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Studies on Vitamin B_1 and Related Compounds. CIX.[1] A Novel Cleavage of Thiamin and Its Homologues by the Reaction with Aromatic Aldehydes[2]

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A novel cleavage reaction of thiamin to give 4-amino-2,5-dimethylpyrimidine (III) and 2-benzoyl-5-(2-hydroxyethyl)-4-methylthiazole (IV) has been discovered. The reaction involves the treatment of thiamin chloride hydrochloride (I) with two mole equivalents of triethylamine and an excess of benzaldehyde in methanol. The finding was further extended to the reactions of thiamin and its homologs with a variety of aromatic aldehydes to give several new 2-acylthiazoles. The pyrimidine moiety of thiamin was indispensable for these reactions. On the other hand, the reaction of thiamin with cinnamaldehyde gave 2,9a-dimethyl-9-(2-hydroxyethyl)-7-(3-methoxy-3-phenylpropionyl)-5,9,9a,-10-tetrahydro-7H-pyrimido[4,5-*d*]thiazolo[3,4-*a*]pyrimidine (XV).

The reaction of thiamin with acetaldehyde provides an excellent means for the synthesis of biochemically important α-hydroxyethylthiamin (HET).[4] Several other α-hydroxyalkylthiamins have also been synthesized by essentially the same method using aliphatic aldehydes.[4,5]

During the course of our investigations on the reactivity at the 2-position of the thiazole ring of thiamin towards aromatic aldehydes, we found that hitherto unknown degradation or rearrangement had occurred giving rise to unidentified products rather than the expected α-hydroxyalkylthiamins under certain conditions. This paper deals with the novel cleavage reaction of thiamin and its homologs, together with the characterization of the reaction products.

When thiamin chloride hydrochloride (I) was treated with two mole equivalents of triethylamine in methanol and the solution was refluxed with an excess of benzaldehyde, the expected α-hydroxybenzylthiamin (HBzT) (II) was detected in a trace quantity; instead, three compounds were obtained along with a considerable amount of benzoin.

One of them, which melted at 202°, was identified as 4-amino-2,5-dimethylpyrimidine (III) from the inspection of the nuclear magnetic resonance (NMR) spectrum, the elementary analysis and the direct comparison with an authentic sample.[6]

The second compound, mp 84°, $C_{13}H_{13}O_2NS$ (IV), was a neutral ketone. Reduction of the ketone with sodium borohydride, however, afforded a basic compound. The compound was identical with 2-(α-hydroxybenzyl)-5-(2-hydroxyethyl)-4-methylthiazole (V) prepared by the sulfite cleavage of HBzT.[7] This result indicates that IV is 2-benzoyl-5-(2-hydroxyethyl)-4-methylthiazole; the fact was further confirmed by the NMR and infrared (IR) spectra

1) Part CVIII: Y. Oka, E. Imamiya and K. Masuda, *Vitamins* (Kyoto), **39**, 324 (1969).
2) Part of this work was presented at the 24th Meeting of Pharmaceutical Society of Japan, Kyoto, April 1967.
3) Location: *Juso, Higashiyodogawa-ku, Osaka.*
4) C.S. Miller, J.M. Sprague and L.O. Krampitz, *Ann. N.Y. Acad. Sci.*, **98**, 401 (1962).
5) Y. Oka and S. Yurugi, *Vitamins* (Kyoto), **32**, 570 (1965).
6) R.R. Williams, A.E. Ruehle and J. Finkelstein, *J. Am. Chem. Soc.*, **59**, 526 (1937).
7) Y. Oka, E. Imamiya and H. Hirano, *Chem. Pharm. Bull.* (Tokyo), **15**, 448 (1967).

(Fig. 1 and 2) and the formation of the 2,4-dinitrophenylhydrazone. The extremely weak basisity of IV might be explained in terms of a strong inductive effect of the benzoyl group at the 2-position.

