UDC 547.853

181. Akira Takamizawa, Kentaro Hirai, and Yoshio Hamashima: Studies on the Pyrimidine Derivatives. XXI.*1 On the Studies of S-Alkoxycarbonylthiamine. (2).

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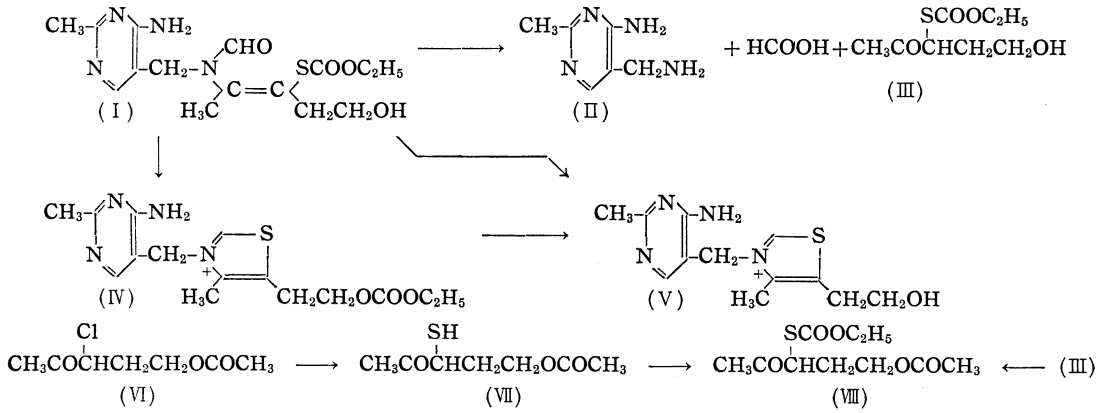
In the previous paper of this series, the syntheses of S-alkoxycarbonylthiamine derivatives were reported. This paper deals with the decomposition of S-ethoxycarbonylthiamine (I) and O,S-bis(ethoxycarbonyl)thiamine (IX) with hydrochloric acid, and with the syntheses of various kinds of O,S-bis(alkoxycarbonyl)thiamine.

S-Ethoxycarbonylthiamine (I) was heated with dil. hydrochloric acid on a boiling water bath for 30 min. After an extraction of the reaction mixture with ether, the aqueous layer was submitted to steam distillation. The distillate was positive to the formic acid reaction.[1] The paper chromatography[2] of the residue showed spots of 2-methyl-4-amino-5-aminomethylpyrimidine (II), thiamine (V) and Rf 0.26. (II) and (V) were isolated as crystals. The spot of Rf 0.26 was found to be identical with that of O-ethoxycarbonylthiamine (IV), prepared by another method.



Ether extract was evaporated to give an oil and acetylated with acetic acid to give an oil of b.$p_{0.5}$ 106~109°. By another method, 3-chloro-5-acetoxy-2-pentanone (VI) was converted to 3-mercapto-5-acetoxy-2-pentanone (VII) by NaSH and was reacted with ethyl chlorocarbonate in pyridine to give 3-ethoxycarbonylthio-5-acetoxy-2-pentanone (VIII). The

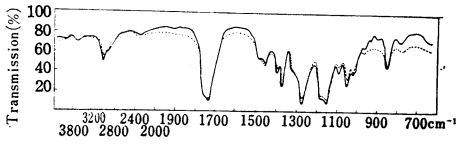


Fig. 1. Infrared Absorption Spectra (film)
—— 3-Ethoxycarbonylthio-5-acetoxy-2-pentone (VIII)
------ b.$p_{0.5}$ 106~109° derived from (I)

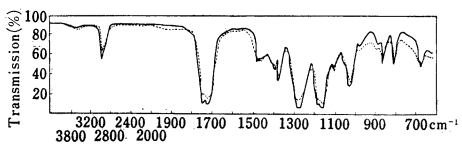


Fig. 2. Infrared Absorption Spectra (film)
—— 3-Ethoxycarbonylthio-5-carboethoxy-2-pentanone (X)
------ b.$p_{0.5}$ 135~141° obtained from (IX)

