) of the specific activity of I*. Radioactivity of the recorved I* was reduced to 49% (3.06×10^9 dpm/mole). This fact suggested that the migration caused by hydrogen bromide was intermolecular in nature. If the 3-methyl-2-butenyl group was released as a cation the isomerization of the migrating group would occur to give the mixture of 3-methyl-2-butenyl- and 1,1-dimethyl-2-propenyl cations. However, no 9-(1,1-dimethyl-2-propenyl)-N^6-benzoyladenine (VIII) was isolated. Consequently, in prat, 3-methyl-2-butenyl bromide might be formed in this migration reaction as was benzyl bromide in the benzyl migration.

(2) Intramolecular Nature of Migration by Mercuric Bromide or Cyanide : The distribution of radioactivity among the products of the migration catalyzed by mercuric bromide was distinctly different from that by hydrogen bromide. A mixture of the equimolar amounts of VI, I* (3.08×10^9 dpm/mole) and mercuric bromide was heated in DMA at 95° for 17 hr and subsequently at 120° for 4 hr. The specific radioactivity of the major product, 9-(1,1-dimethyl-2-propenyl) isomer (VIII*) (39% yield), was 0.31×10^9 dpm/mole. This was extremely low as compared to that of the minor product, 9-(3-methyl-2-butenyl) derivative (8% yield) which had a considerably high specific activity (2.66×10^9 dpm/mole). The remaining 3-(3-methyl-2-butenyl) derivative (10%) also had a low specific activity, 0.32×10^9 dpm/mole. The recovered I* had been diluted to 2.33×10^9 dpm/mole in its radioactivity. The migration reaction at higher temperature such as 120° for 32 hr gave a smaller amount of radioactive VIII and a larger amount of radioactive VII, as shown in Table II (A,B), and at lower tem-

10) N.J. Leonard and J.A. Deyrup, *J. Am. Chem. Soc.*, **84**, 2148 (1962); A.S. Belikov, A.I. Bankowsky, and M.V. Tsarev, *Zh. Obsch. Khim.*, **24**, 919 (1954); M.M. Janot, A. Care, and R. Goutharell, *Bull. Soc. Chim. France*, **1960**, 896; X.G. Monseur and E.L. Adriaens, *J. Pharm. Belg.*, **1960**, 279; H. Morimoto and H. Oshio, *Chem. Pharm. Bull.* (Tokyo), **11**, 1320 (1963).

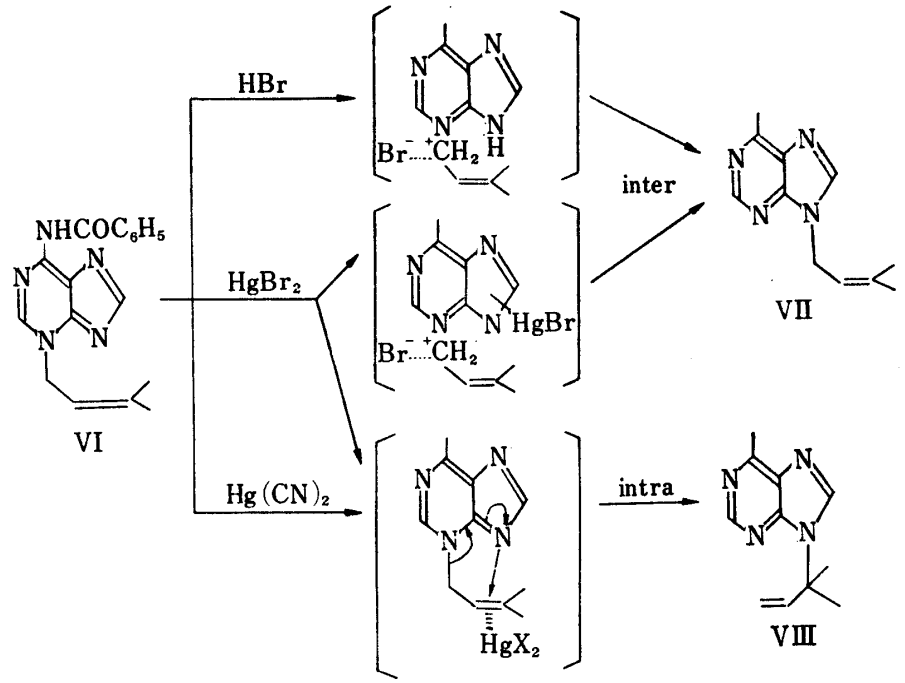


Chart 2

Table II-A Isotope Distribution in 3-Methyl-2-butenyl Migration (Relative Specific Radioactivity)