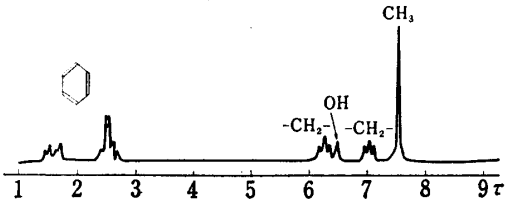

Fig. 1. NMR Spectrum of IV in $CDCl_3$ at 60 Mc

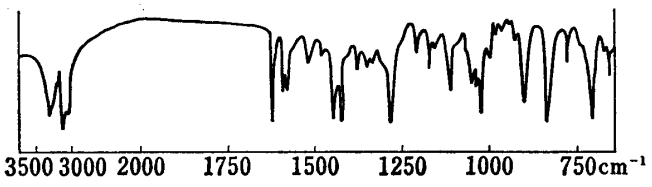

Fig. 2. IR Spectrum of IV (Nujol)

The third compound (VI) having an empirical formula $C_{19}H_{22}O_2N_4S$ was isolated in a minute quantity, and the structure of which will be discussed in the following paper of this series.

These results demonstrate that the reaction of thiamin with benzaldehyde under the aforementioned conditions proceeds mainly to give III and IV. Since such a cleavage reaction does not occur when thiamin chloride hydrochloride or thiamin chloride is allowed to react with benzaldehyde in the absence of triethylamine, it is reasonable to assume that benzaldehyde first reacts at the 2-position of the thiazole ring to give HBzT followed by further rearrangements of the reaction product into III and IV.