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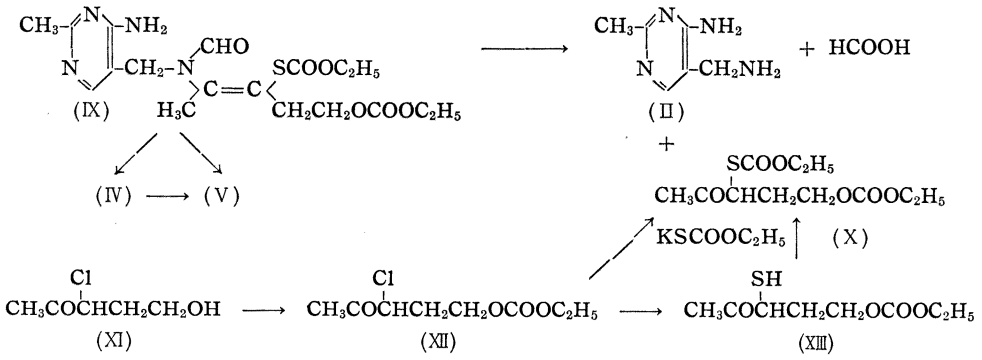
1) T. Matsukawa, H. Kawasaki: Yakugaku Zasshi, **73**, 709 (1953).
2) Paperchromatography: Solvent, BuOH : AcOH : H_2O (4 : 1 : 5). Ascending method. Dragendorff reagent.

infrared spectrum of this compound was identical with that of an oil, b.$p_{0.5}$ 106~109°, derived from HCl decomposition of (I) (Fig. 1). Therefore, HCl decomposition of (I) gave 3-ethoxycarbonylthio-5-hydroxy-2-pentanone (III).

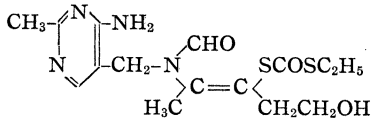
From these facts, it was found that S-ethoxycarbonylthiamine (I), when heated with hydrochloric acid, was decomposed partially into three compounds; 2-methyl-4-amino-5-aminomethylpyrimidine (II), formic acid and 3-ethoxycarbonylthio-5-hydroxy-2-pentanone (III). Further, a part underwent S → O Rearrangement to give O-ethoxycarbonylthiamine (IV) followed by either hydrolysis to thiamine (V) or directly hydrolyzed to thiamine (V). In a simillar manner, when O, S-bis(ethoxycarbonyl)thiamine (IX) was heated with hydrochloric acid, (II) was isolated and (IV), (V) and formic acid were detected.



The ether extract gave an oil of b.$p_{0.5}$ 135~141°. In another method, 3-chloro-5-hydroxy-2-pentanone (XI) was carboethoxylated with ethyl chlorocarbonate in pyridine to 3-chloro-5-ethoxycarbonyl-2-pentanone (XII), which was converted to the SH compound (XIII) and reacted with ethyl chlorocarbonate to give 3-ethoxycarbonylthio-5-ethoxycarbonyl-2-pentanone (X). (X) was also obtained from (XIII) by the action of potasium ethylmonothiocarbonic acid, and its infrared spectrum was found to be identical with an oil of b.$p_{0.5}$ 135~141°, obtained from HCl decomposition of (IX) (Fig. 2).

From these results, the structure O, S-bis(ethoxycarbonyl)thiamine is proposed for (IX).

Further, S-ethylthiocarbonylthiamine (XIV) was prepared by the action of S-ethyl chlorothiocarbonate on thiamine in alkaline solution.



