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STUDIES ON PYRIMIDINE DERIVATIVES AND RELATED COMPOUNDS. LIV.*1 REACTION OF THIAMINE WITH PHENYLGLYOXAL AND SYNTHESIS OF HYDROXYMETHYLTHIAMINE

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In relation to the mechanism of thiamine action, the chemical reactivity of C-2 carbon atom of the thiazolium portion of thiamine molecule has recently attracted considerable attentions (1). Previously, the authors reported on the reaction of thiamine with aldehydes involving anomalous C-acylation at the 2-position of the thiazolium nucleus (2). This communication describes a new reaction of thiamine with phenylglyoxal and a synthesis of hydroxymethylthiamine, a postulated intermediate in the glyoxylate metabolism (3).

Ethanolic suspension of thiamine-sodium salt (I) readily reacted with phenylglyoxal in the presence of carbon dioxide at room temperature to give a yellow crystalline compound II, m.p. 150-158° (d.), as hemihydrate $C_{20}H_{22}N_4O_3S \cdot \frac{1}{2}H_2O$,*2 in 50-60% yield accompanied with thiamine-thiazolone (III) and racemic 1,2-dibenzoylethylene glycol (IV) (4) as minor products. The structure of the compound II was determined to be 2-phenyloxalyl-3-(2-methyl-4-aminopyrimidin-5-yl)methyl-4-methyl-5β-hydroxyethylthiazoline on the basis of the following evidences. By the action of hydrochloric acid, II was converted into thiamine hydrochloride (VI) in almost quantitative yield. In pyridine or dimethylsulfoxide, II readily underwent air oxidation to yield III and phenylglyoxylic acid. When the air oxidation was carried out in the presence of o-phenylenediamine in methanol, III and 3-phenylquinoxalin-2-one (5) were obtained. The property of this facil air oxidation of the compound II suggested the presence of a-diketone group, for the oxidative C-C bond cleavage of this type has been exemplified by the ready air

*1 Part LIII. Chem. Pharm. Bull. (Tokyo), in contribution.

*2 All compounds given by formula in this communication gave correct elementary analysis.

oxidation of benzylphenyl-α-diketone to benzaldehyde and phenylglyoxylic acid (6).

In IR spectrum (nujol), however, II exhibited no carbonyl absorption besides 1660 cm^{-1}, while it showed three UV absorption maxima (in EtOH) at 233 mμ (log ε = 4.09), 274 mμ (log ε = 3.83) and 420 mμ (log ε > 3.90), *but in acidic medium only single maximum was observed at 250 mμ*. These spectral properties indicated that II might predominantly exist in an enolic form II'. In fact, treatment of II with acetic anhydride-pyridine mixture under nitrogen stream afforded a stable diacetate V, m.p. 176-178° (d.), $C_{24}H_{26}N_4O_5S$ [UV λ_{max}^{EtOH} mμ: 234 (log ε = 4.32), 272 (log ε = 3.90) and 391 (log ε = 4.40)]. IR spectrum of V showed absorptions due to three different carbonyl groups at 1763 cm^{-1} (enol-acetate), 1730 cm^{-1} (alcohol-acetate) and 1636 cm^{-1} (α,β-unsaturated ketone) and its NMR spectrum was quite consistent with the structure [(60 M.c.p.s., d_6-DMSO): singlets at 2.86 τ (1H, pyrimidine-H), 3.11 τ