Catalyst	N⁶-Benzoyladenine (I[a])		Substituted N⁶-benzoyladenine		
	Before migr.	After migr.	9-(3-Me-2-butenyl) (VII[a])	9-(1,1-di-Me-2-propenyl) (VIII[a])	3-(3-Me-2-butenyl (VI[a])
HBr	100	49	51	—	—
$HgBr_2$(a)	100	54	65	23	—
$HgBr_2$(b)	100	76	86	10	11

Table II-B Yield of Products in 3-Methyl-2-butenyl Migration

Catalyst	Conditions	Substituted N⁶-benzoyladenine		
		(VII[a])	(VIII[a])	(VI[a])
HBr	110°, 24 hr	37	0	0
$HgBr_2$(a)	120°, 32 hr	29	11	10
$HgBr_2$(b)	95°, 17 hr+120°, 4 hr	8	39	10
$Hg(CN)_2$	140°, 26 hr	0	43*	21

a) containing the debenzoylated derivative

peratures (below 90°), gave only VIII. If the migration proceeded merely by intramolecular mechanism the 9-substituted derivatives would not labeled and, if the reaction proceeded only by intermolecular mechanism, the 9-substituted isomers and the recovered N⁶-benzoyladenine would have the same spicific activity. The present result indicated that the migration proceeded both by an intramolecular rearrangement to give the 9-(1,1-dimethyl-2-propenyl) isomer of low specific activity and by an intermolecular mechanism to give the 9-(3-methyl-2-butenyl) derivative of high specific activity. The labeled 3-(3-methyl-2-butenyl) derivative

was assumed to be regenerated from the intermolecularly released 3-methyl-2-butenyl group and N^6-benzoyladenine moiety.

The reason for the unusual intramolecular rearrangement was attributed to a certain interaction between mercuric bromide and the π-electrons in the allylic systems. To gain more information on this interaction, the migration was examined in the presence of mercuric cyanide which was considered to be less effective as an acid but still having affinity for the π-electrons of the allyl group. When VI was heated with mercuric cyanide at 140° for 26 hr in DMF, only VIII was formed. It is assumed, therefore, that the formation of a certain complex of allylic π-electrons with mercuric compounds initiated the nucleophilic attack of N-9 at the 3-postion of N-3-(3-methyl-2-butenyl) group, and subsequently the C–N bond at N-3 is cleaved, accompanied by the isomerization of the allyl group. On the contrary, attack of mercuric compound at the nitrogen in purine ring is assumed to cause the direct C–N bond cleavage at N-3 of VI. The difference in mode of interaction of these catalysts with VI was also observed in the NMR spectra.[11]

The isotope distribution in 3-methyl-2-butenyl migration and the yield of migration products are summarized in Table II–A and B. From these results, the mechanisms of the migrations can be illustrated in Chart 2. Pyrotriacanthine[10] was reported to be formed when triacanthine hydrochloride was heated in the absence of any solvent. The mechanism of this reaction seems to differ from the present migration reaction.

Experimental[12]