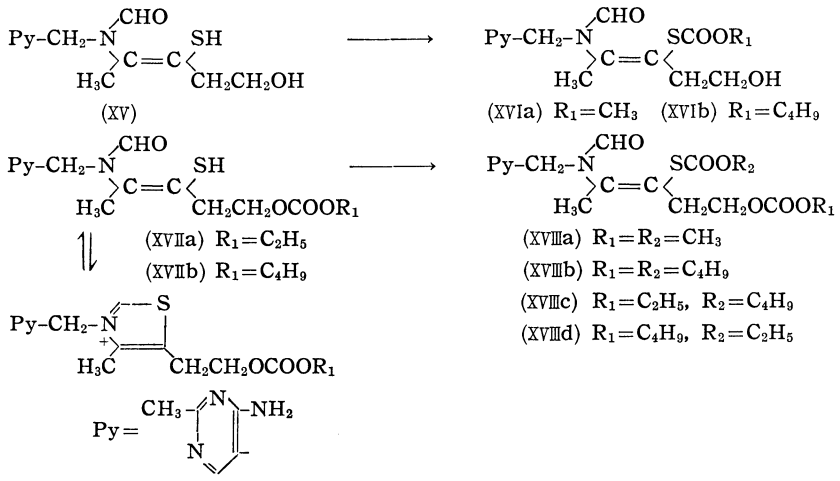
(XIV)

When 2 moles of ethyl chlorocarbonate was reacted with thiamine in alkaline solution, a viscous oil was separated, which, however, did not solidify readily. The paper chromatography of this oil showed a spot of Rf 0.80 besides 0.72. The former was assumed to be the spot of (IX) and the latter was found to be (I). (IX) was isolated from the reaction mixture as the hydrochloride in poor yield. When this reaction was carried out in alcoholic solution with triethylamine, (IX) hydrochloride was obtained in considerable yield. When reacted with methyl chlorocarbonate, O, S-bis(methoxycarbonyl)thiamine(XVIIIa) was obtained and butyl chlorocarbonate provided O, S-bis(butoxycarbonyl)thiamine (XVIIIb).

When thiamine is carbalkoxylated in alkaline solution, it is considered that, at the first step, S-alkoxycarbonylthiamine (XVI) would be formed, and then S → O rear-

rangement occured to give O-alkoxycarbonylthiamine (XVII). Moreover the SH group of (XVII) is carbalkoxylated to yield O, S-bis(alkoxycarbonyl)thiamine (XVIII).

O-Alkoxycarbonylthiamine was obtained from S-alkoxycarbonylthiamine even in acidic solution.



The probable mechanism for this rearrangement could be suggested as shown Chart 1.

For example, S-ethoxycarbonylthiamine (I) was converted to O-ethoxycarbonylthiamine (XVIIa) in sodium ethylate readily. When (I) was dissolved in sodium ethylate and reac ted with butyl chlorocarbonate S-butoxycarbonyl-O-ethoxycarbonylthiamine (XVIIIc) was afforded. This compound was confirmed to be identical with the one obtained by the reaction of butyl chlorocarbonate with O-ethoxycarbonylthiamine (XVIIa) in alkaline solution.

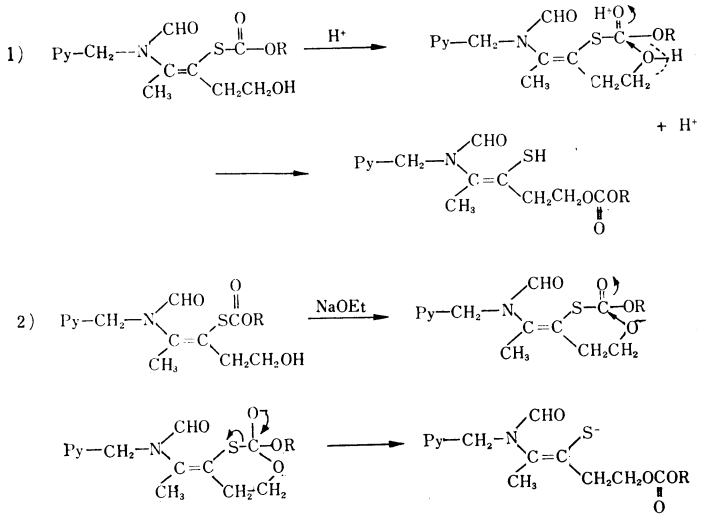
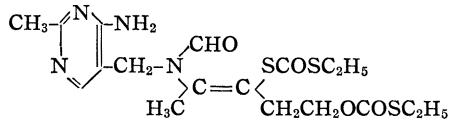


Chart 1.