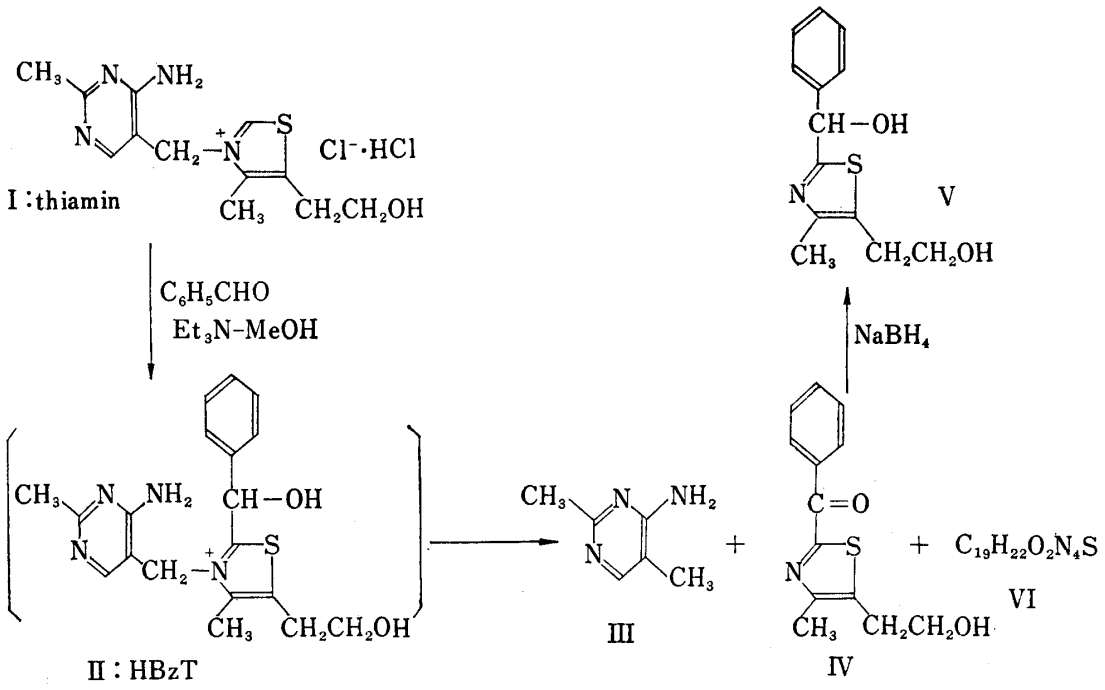

Chart 1

In order to confirm this assumption we examined the behavior of HBzT in alkaline solutions. It is already known that in alkaline media the thiazolium ring of HET is cleaved to give a thiol–form in a similar manner as in thiamin. When HBzT chloride hydrochloride[4] was neutralized with sodium hydroxide, the compound was readily cleaved into benzaldehyde and thiol–form thiamin and only a small amount of HBzT was recovered on acidification. On the other hand, when HBzT chloride hydrochloride was refluxed with two mole equivalents of triethylamine in methanol, III, IV and VI were obtained along with thiamin and benzoin. These observations indicate that the chemical properties of HBzT in alkaline solutions are

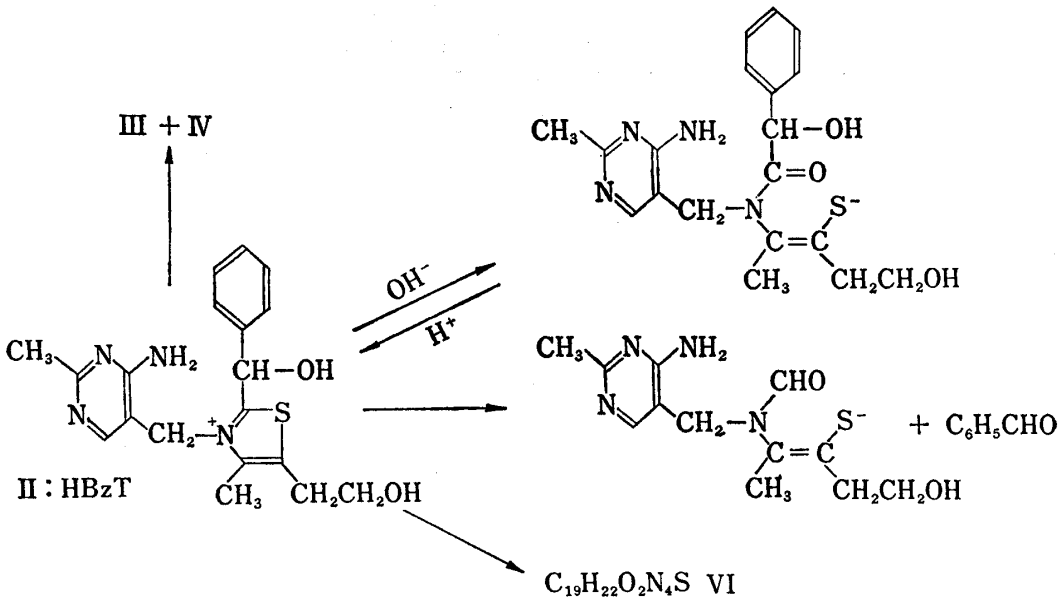

Chart 2

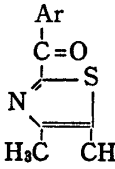

TABLE I.

No.	Ar	R	Formula	mp (°C)	Yield (%)
1	C_6H_5	CH_2OH	$C_{13}H_{13}O_2NS$	84	40
2	p-$CH_3C_6H_4$	CH_2OH	$C_{14}H_{15}O_2NS$	76—78	25
3	p-ClC_6H_4	CH_2OH	$C_{13}H_{12}O_2NSCl$	86	36
4	p-BrC_6H_4	CH_2OH	$C_{13}H_{12}O_2NSBr$	99—100	45
5	p-$CH_3OC_6H_4$	CH_2OH	$C_{14}H_{15}O_3NS$	67—68	20
6	2-Furyl	CH_2OH	$C_{11}H_{11}O_3NS$	87—89	20
7	C_6H_5	$CH_2OCOC_6H_5$	$C_{20}H_{15}O_3NS$	107	21
8	C_6H_5	H	$C_{12}H_{11}ONS$[a]	88—89	30
9	p-ClC_6H_4	H	$C_{12}H_{10}ONSCl$	97—99	54
10	p-BrC_6H_4	H	$C_{12}H_{10}ONSBr$	90—94	35