Benzylation of N^6-Benzoyladenine (I)——a) A mixture of I (4.5 g) and benzyl bromide (9.0 g) in DMF (5 ml) was heated at 100° for 3 hr and then evaporated to dryness *in vacuo*. Crystallization of the residue from EtOH gave II-HBr (1.42 g, 18.4%), mp 250°. UV $\lambda_{max}^{H_2O}$ mμ: pH 1, 300; pH 7, 301; pH 13, 330. *Anal.* Calcd. for $C_{19}H_{15}N_5O \cdot HBr$: C, 55.61; H, 3.66; N, 17.07. Found: C, 55.70; H, 3.95; N, 17.10. The mother liquor was treated with NH_4OH and evaporated to dryness, and the residue was crystallized from EtOH to give II (1.60 g, 25.9%), mp 195—196°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 300 (29500); pH 7, 301 (17400); pH 13, 330 (18600). *Anal.* Calcd. for $C_{19}H_{15}ON_5$: C, 69.28; H, 4.59; N, 21.27. Found: C, 69.18; H, 4.53; N, 21.13. The mother liquor was evaporated to dryness and the residue was chromatographed on a silica gel column (80 g). The first fraction eluted with $CHCl_3$–MeOH (99.5:0.5) on recrystallization from benzene gave III (0.91 g, 14.7%), mp 159—161°. UV $\lambda_{max}^{H_2O}$ mμ: pH 1, 290; pH 7, 281; pH 13, 299. *Anal.* Calcd. for $C_{19}H_{15}ON_5$: C, 69.28; H, 4.59; N, 21.27. Found: C, 69.29; H, 4.78; N, 21.40. The second fraction eluted with the same solvent on recrystallization from EtOH gave 3,7-dibenzyl-N^6-benzoyladenine (0.41 g, 5.2%), mp 218—220°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 302 (18500); pH 7, 302 (16500). *Anal.* Calcd. for $C_{26}H_{21}ON_5$: C, 74.44; H, 5.05; N, 16.70. Found: C, 74.39; H, 5.16; N, 16.61. The third fraction eluted with the same solvent on recrystallization from EtOH gave II (0.16 g, 2.5%). The fraction eluted with $CHCl_3$–MeOH (99:1) on recrystallization from EtOH gave 9-benzyladenine (0.20 g, 4.7%), mp 233°. UV $\lambda_{max}^{H_2O}$ mμ: pH 1, 259; pH 7, 261; pH 13, 261. *Anal.* Calcd. for $C_{12}H_{11}N_5$: C, 63.98; H, 4.92; N, 31.09. Found: C, 63.61; H, 4.93; N, 30.78. The fraction eluted with $CHCl_3$–MeOH (98:2) on recrystallization from EtOH gave 3-benzyladenine (0.10 g, 2.3%), mp 275—277°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 274.5 (16900); pH 7, 272 (12700); pH 12.8, 272 (12500). *Anal.* Calcd. for $C_{12}H_{11}N_5$: C, 63.98; H, 4.92; N, 31.09. Found: C, 63.58; H, 5.06; N, 31.12.

b) A mixture of I (1.25 g) and benzyl bromide (0.89 g) in DMF (15 ml) was heated under reflux for 3 hr, treated with NH_4OH and then evaporated to dryness. Chromatography of the residue on silica gel gave III (0.97 g, 45.6%), II (0.065 g, 3.0%) and 9-benzyladenine (0.053 g, 3.6%). Each of the products was identical in all respects to the compound prepared by method (a).

Benzylation of N^6-Acetyladenine——a) A mixture of N^6-acetyladenine (7.1 g) and benzyl bromide (20.5 g) in DMF (200 ml) was heated at 60—65° for 20 hr and evaporated to dryness *in vacuo*. The residue was treated with NH_4OH and extracted with $CHCl_3$. The extract was washed with H_2O, dried over Na_2SO_4 and allowed to stand at 0°. The crystals of 3-benzyl-N^6-acetyladenine appeared were collected by filtration, washed with $CHCl_3$ and dried *in vacuo* (4.24 g, 39.8%), mp 214°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 290 (19600); pH 7, 223 (20800) and 288.5 (14900); pH 13, 235 (11500) and 318 (17700). *Anal.* Calcd. for $C_{14}H_{13}ON_5$: C, 62.91; H, 4.90; N, 26.20. Found: C, 62.81; H, 4.92; N, 26.35. The mother liquor was evaporated to dryness

11) The spectra is described in ref. 1.

12) Melting points are not corrected. NMR spectra were measured on a Varian A60 Spectrometer.

and the residue was chromatographed on silica gel (200 g). The fraction eluted with $CHCl_3$–MeOH (99:1) on recrystallization from benzene gave 9-benzyl-N^6-acetyladenine (0.52 g, 4.9%), mp 166—168°. UV $\lambda_{max}^{H_2O}$ mμ: pH 1, 282; pH 7, 274; pH 13, 289. The fraction eluted with $CHCl_3$–MeOH (97.5:2.5) and $CHCl_3$–MeOH (95:5) gave 9-benzyl-(2.19 g, 24.3%) and 3-benzyladenine (1.38 g, 15.3%) respectively. These deacetylated compounds were considered to be formed during the chromatography.

b) A mixture of N^6-acetyladenine (5.0 g) and benzyl bromide (14.0 g) in DMF (150 ml) was heated at 110° for 140 hr and evaporated to dryness. Similar treatment of the residue as described above gave 9-benzyl-N^6-acetyladenine (3.23 g, 43.2%) and 9-benzyladenine (0.45 g, 7.1%) which were identical in all respects with the products obtained by method (a).