In a simillar manner, (IX), (XVIIIb) and O,S-bis(ethylthiocarbonyl)thiamine (XIX) were prepared from (I), (XVIb) and (XIV) respectively.



(XIX)

All of these O,S-bis(alkoxycarbonyl)thiamine derivatives and compounds (XIV) and (XIX) showed excellent thiamine activity in absorption from the intestine and thiamine blood level as shown Table I.[3]

Table I. Blood B_1 levels following oral administration of 5 mg./kg. alkoxycarbonylthiamine to rabbits

		Blood B_1 level (γ/dl)					
	hr. / No. of exp.	0	0.5	1	3	5	8
B_1-HCl	4	22.2	24.2	27.5	32.9	34.4	28.7
(XVIIIa)-HCl	4	22.1	62.1	81.2	74.4	62.2	44.0
(XVIIIc)-HCl	3	23.1	32.6	45.0	62.6	60.8	52.1
(XVIIId)-HCl	4	22.1	46.2	61.2	67.2	58.8	53.0
(XIX)-HCl	2	25.4	44.9	61.9	75.0	75.2	66.1

Experimental*3

Decomposition of S-ethoxycarbonylthiamine (I) by HCl——5 g. of (I) was dissolved in 100 cc. of 20% HCl and heated on a boiling water bath for 30 min. After cooling, the reaction mixture was extracted with Et_2O. The Et_2O residue, 1.7 g. of a brown oil, was refluxed with 5.1 g. of glacial AcOH, which was then evaporated *in vacuo* and the residue was distilled to give a colorless oil (0.3 g) of b.$p_{0.5}$ 106~109°. 3-Ethoxycarbonylthio-5-acetoxy-2-pentanone (VIII). *Anal.* Found : C, 48.27; H, 6.57. The IR spectrum was shown in Fig. 1. Mother liquor of the Et_2O extract was submitted to steam distillation, the distillate showing a positive formic acid reaction by chromotropic acid. Residue showed a spot of Rf 0.26, thiochrome reaction positive, besides thiamine. This spot was later identified with O-ethoxycarbonylthiamine. The residue was evaporated *in vacuo* to give a crystalline powder. To this powder, 3 cc. of conc. HCl and 3 cc. of EtOH were added and the undissolved substance was filtered and recrystallized from dil. EtOH to give colorless prisms, m.p. 260~261° (decomp.), which was identified with 2-methyl-4-amino-5-aminomethylpyrimidine (II) dihydrochloride. From the filtrate, colorless needles, m.p. 248~249° (decomp.) were separated and identified with thiamine hydrochloride.

3-Ethoxycarbonylthio-5-acetoxy-2-pentanone (VIII)——To the solution of 1.1 g. of NaOH in 10 cc. of MeOH, H_2S gas was saturated under ice-cooling and a solution of 5 g. of 3-chloro-5-acetoxy-2-pentanone (VI) in 15 cc. of EtOH was added with stirring. The separated NaCl was filtered off and the filtrate was evaporated *in vacuo*.

Water was added to the residual oil and extracted with benzene, which was after drying evaporated to leave 5 g. of crude 3-mercapto-5-acetoxy-2-pentanone (VII), to which was added a solution of 1.1 g. of NaOH in 25 cc. of H_2O and 3.5 g. of ClCOOEt. After stirring for 2 hr., the oil separated was extracted with benzene. The extract was washed with water, dried and evaporated leaving a brown oil. A distillation gave a faint yellow oil of b.$p_{0.5}$ 108~110° (1.4 g.). *Anal.* Calcd. for $C_{10}H_{16}O_5S$: C, 48.35; H, 6.94. Found : C, 48.59; H, 6.73. The infrared spectrum was identical with that of the product derived from HCl decomposition of (I) (Fig. 1).