No.	UV λ_{max}^{EtOH} mμ (ε)	Analysis (%) Calcd. C	Calcd. H	Calcd. N	Found C	Found H	Found N
1	274 (6780), 326 (10800)	63.13	5.30	5.67	63.13	5.23	5.41
2	285 (9400), 330 (14000)	64.34	5.78	5.36	64.73	5.61	5.07
3	277 (9500), 334 (13100)	55.41	4.29	4.97	55.27	4.27	4.81
4	280 (10300), 332 (13100)	47.86	3.71	4.29	47.88	3.48	4.13
5	232 (7800), 332 (18900)	60.62	5.45	5.05	60.61	5.45	4.91
6	242 (3500), 341 (14900)	55.68	4.67	5.90	55.01	4.73	5.67
7	231 (17500), 271 (8100), 326 (11100)	68.16	5.15	3.97	68.45	4.87	3.94
8	267 (7200), 328 (10700)	66.33	5.10	6.45	66.41	4.89	6.53
9		57.25	4.00	5.56	56.84	3.96	5.62
10		48.65	3.40	4.73	48.96	3.26	4.66

a) lit. 88—89° [8]

8) M. Erne and H. Erlenmeyer, *Helv. Chim. Acta*, 31, 652 (1948).

much more complicated compared with those of HET and thiamin as is shown in Chart 2, and that III, IV and VI would have probably arisen from HBzT.

Since the preparation of 2-acylthiazoles by the known methods was not so easy, the present reaction appears to be useful as a general method for the synthesis of 2-acylthiazoles. In order to see if the reaction can be applicable to reactions between other thiazolium compounds and aldehydes, we have explored the scope and limitation of the reaction. First of all, we investigated the reaction of thiamin with other alkyl aldehydes. However, all these alkyl aldehydes gave nothing but the corresponding α-hydroxyalkylthiamin by the reaction with thiamin. On the other hand, it was found that a variety of 2-acyl-5-(2-hydroxyethyl)-4-methylthiazoles were obtained by the reaction with various aromatic aldehydes; *e.g.*, *p*-tolualdehyde, *p*-chlorobenzaldehyde, anisaldehyde and furfural. This reaction also proceeded in the same way when thiamin homologues such as VII and VIII, in which the substituents on the thiazole are modified, were reacted with aromatic aldehydes. Thus, several new 2-acyl derivatives of thiazoles could be prepared by this reaction. The results are summarized in Table I.

It should be pointed out, however, that the reaction is markedly affected by modification of the pyrimidine moiety of the compound. When 3,4-dimethyl-5-(2-hydroxyethyl)thiazolium iodide (IX), 3-benzyl-5-(2-hydroxyethyl)-4-methylthiazolium chloride (X) and 5-(2-hydroxyethyl)-4-methyl-3-(4-nitrobenzyl)thiazolium chloride (XI), in which the pyrimidinylmethyl moiety was replaced by one of methyl, benzyl and 4-nitrobenzyl groups, were allowed to react with benzaldehyde, all the reaction failed to give IV; instead 3,4-dimethyl-2-(α-hydroxybenzyl)-5-(2-hydroxyethyl)thiazolium iodide (XII) was obtained only in the case of IX. The compound (XII) thus obtained showed a complete identity with an authentic sample which was prepared by methylation of V with methyl iodide. Furthermore it was found that oxythiamin (XIII), in which the 4-amino group in the pyrimidine moiety of thiamin is replaced by the hy-