Allylation of I——A mixture of I (2.39 g) and allyl bromide (1.82 g) in DMA (5 ml) was heated at 100° for 3 hr and evaporated to dryness *in vacuo*. The residue was treated with NH_4OH and extracted with $CHCl_3$. The $CHCl_3$ solution was washed with H_2O, dried over Na_2SO_4 and evaporated to dryness. Chromatography of the residue on silica gel gave 9-allyl-N^6-benzoyladenine (0.6 g, 16.6%) and 3-allyl-N^6-benzoyladenine (1.25 g, 48.6%). 9-Allyl-N^6-benzoyladenine: mp 134—135°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 289 (22800); pH 7, 282 (17900); pH 13, 300 (11200). *Anal.* Calcd. for $C_{15}H_{13}ON_5$: C, 64.50; H, 4.69; N, 25.08. Found: C, 64.74; H, 4.59; N, 24.99. 3-Allyl-N^6-benzoyladenine: mp 184°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 299.5 (32800); pH 7, 240 (19000) and 300.5 (20700); pH 13, 330 (21400).

3-Methyl-2-butenylation of I——a) A mixture of I (956 mg) and 3-methyl-2-butenyl bromide (894 mg) in DMA (29 ml) was heated at 80° for 2 hr and evaporated to dryness. When the EtOH solution of the residue was kept at 0° for 15 hr the crystals of 3-(3-methyl-2-butenyl)-N^6-benzoyladenine. HBr (VI-HBr) were appeared (276 mg, 17.8%), mp 181°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 299 (29400); pH 7, 240 (18300) and 300 (19300); pH 13, 329 (18000). *Anal.* Calcd. for $C_{17}H_{17}ON_5 \cdot HBr$: C, 52.57; H, 4.38; N, 18.04; Br, 20.89. Found: C, 52.20; H, 4.75; N, 18.35; Br, 20.61. The filtrate and the mother liquor from recrystallization were treated with NH_4OH and evaporated to dryness. The residue was extracted with $CHCl_3$ and the $CHCl_3$ solution was washed with H_2O, dried over Na_2SO_4, concentrated and chromatographed on silica gel (16 g). The fraction eluted with $CHCl_3$–MeOH (99.5:0.5) on recrystallization from benzene gave 9-(3-methyl-2-butenyl)-N^6-benzoyladenine (VII) (170 mg, 13.8%), mp 125—127°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 250 (11000) and 288 (25800); pH 7, 250 (11000, sh) and 280 (20200); pH 13, 300 (12900). *Anal.* Calcd. for $C_{17}H_{17}ON_5$: C, 66.43; H, 5.58; N, 22.79. Found: C, 66.22; H, 5.50; N, 22.46. The fraction eluted with $CHCl_3$–MeOH (99:1) on recrystallization from EtOH–benzene gave VI (200 mg, 16.3%), mp 167—168°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 299 (32800); pH 7, 239 (19800) and 301 (21400); pH 13, 328 (20800). *Anal.* Found: C, 66.64; H, 5.80; N, 22.56. The fraction eluted with $CHCl_3$–MeOH (97:3) on recrystallization from EtOH gave 9-(3-methyl-2-butenyl)adenine (80 mg, 9.8%), mp 163°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 260.5 (14500); pH 7, 262.5 (15000); pH 13, 262 (15150). *Anal.* Calcd. for $C_{10}H_{13}N_5$: C, 59.09; H, 6.45; N, 34.46. Found: C, 59.00; H, 6.58; N, 34.47.

b) A mixture of N^6-benzoyladenine (2.0 g) and 3-methyl-2-butenyl bromide (1.86 g) in DMA (60 ml) was heated at 90° for 57 hr and treated as described above to obtain VII (1.2 g, 46.7%).