Decomposition of O,S-Bis(ethoxycarbonyl)thiamine (IX) by HCl—— 1 g. of (IX) was dissolved in 20 cc. of 15% HCl and heated on a boiling water bath for 30 min. After cooling, the reaction mixture was extracted with Et_2O. The Et_2O residue was distilled to give a colorless oil of b.$p_{0.8}$ 135~141°. 3-Ethoxycarbonylthio-5-ethoxycarbonyl-2-pentanone (X). *Anal.* Found : C, 47.78; H, 6.68. Mother liquor of the Et_2O extract was submitted to steam distillation, showing a positive formic acid reaction with the distillate. Residue was evaporated to dryness and triturated with EtOH to give 0.3 g. of crystals, m.p. 233~235° (decomp.). This was recrystallized from dil. EtOH to form colorless prisms, m.p. 262~264° (decomp.), which was identified with 2-methyl-4-amino-5-aminomethylpyrimidine (II) dihydrochloride. Paper chlomatography of filtrate showed a spot of thiamine and a small spot assumed to be O-ethoycarbonylthiamine.

*3 All melting points are not corrected.

3) Biological tests were undertaken by Dr. T. Mineshita *et al.* of this laboratory. A detailed report will be presented elsewhere.

3-Chloro-5-ethoxycarbonyl-2-pentanone (XII)——70 g. of 3-chloro-5-hydroxy-pentanone (XI) was dissolved in 140 cc. of anhyd. pyridine and to this solution was added 67 g. of $ClCOOC_2H_5$ under ice-cooling and the mixture was allowed to stand over night in a refrigerator. To this was added dil. H_2OSO_4 and extracted with $CHCl_3$, which was dried over anhyd. $MgSO_4$ and $CHCl_3$ was removed. The residue was distilled to give a colorless oil, 44.6 g. of $b.p_7$ 107~111°. *Anal.* Calcd. for $C_8H_{13}O_4Cl$: C, 46.05; H, 6.28. Found: C, 46.48; H, 6.41. IR (Film) cm^{-1}: 1740, 1260, 1020, 790 (OCOO).

3-Ethoxycarbonylthio-5-ethoxycarbonyl-2-pentanone (X)——a) To a solution of 5.3 g. of KOH in 26 cc. of MeOH, H_2S gas was saturated under ice-cooling and an EtOH solution of 13.9 g. of (XII) was added and stirred for 2 hr. The separated KCl was filtered off, and the filtrate was concentrated *in vacuo* to give a residual oil, to which was added H_2O and extracted with benzene. The benzene extract, after drying, was evaporated to yield 14 g. of crude 3-mercapto-5-ethoxycarbonyl-2-pentanone (XIII). To this was added a solution of 6.5 g. of KOH in 58 cc. of H_2O and 8.6 g. of ClCOOEt with stirring. After the reaction, it was extracted with benzene and the extract was dried and evaporated. The residue was distilled to give 4 g. of a colorless oil of $b.p_{0.3}$ 118~122°. *Anal.* Calcd. for $C_{11}H_{18}O_6S$: C, 47.47; H, 6.52. Found: C, 47.88; H. 6.68.

b) To a solution of 3.2 g. of (XII) in 15 cc. of MeOH, 2.2 g. of KSCOOEt was added in small portions. After stirring for 1.5 hr., the separated KCl was filtered off and the filtrate was evaporated *in vacuo*. H_2O was added to the residue and extracted with benzene, which was dried and the solvent was distilled to give 2.8 g. of residual oil. The infrared spectrum was identical with the oil obtained in a).

S-Ethylthiocarbonylthiamine (XIV)——3.3 g. of thiamine hydrochloride was dissolved in a solution of 1.2 g. of NaOH and 9 cc. of H_2O. After standing for 30 min., 1.25 g. of S-ethylthiochloroform was added with stirring for 2 hr. under cooling. The reaction mixture was extracted with $CHCl_3$, the extract dried and the solvent was removed. The residue was triturated with AcOEt and recrystallized from benzene to colorless prisms, m.p. 136~137° (decomp.), yield, 1.4 g. It gave a negative thiochrome reaction but became positive after treatment with alkali. Rf 0.78. *Anal.* Calcd. for $C_{15}H_{22}O_3N_4S_2$: C, 48.62; H, 5.99; N, 15.12; S, 17.31. Found: C, 48.63; H, 6.19; N, 14.88; S, 17.53.