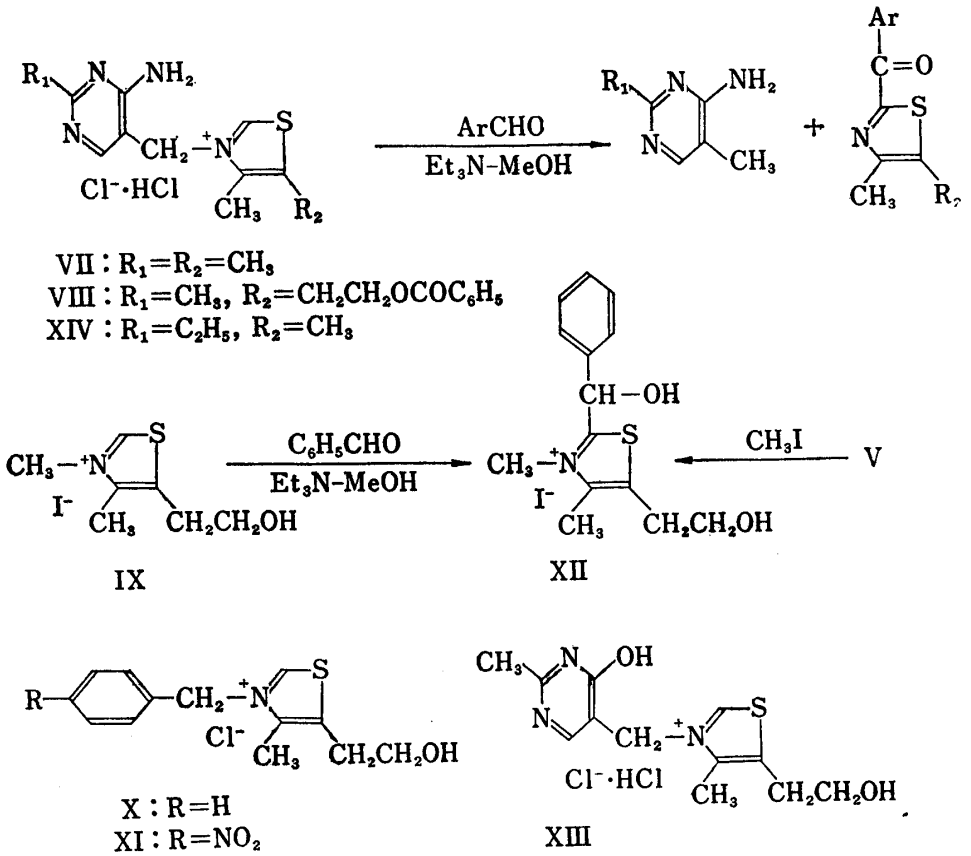

Chart 3

droxy group, failed to undergo this reaction, while 2-ethyl homolog (XIV) afforded 2-benzoyl-4,5-dimethylthiazole.

In view of these results it may be concluded that the 4-amino group in the pyrimidine ring is indispensable for this cleavage reaction.

In the course of these investigations an anomalous finding was made when cinnamaldehyde, a vinylog of benzaldehyde, was allowed to react with thiamin expecting to obtain a 2-cinnamoyl thiazole derivative. From the empirical formula, $C_{22}H_{28}O_3N_4S$, the reaction product was assumed to be an addition product of one mole each of thiamin, cinnamaldehyde and methanol. The NMR spectrum of this compound (Fig. 3) showed that the methyl signal at the 4-position of the thiazole moiety had shifted to 8.75 τ and that the –NH– signal was observed at 2.48 τ. These data together with the carbonyl band at 1730 cm^{-1} in the IR spectrum strongly suggest that the compound is a pseudo-2-acyldihydrothiamin derivative.[7,9] In addition, the absence of a vinyl signal in the NMR indicated that the addition of methanol had occurred at the double bond of the cinnamoyl group. These evidence led us to assign the structure XV (Chart 4) to the compound. It remains to be seen if the addition of methanol takes place before or after the reaction of aldehydes with thiamin.