Benzylation of Chloromercuri-N^6-benzoyladenine——A mixture of N^6-benzoyladenine-HgCl (1.9 g) and benzyl bromide (0.9 g) in DMF was heated under reflux for 3 hr and evaporated to dryness *in vacuo*. The residue was extracted with $CHCl_3$ and the $CHCl_3$ solution was washed with 30% KI and H_2O, dried over Na_2SO_4 and evaporated to dryness. The residue (1.1 g) was chromatographed on silica gel (20 g). The fraction eluted with $CHCl_3$ on recrystallization from benzene gave III (0.29 g, 22%). The fraction eluted with $CHCl_3$–MeOH (99:1) on recrystallization from EtOH gave II (0.26 g, 19.7%) and 3-benzyladenine (0.07 g, 7.7%).

NMR Measurement of the Reaction Mixture of Benzylation and Benzyl Migration——a) A 1:1 mixture of 0.05M solutions of I and benzyl bromide in DMF was heated at 110°. To each 9.6 ml of the reaction mixture was added 1N NH_4OH (0.8 ml) at 0° and the mixture was evaporated to dryness. The residue was dissolved in d_6-DMSO (0.4 ml) and measured by NMR spectrometer at 60M cps. The ratio of intensity of signal[11] at δ 5.56 (benzyl methylene of III) to that at 5.75 (benzyl methylene of II) changed as shown in Fig. 1.

b) 0.05M solution of II-HBr in DMF was heated at 110° and treated as described above. The ratio of the yield of III to II is shown in Fig. 1.

Benzyl Migration of II——a) A solution of II-HBr (1.0 g) in DMF (40 ml) was heated at 110° for 70 hr and evaporated to dryness. The residue was extracted with a mixture of $CHCl_3$ (30 ml) and 15% NH_4OH (10 ml), the $CHCl_3$ layer was washed with H_2O, dried over Na_2SO_4 and evaporated to dryness. The residue (0.8 g) was chromatographed on silica gel (35 g). The fraction eluted with $CHCl_3$ and $CHCl_3$–MeOH (99.5:0.5) on recrystallization from benzene gave 9-benzyl derivative, III (485 mg, 60%), mp 161°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 290 (21700); pH 7, 281 (18200); pH 13, 299 (13300). *Anal.* Calcd. for $C_{19}H_{15}ON_5$: C, 69.28; H, 4.59; N, 21.27. Found: C, 69.52; H, 4.74; N, 20.81. This compound was identical in all respects with the sample prepared by benzylation of I. The crystals of III (425 mg) was heated in 0.25N NaOMe/MeOH (10 ml) under reflux for 1 hr and evaporated to dryness. Crystallization of the residue from EtOH gave 9-benzyladenine (240 mg, 83%) which was identical with the authentic sample in mp, UV and IR spectra.

b) II-HCl (510 mg) was heated in DMA (20 ml) at 110° for 20 hr and at 140—150° for 20 hr and evaporated to dryness. The residue (373 mg) was treated as described above and chromatographed on silica gel (2.5 g). The fraction eluted with $CHCl_3$ on recrystallization from benzene gave III (100 mg, 21.7%). The fraction eluted with $CHCl_3$–MeOH (99.5:0.5) on recrystallization from EtOH gave 7-benzyl-N^6-benzoyladenine (11 mg, 2.4%), mp 220—228°, which was identical with the authentic sample[13] in mp, UV and IR spectra and *Rf* value of TLC. The fraction eluted with $CHCl_3$–MeOH (98:2) on recrystallization from EtOH gave 9-benzyladenine (40 mg, 12.7%).

Benzyl Migration of 3-Benzyl-N^6-acetyladenine——3-Benzyl-N^6-acetyladenine-HBr (800 mg) was heated at 110° for 72 hr in DMF (40 ml) and evaporated to dryness. The residue was treated as described above and chromatographed on silica gel (12 g). The first fraction eluted with $CHCl_3$–MeOH (99.8:0.2) gave 9-benzyl-N^6-acetyladenine (70 mg, 11%), mp 166—168°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 282 (19050); pH 7, 274 (16600); pH 13, 289 (11300). *Anal.* Calcd. for $C_{14}H_{13}ON_5$: C, 62.91; H, 4.90; N, 26.20. Found: C, 62.82; H, 4.67; N, 26.60. This compound was identified with the sample obtained by benzylation of N^6-acetyladenine. The second fraction on recrystallization from EtOH gave 9-benzyladenine (110 mg, 21%). 3-Benzyladenine (101 mg, 19.5%) was recovered from the fraction eluted with $CHCl_3$–MeOH (95:5).