O,S-Bis(ethoxycarbonyl)thiamine (IX)——a) To a solution of 3.5 g. of Na in 200 cc. of 99% EtOH was added the solution of 17 g. of thiamine in 17 cc. of H_2O. After standing for 30 min., 5 g. of $(C_2H_5)_3N$ was added and further 10.8 g. of ClCOOEt in small portions. Stirring was continued for 2 hr. at 45~48°. After a filtration, the filtrate was concentrated under reduced pressure. The oily residue was dissolved in $CHCl_3$ and washed with dil AcOH and H_2O. After shaking with 15% HCl, the $CHCl_3$ layer was dried over anhyd. $MgSO_4$ and evaporated. The oily residue solidified gradually, yield, 10.3 g., which was recrystallized from Me_2CO to colorless prisms, m.p. 121~123° (decomp.). The free base was recrystallized from AcOEt-pet. ether to colorless prisms m.p. 113.5~114.5°. *Anal.* Calcd. for $C_{18}H_{26}O_6N_4S$: C, 50.70; H, 6.15; N, 13.14. Found: C, 50.83; H, 6.36; N, 13.10.

b) To the solution of 0.65 g. of Na in 100 cc. of 99% EtOH was added 10 g. of S-ethoxycarbonylthiamine. After standing for several min., 3.1 g. of ClCOOEt was added and the mixture was warmed to 45~50° for 10 min. After concentration, it was extracted with $CHCl_3$ and shaken with 15% HCl. The $CHCl_3$ layer was dried, evaporated and the residue was recrystallized from Me_2CO to colorless prisms, m.p. 121~122° (decomp.), yield, 11 g.

O,S-Bis(methoxycarbonyl)thiamine (XVIIIa)——a) To the solution of 2.7 g. of Na in 100 cc. of MeOH was added the solution of 10 g. of thiamine hydrochloride in 10 cc of H_2O. After standing for 30 min., 5.6 g. of ClCOOMe was added and stirred for 3 hr. at 40°, then treated as above. The product obtained was recrystallized from Me_2CO to colorless prisms, m.p. 135° (decomp.), yield, 2.1 g.

b) 10 g. of (XVIa) was dissolved in a solution of 0.64 g. of Na and 100 cc. of MeOH and to this solution was added 2.66 g. of ClCOOMe. After 2 hr. at 40°, this mixture was concentrated and extracted with $CHCl_3$. Recrystallization of the residue from Me_2CO gave 11 g. of colorless prisms, m.p. 139°, yield, 7.2. *Anal.* Calcd. for $C_{16}H_{22}O_6N_4S$: C, 48.24; H, 5.57; N, 14.07. Found: C, 48.20; H, 5.68; N, 14.02.

O,S-Bis(butoxycarbonyl)thiamine (XVIIIb)——a) 17.3 g. of thiamine hydrochloride and 13.7 g. of ClCOOBu were worked up as above to give 4.8 g. of colorless prisms, m.p. 83~84° (hydrochloride).

b) 10 g. of (XVIb) was treated in a similar manner to give 8.5 g. of colorless prisms, m.p. 85° (from AcOEt). *Anal.* Calcd. for $C_{22}H_{35}O_6N_4SCl \cdot H_2O$: C, 49.19; H, 6.94; N, 10.43. Found: C, 49.32; H, 7.09; N, 10.48.

O,S-Bis(ethoxycarbonylthio)thiamine (XIX)——4.06 g. of (XIV) hydrochloride reacted with 1.25 g. of ClCOSEt by the usual method to give 1 g. of colorless prisms (from AcOEt), m.p. 155~157° (decomp.) (hydrochloride). *Anal.* Calcd. for $C_{18}H_{26}O_4N_4S \cdot HCl$: C, 43.70; H, 5.47; N, 11.32. Found: C, 43.45; H, 5.73; N, 11.08.