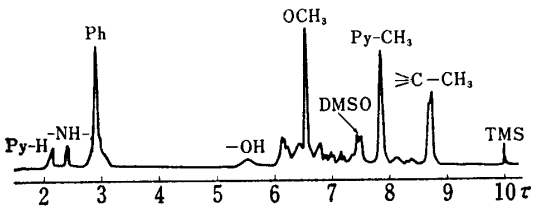

Fig. 3. NMR Spectrum in DMSO-d_6 at 100 Mc

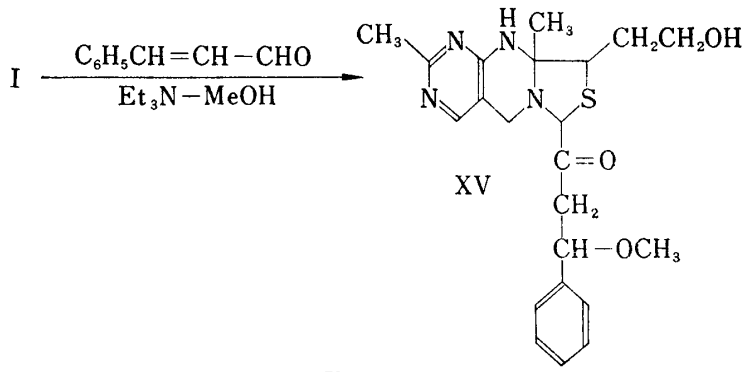

Chart 4

Experimental

Reaction of Thiamin with Benzaldehyde——To a suspension of thiamin Cl. HCl (I) (16.8 g) in MeOH (125 ml) was added 10.5 g of triethylamine and the mixture was stirred until I had completely dissolved. To this solution was added 10.5 g of benzaldehyde and the mixture was refluxed for 5 hr. After removal of the solvent *in vacuo*, the residue was treated with 100 ml of AcOEt and extracted with 10% HCl. From the AcOEt layer was obtained some amounts of benzoin in crystalline form, which was removed by filtration. The filtrate was washed with water, dried over Na_2SO_4 and evaporated to dryness. The residue was dissolved in 20 ml of benzene. On standing an additional amount of benzoin crystallized out, which was filtered and the filtrate was evaporated to give 10 g of syrup. This was then chromatographed on silica gel (Mallincrodt, 100 mesh: 200 g). After benzoin had been eluted with a solvent system of 550 ml of benzene–acetone (4:1), elution with 700 ml of benzene–acetone (4:1) and evaporation of the solvent afforded 6.2 g of colorless syrup which crystallized on standing in benzene–petroleum ether. Recrystallization from benzene–petroleum (1:1) gave 4.95 g (40%) of 2-benzoyl-5-(2-hydroxyethyl)-4-methylthiazole (IV) as colorless prisms, mp 84°. *Anal.* Calcd. for $C_{13}H_{13}O_2NS$: C, 63.13; H, 5.30; N, 5.67. Found: C, 63.13; H, 5.23; N, 5.41. UV λ_{max}^{EtOH} mμ (ε): 263 (6780), 326 (10800).

2,4-Dinitrophenylhydrazone of IV was prepared according to the method by Parrick and Rasburn;[10] mp 236° (decomp.). *Anal.* Calcd. for $C_{19}H_{17}O_5N_5S$: C, 53.39; H, 4.01; N, 16.59. Found: C, 53.10; H, 3.97; N, 16.45.

On the other hand, the above HCl solution was neutralized with $NaHCO_3$ and extracted with AcOEt. The AcOEt phase was dried over Na_2SO_4 and evaporated to dryness. Then the residue was dissolved in 10 ml of acetone and set aside overnight to afford VI as needles, which were recrystallized from acetone. Yield 0.7 g, mp 171—172° (decomp.). *Anal.* Calcd. for $C_{19}H_{22}O_2N_4S$: C, 61.59; H, 5.99; O, 8.64; N, 15.12; S, 8.66. Found: C, 61.38; H, 5.95; O, 9.16; N, 15.40; S, 8.96.

9) Y. Oka, K. Yoshioka and H. Hirano, *Chem. Pharm. Bull.* (Tokyo), **15**, 119 (1967).
10) J. Parrick and J.W. Rasburn, *Can. J. Chem.*, **43**, 3453 (1965).