Concentration effect on the Rate of Benzyl Migration——Each of solutions of II-HBr in DMF was heated at 100—110° for 40 hr, treated with NH_4OH, and then evaporated to dryness. The spots of III, II, and I on TLC were extracted with $CHCl_3$–EtOH. The quantity of III, II, or I in each reaction mixture was determined by measurement of the optical density at UV absorption maximum in the EtOH solution of each extract.

Concentration (M) of II-HBr	Quantity (%)		
	III	II	I
5×10^{-2}	28	16	19
5×10^{-3}	2	26	13
5×10^{-4}	<1	22	6

N^6-Benzoyladenine-8-^{14}C——A mixture of Adenine-8-^{14}C (0.5 mc/18 mg), cold adenine (1 g) and benzoic anhydride (3 g) was fused at 160° for 80 min and EtOH (4 ml) was added. After being cooled, the mixture was triturated with ether (20 ml) and the insoluble crystals were recrystallized from EtOH to give N^6-benzoyladenine-8-^{14}C (1.5 g, 85%), mp 239—240°.

Exchange of Benzyl and Allyl Groups during N-3 N-9 Migration——A mixture of II*-HBr (2.38×10^9 dpm/mole, 594 mg) and 3-allyl-N^6-benzoyladenine-HBr (IV-HBr) (520 mg) in DMF (70 ml) was heated at 90—110° for 61 hr. The reaction mixture was treated with NH_4OH at 0° and evaporated to dryness, and the residue was chromatographed on silica gel (20 g). The former fraction eluted with $CHCl_3$ on recrystallization from benzene-EtOH gave 9-benzyl-N^6-benzoyladenine-8-^{14}C (III*) (200 mg, 42%), mp 159—160°, which showed the radioactivity of 1.47×10^9 dpm/mole. The latter fraction on recrystallization from benzene–EtOH gave 9-allyl-N^6-benzoyladenine-8-^{14}C (V*) (36 mg, 9%), mp 133—135.5°, which showed the radioactivity of 1.44×10^9 dpm/mole. The small amounts of radioactive 3-benzyl-N^6-benzoyladenine and 3-allyl-N^6-benzoyladenine were recovered.

Benzyl Migration in the presence of labeled N^6-Benzoyladenine (I*)——A mixture of II-HBr (1.23 g) and I* (2.80×10^9 dpm/mole, 717 mg) in DMA (60 ml) was heated at 110° for 70 hr and treated with NH_4OH at 0°. The reaction mixture was evaporated to dryness and chromatographed on silica gel (18 g). The fraction eluted with $CHCl_3$–MeOH (99.8:0.2) on recrystallization from benzene–EtOH gave labeled 9-benzyl derivative (III*) (460 mg, 46.7%), mp 160°, which showed activity of 1.46×10^9 dpm/mole. The fraction eluted with $CHCl_3$–MeOH (99:1) on recrystallization from EtOH gave 7-benzyl-N^6-benzoyladenine-8-^{14}C (46 mg, 4.7%), mp 220—230°, which showed radioactivity of 1.45×10^9 dpm/mole. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 281 (13100) and 330 (2940); pH 7, 280.5 (11900) and 331 (3580); pH 13, 299 (11500). *Anal.* Calcd. for $C_{19}H_{15}ON_5$: C, 69.28; H, 4.59; N, 21.27. Found: C, 69.00; H, 4.74; N, 21.41.

An Attempt to Migrate 9-Benzyl-N^6-benzoyladenine——A mixture of III (329 mg), HBr/AcOH (0.25 ml) and N^6-benzoyladenine-8-^{14}C (I*) (6.35×10^9 dpm/mole, 239 mg) in DMF (20 ml) was heated at 110° for 70 hr. The reaction mixture was treated with NH_4OH and evaporated to dryness, and the residue was chromatographed on silica gel. The fraction eluted with $CHCl_3$ on recrystallization from benzene–EtOH gave 9-benzyl-N^6-benzoyladenine (160 mg, 48.5%), showing activity of 5.96×10^7 dpm/mole.