O-Ethoxycarbonylthiamine (XVIIa)——a) To a solution of 0.65 g. of Na in 100 cc. of EtOH was added 10 g. of (I). After stirring for 5 min. under ice-cooling, 9.9 g. of conc. HCl was added. This was concentrated *in vacuo* and recrystallized from EtOH to give colorless needles, m.p. 207° (decomp.), yield, 3.9 g.

b) 8 g. of (I) hydrochloride was dissolved in 160 cc. of H_2O and heated for 4 hr. on a boiling water bath. After cooling, 2.1 g. of conc. HCl was added and concentrated *in vacuo* below 45°. The residue was dissolved in EtOH, allowed to stand in a refrigerator, and the crystalline precipitate was separated. It was recrystallized from EtOH to give 2.8 g. of colorless needles, m.p. 207° (decomp.). *Anal.* Calcd. for $C_{15}H_{21}O_3N_4SCl \cdot HCl \cdot H_2O$: C, 42.15; H, 5.62; N, 13.11. Found : C, 42.58; H, 5.90; N, 12.89. Rf 0.26. IR ν_{max}^{Nujol} cm^{-1} : 1750, 1260, 1010 (OCOO).

O-Butoxycarbonylthiamine (XVIIb)——10 g. of (XVIb) was treated as above to give 3.5 g. of colorless prisms, m.p. 231° (from C_2H_5OH). *Anal.* Calcd. for $C_{17}H_{25}O_3N_4SCl$: N, 12.81. Found : N, 12.84.

S-Butoxycarbonyl-O-ethoxycarbonylthiamine (XVIIIc)——a) 10 g. of (I) was dissolved in NaOEt and reacted with 3.9 g. of ClCOOBu to give 13 g. of crude crystals. Recrystallization from Me_2CO gave colorless prisms, m.p. 136° (decomp.). *Anal.* Calcd. for $C_{20}H_{30}O_6N_4S \cdot HCl \cdot \frac{1}{2}H_2O$: C, 48.04; H, 6.45; N, 11.21. Found : C, 48.08; H, 6.56; N, 11.19.

b) To a solution of 2 g. of (XVIIa) in 50 cc. of H_2O was added dil. NaOH and reacted with 0.6 g. of ClCOOBu to give 0.9 g. of colorless prisms, which were identical with the sample obtained as above in IR spectra.

S-Ethoxycarbonyl-O-butoxycarbonylthiamine (XVIIId)——a) 10 g. of (XVIb) was dissolved in NaOEt and reacted with 2.8 g. of ClCOOEt. The reaction mixture was treated by the usual method to give 10 g. of crude crystals. Recrystallization from AcOEt gave colorless prisms, m.p. 105~106° (decomp.). *Anal.* Calcd. for $C_{20}H_{30}O_6N_4S \cdot HCl \cdot H_2O$: C, 47.19; H, 6.50; N, 11.01. Found : C, 47.21; H, 6.69; N, 11.01.

b) 2 g. of (XVIIb) was dissolved in dil. NaOH and reacted with 0.5 g. of ClCOOEt to give 0.5 g. of colorless prisms, which were identical with the sample obtained as above in IR spectra.

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Summary

HCl decomposition of S-ethoxycarbonylthiamine (I) gave 2-methyl-4-amino-5-aminomethylpyrimidine (II), formic acid and 3-ethoxycarbonylthio-5-hydroxy-2-pentanone (III). 3-Ethoxycarbonylthio-5-acetoxy-2-pentanone (VIII) was prepared. A part of it was decomposed into O-ethoxycarbonylthiamine (IV) and thiamine (V). Also O,S-bis(ethoxycarbonyl)thiamine (IX) decomposed into (II), formic acid and 3-ethoxycarbonylthio-5-ethoxycarbonyl-2-pentanone (X) by heating with HCl. A part was also decomposed into (IV) and (V). (X) was characterized by synthesis. Further, ethylthiocarbonylthiamine (XIV) was prepared, and O-alkoxycarbonylthiamine was obtained by rearrangement of S-alkoxycarbonylthiamine. By the use of this rearrangement, various kinds of O,S-bis(alkoxycarbonyl)thiamine were prepared. All of these thiamine derivatives showed excellent thiamine activity in absorption from intestine and in thiamine blood level.

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