The aqueous layer was acidified again with HCl and evaporated to dryness *in vacuo*. The residue was extracted with hot EtOH (40 ml), and the extract was evaporated to dryness. To the residue was added 50 ml of acetone and an undissolved crystalline mixture of III·HCl and triethylamine hydrochloride was collected by filtration. Washing of the mixture with $CHCl_3$ several times removed triethylamine hydrochloride and there remained 3.5 g (44%) of III·HCl, which was dissolved in 5 ml of water and neutralized with $NaHCO_3$. The resulting precipitate was collected and recrystallized from acetone to give IV as colorless prisms, mp 202° (lit.[6] mp 201—202°). *Anal.* Calcd. for C_6H_9N: C, 58.51; H, 7.36; N, 34.12. Found: C, 58.31; H, 7.45; N, 33.89. NMR (in CF_3COOH) τ: 7.67 (CH_3), 7.17 (CH_3), 2.04 (CH).

2-(α-Hydroxybenzyl)-5-(2-hydroxyethyl)-4-methylthiazole (V)——To a stirred solution of IV (1.0 g) in MeOH (30 ml) was added 0.2 g of $NaBH_4$ dissolved in MeOH (5 ml). The stirring was continued at room temperature for 40 min, and to the solution was added a few drops of AcOH to decompose excess $NaBH_4$. After evaporation *in vacuo*, the residue was made alkaline with aq. NaOH and extracted with AcOEt. The extract was dried over Na_2SO_4 and evaporated. The residue was set aside in ether to afford crystalline solid which was filtered and recrystallized from AcOEt–ether to give V as colorless prisms, mp 101—102°. This substance showed no depression of melting point on admixture with an authentic sample;[7] IR spectrum of V also showed complete identity with that of an authentic specimen.

Reactions of HBzT with Bases——i) To a stirred solution of II·Cl·HCl (4.6 g) in water (40 ml) was added 12 ml of aq. NaOH to effect an yellow clear solution. Within a minute there appeared an oily substance which was taken in AcOEt and evaporated to give 250 mg of oil. Treatment of the oil with 2,4-dinitrophenylhydrazine in DMF gave orange-yellow crystals, mp 239—240°, which showed no depression of melting point on admixture with an authentic sample of benzaldehyde 2,4-dinitrophenylhydrazone. The aqueous layer was adjusted to pH 1 with HCl and concentrated *in vacuo* to dryness. The residue was extracted with 15 ml of hot MeOH and the extract was diluted with 15 ml of acetone. On cooling 0.8 g (24%) of thiamin chloride HCl (I) precipitated which after recrystallization showed complete identity with an authentic sample of I in every respect. The mother liquor of I was evaporated to dryness and the residue was recrystallized from MeOH–acetone to recover 0.2 g (4%) of the starting material, II·Cl·HCl.

ii) A solution of II·Cl·HCl[7] (2.3 g) and triethylamine (1.1 g) in 13 ml of MeOH was refluxed for 5 hr and evaporated to dryness. The residue was acidified with 10% HCl and extracted with ether. After drying over Na_2SO_4, the solvent was removed by evaporation to afford 0.6 g of yellow oil which crystallized gradually. Washing with a small portion of ether and recrystallization from benzene–petroleum ether gave colorless prisms of IV (0.2 g), mp 84°, which showed no depression of melting point on admixture with an authentic sample of IV. The aqueous layer was neutralized with $NaHCO_3$ and extracted with AcOEt. After evaporation of AcOEt, the residue was chromatographed on silica gel (45 g) and eluted with a solvent system of benzene–acetone–MeOH (3:3:1) to afford 60 mg of VI. Evaporation of the remaining aqueous phase gave thiamin, which was isolated as the monothiocyanate,[9] and III (0.1 g).