Allyl Migration of 3-Allyl-N^6-benzoyladenine (IV) (by HBr)——IV-HBr (mp 216—218°, 360 mg) was heated in DMA (4 ml) at 110° for 30 hr. The reaction mixture was treated with NH_4OH at 0°, evaporated

13) It was kindly supplied by Dr. J.A. Montgomery.

to dryness and chromatographed on silica gel. The fraction eluted with $CHCl_3$ on recrystallization from benzene gave 9-Allyl-N^6-benzoyladenine (V) (53 mg, 19%) which was identical with the sample prepared by allylation of I in UV spectra and mp.

Allyl Migration of IV (by $HgBr_2$ or $HgCl_2$)——A mixture of IV (100 mg) and $HgBr_2$ (129 mg) in DMF (1 ml) was heated at 100° for 24 hr and evaporated to dryness. The residue was extracted with $CHCl_3$ and the solution was washed with 30% KI and H_2O, dried (Na_2SO_4) and chromatographed on silica gel. The fraction eluted with $CHCl_3$ on recrystallization from benzene gave V (31 mg, 31%), mp 134°. When a mixture of IV (93 mg) and $HgCl_2$ (90 mg) in DMF (1 ml) was heated at 100° for 30 hr and treated as described above, V was obtained in 42% yield.

3-Methyl-2-butenyl Migration of N^6-Benzoyltriacanthine (VI) (by HBr)——VI-HBr (2.8 g) was heated in DMF (20 ml) at 95—100° for 40 hr. The reaction mixture was treated with NH_4OH and evaporated to dryness. The $CHCl_3$ extract of the residue was washed with H_2O, dried, concentrated and chromatographed on silica gel (15 g). The fraction eluted with $CHCl_3$ on recrystallization from benzene gave 9-(3-methyl-2-butenyl)-N^6-benzoyladenine (VII) (390 mg, 17.6%), mp 125—127°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 250 (10400) and 288 (24500); pH 7, 250 (10400, sh) and 281 (20100); pH 13, 299.5 (12500). *Anal.* Calcd. for $C_{17}H_{17}ON_5$: C, 66.43; H, 5.58; N, 22.79. Found: C, 66.19; H, 5.60; N, 22.55. The fraction eluted with $CHCl_3$–MeOH (99:1) gave 9-(3-methyl-2-butenyl)adenine (35 mg, 2.4%), mp 163—166°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 260 (14500); pH 7, 262 (14900); pH 13, 262 (15000). *Anal.* Calcd. for $C_{10}H_{13}N_5$: C, 59.09; H, 6.45; N, 34.46. Found: C, 58.85; H, 6.39; N, 34.32.

Migration of N^6-Benzoyltriacanthine (VI) (by $HgBr_2$)——A mixture of VI (212 mg) and $HgBr_2$ (250 mg) in DMF (1.5 ml) was heated at 110° for 30 hr and evaporated to dryness. The residue was extracted with $CHCl_3$ and the $CHCl_3$ solution was washed with 30% KI and H_2O, dried, and chromatographed on silica gel (5 g). The former fraction eluted with $CHCl_3$ on recrystallization from *n*-hexane–benzene gave 9-(1,1-dimethyl-2-propenyl)-N^6-benzoyladenine (VIII) (85 mg, 40%), mp 153—157°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 251 (10500) and 288.5 (24600); pH 7, 251 (10500, sh) and 282 (20000); pH 13, 299 (12900). *Anal.* Calcd. for $C_{17}H_{17}ON_5$: C, 66.43; H, 5.58; N, 22.79. Found: C, 66.14; H, 5.62; N, 22.65. The latter fraction eluted with $CHCl_3$ on recrystallization from benzene gave VII (6 mg, 2.8%) which was identical in all respects with the sample obtained from the migration reaction induced by HBr. The NMR spectra of these compounds are shown in Fig. 3. In the spectrum of VII, the triplet at 5.45 (1-proton) and the doublet at 4.86 ppm (2-protons) are characteristic of the $=CH-CH_2-$ group, and J-value for the two multiplets is 7 cps. The peaks at 1.82 (3-protons) and 1.72 ppm (3-protons) are due to $=C\langle^{CH_3}_{CH_3}$ group. Whereas in the spectrum of VIII, the quartet at 6.48 ppm (1-proton) and the doublets at 5.28 (1-proton) and 5.16 ppm (1-proton) are characteristic of vinyl protons, $^{H}\rangle C=C\langle^{H}_{H}$ and J-values are 11 and 17 cps. The peaks at 1.85 ppm (6-protons) due to the $\rangle C\langle^{CH_3}_{CH_3}$ group.

3-Methyl-2-butenyl Migration of VI in the Presence of Labeled N^6-Benzoyladenine (I*) (by HBr)——A mixture of VI-HBr (600 mg) and I* (6.19×10^9 dpm/mole) (369 mg) in DMF (6 ml) was heated at 110° for 24 hr, treated with NH_4OH and evaporated to dryness. The $CHCl_3$–extract of the residue was allowed to stand at room temperature for 18 hr. Recrystallization of the resulting precipitates from EtOH gave N^6-benzoyladenine-8-C^{14} (127 mg) revealing radioactivity of 3.06×10^9 dpm/mole. Chromatography of the mother liquor on silica gel (12 g) gave the labeled VII* (175 mg, 37%) mp 125—127°, which showed activity of 3.11×10^9 dpm/mole.

Migration of VI in the Presence of Labled N^6-Benzoyladenine (I*) (by $HgBr_2$)——a) A mixture of VI (154 mg), $HgBr_2$ (180 mg) and I* (3.08×10^9 dpm/mole, 120 mg) in DMF (2 ml) was heated at 95° for 17 hr and subsequently at 120° for 4 hr, and evaporated to dryness. The residue was extracted with $CHCl_3$, and the $CHCl_3$ solution was washed with 30% KI and H_2O, dried and chromatographed on silica gel (4 g). The fraction eluted with benzene–$CHCl_3$ (50:50) on recrystallization from *n*-hexane–benzene gave VIII* (60 mg, 39%), mp 158—160°, radioactivity: 3.1×10^8 dpm/mole. The fraction eluted with $CHCl_3$ gave VII* (12 mg, 8%), mp 125°, radioactivity: 2.66×10^9 dpm/mole. The fraction eluted with $CHCl_3$–MeOH (99:1) gave VI* (16 mg, 10%), mp 168°, radioactivity: 3.2×10^8 dpm/mole. The fraction eluted with $CHCl_3$–MeOH (98:2) gave N^6-benzoyladenine (87 mg, 73%), mp 239°, radioactivity: 2.33×10^9 dpm/mole.

b) A mixture of VI (500 mg), $HgBr_2$ (585 mg) and I* (6.2×10^9 dpm/mole, 389 mg) in DMF (5 ml) was heated at 120° for 32 hr and treated as described above. The yield and radioactivity of each product was as follows: VIII* (55 mg, 11%), 1.43×10^9 dpm/mole; VII* (145 mg, 29%), 4.04×10^9 dpm/mole; I* (270 mg), 3.33×10^9 dpm/mole.

Migration of VI by $Hg(CN)_2$——A mixture of VI (250 mg) and $Hg(CN)_2$ (205 mg) in DMA (4 ml) was heated at 140° for 26 hr. Metal Hg (12 mg) was filtered off and the filtrate was evaporated to dryness. The residue was extracted with $CHCl_3$ and the extract was treated as described above and chromatographed on silica gel (7.5 g). The fraction eluted with $CHCl_3$ gave VIII (23 mg, 9%) which was identical with the authentic sample in all respects. The fraction eluted with $CHCl_3$–MeOH (99:1) gave 9-(1,1-dimethyl-2-propenyl)adenine (56 mg, 34%), mp 185°. UV $\lambda_{max}^{H_2O}$ mμ (ε): pH 1, 260 (14400); pH 7, 261.5 (14750);

pH 13, 261.5 (14750). The fraction eluted with $CHCl_3$–MeOH (97:3—95:5) gave 3-(3-methyl-2-butenyl)-adenine (35 mg, 21%).

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