General Procedure for the Syntheses of 2-Acylthiazoles by the Reaction of Thiamin or Its Homologs with Aromatic Aldehydes——A variety of 2-acylthiazoles listed in Table I were prepared by a similar procedure to the reaction of thiamin and benzaldehyde. The procedure is as follows: To a solution of thiamin or its homologs (0.1 mole) and triethylamine (20.2 g, 0.2 mole) in MeOH (250 ml) was added 0.2 mole of an aromatic aldehyde and the mixture was refluxed for 5 hr. After MeOH had been evaporated *in vacuo*, the residue was acidified with 10% HCl and extracted with AcOEt. The AcOEt layer was dried over Na_2SO_4 and evaporated to dryness. The residue was dissolved in a small portion of benzene. If necessary benzoin and its homologs were removed by filtration. The benzene solution was chromatographed on silica gel and eluted with a mixture of benzene, acetone and methanol to afford 2-acylthiazole. Physical properties and yields of the thiazoles are listed in Table I.

3,4-Dimethyl-2-(α-hydroxybenzyl)-5-(2-hydroxyethyl)thiazolium Iodide (XII)——i) To a solution of 3,4-dimethyl-5-(2-hydroxyethyl)thiazolium iodide (IX) (7 g) in 40 ml of MeOH was added triethylamine (2.5 g) and benzaldehyde (20 g), and the solution was refluxed for 5 hr. After evaporation of the solvent, the residue was treated with 30 ml of water and the mixture was extracted with 30 ml of AcOEt. The aqueous layer was concentrated to 10 ml and extracted twice with each 10 ml of *n*-butanol. The combined butanol solution was dried over Na_2SO_4 and evaporated to dryness under reduced pressure. The residue was then chromatographed on silica gel (80 g). The elution with a solvent system of benzene–acetone–methanol (2:2:1) afforded 0.95 g (10%) of XII. Recrystallization from EtOH–acetone gave colorless prisms, mp 111—113°. *Anal.* Calcd. for $C_{14}H_{18}O_2NS$: C, 42.97; H, 4.63; N, 3.58. Found: C, 42.96; H, 4.61; N, 3.51. UV λ_{max}^{EtOH} mμ (ε): 215 (20400), 266 (7660). NMR (in D_2O) τ: 7.53 (CH_3), 6.86 (triplet, CH_2), 6.24 (CH_3), 6.12 (triplet, CH_2), 3.56 (CH), 2.48 (C_6H_5).

ii) A solution of V (0.4 g) and methyl iodide (4 ml) in 5 ml of THF was refluxed for 5 hr, and excess of methyl iodide and THF were removed by evaporation to afford an yellow crystalline mass, which was dissolved in water and treated with charcoal. The solution was evaporated again to dryness *in vacuo*, and the residue was recrystallized with EtOH–acetone to give colorless prisms, mp 111—113°, which showed no depression of melting point on admixture with the sample obtained by the method of i). IR spectrum of this compound was also identical with an authentic specimen.

2,9a-Dimethyl-9-(2-hydroxyethyl)-7-(3-methoxy-3-phenylpropionyl)-5,9,9a,10-tetrahydro-7H-pyrimido-[4,5-*d*]thiazolo[3,4-*a*]pyrimidine (XV)——To a solution of I (8.4 g) and triethylamine (5.3 g) in MeOH (63 ml) was added 6.6 g of cinnamaldehyde and the mixture was refluxed for 5 hr. The solvent was evaporated and to the residue was added 50 ml of water and 50 ml of AcOEt. Undissolved crystals were collected by filtration, washed with acetone and dried to give 2.3 g of crude crystals of XV. AcOEt extract of the filtrate was dried over Na_2SO_4 and evaporated to dryness. The residue was dissolved in 5 ml of acetone to give further crystals of XV (0.5 g). Total yield 2.8 g (26%). Recrystallization from EtOH gave colorless prisms, mp 197—199° (decomp.). *Anal.* Calcd. for $C_{22}H_{28}O_3N_4S$: C, 61.65; H, 6.59; N, 13.07. Found: C, 61.56; H, 6.71; N, 13.04.

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