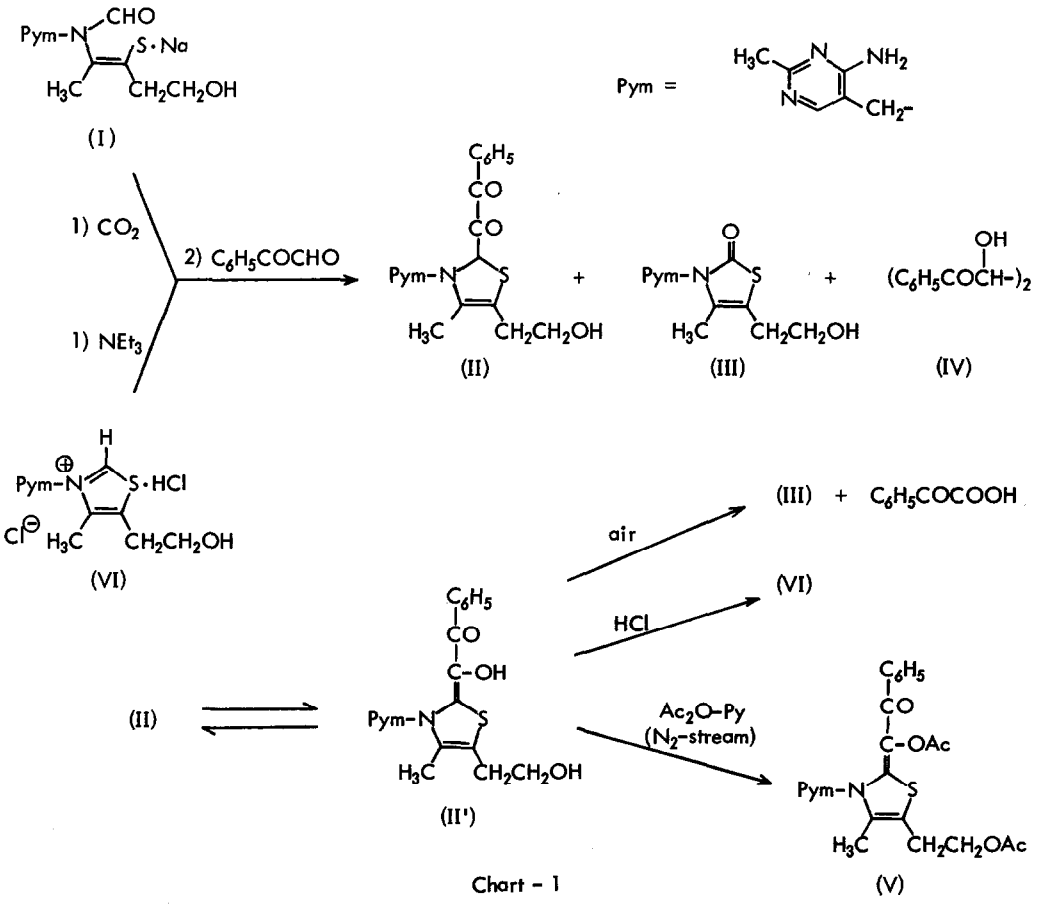

Chart - 1

(broad, 2H, NH_2), 7.68 τ, 7.91 τ, 7.97 τ and 8.43 τ (each 3H, pyrimidine-CH_3, =C-CH_3, -$CH_2OCOC\underline{H}_3$ and =C-$OCOCH_3$ respectively), quartet at 4.90 τ (2H, pyrimidine-CH_2) and a pair of triplets at 5.77 τ and 7.04 τ (each 2H, =C-CH_2CH_2O-)]. These data cited above led to the conclusion that the aldehydic carbonyl group of phenylglyoxal was exclusively condensed with the "active site" of thiamine to give II as a major product which was proved to exist preferably in the enolic form II'.

Reaction of thiamine hydrochloride (VI) with phenylglyoxal in the presence of triethylamine was also found to proceed analogously providing II in a moderate yield accompanied with a small amount of III and IV. By the action of ethanolic hydrogen chloride, V was converted into hydroxymethylthiamine hydrochloride (VII), m.p. 225-226° (d.), as monohydrate, $C_{13}H_{20}N_4O_2SCl_2 \cdot H_2O$ [UV λ_{max}^{EtOH} mμ: 239 (log ε = 4.12) and 265 (log ε = 4.20), NMR (60 M.c.p.s., D_2O): singlets at 2.55 τ (1H, pyrimidine-H), 4.40 τ (2H, pyrimidine-CH_2), 4.80 τ (2H, $^{+}$N/S>-$\underline{CH_2}$OH), 7.33 τ and 7.52 τ (each 3H, pyrimidine-CH_3 and =C-CH_3), a pair of triplets at 6.05 τ and 6.76 τ (each 2H, =C-$\underline{CH_2}$-$\underline{CH_2}$-OH)]. The structure of VII was confirmed by the formation of tribenzoate VIII, m.p. 160-162.5° $C_{34}H_{32}N_4O_6S$ [IR ν_{max}^{Nujol} cm^{-1}: 1725, 1669 (each CO), UV λ_{max}^{EtOH} mμ: 232.5, 275, NMR (60 M.c.p.s., $CDCl_3$): singlet at 5.33 τ (2H,

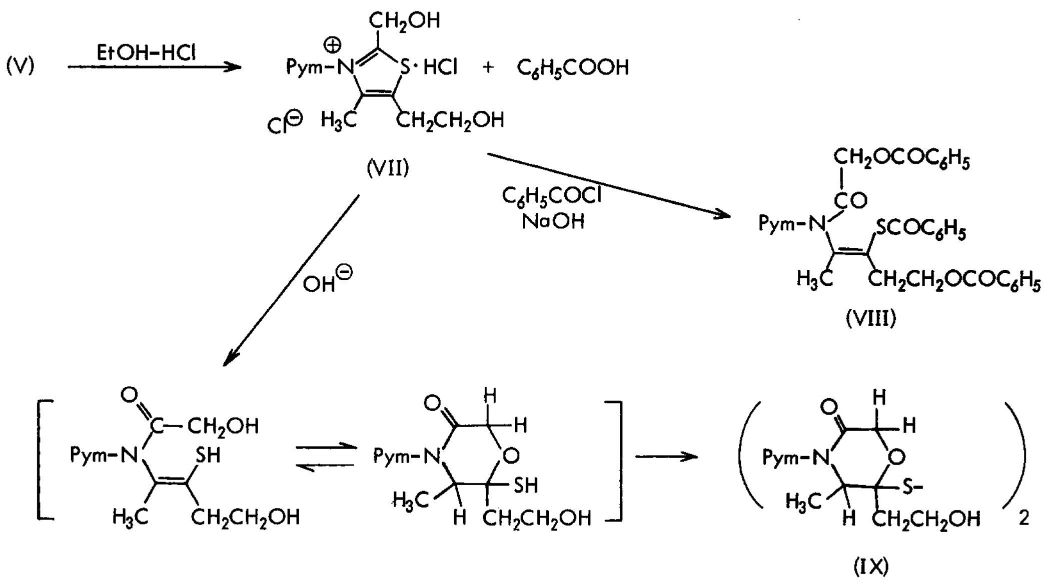

Chart - 2

-N(CO-CH$_2$-OCOPh)] on the treatment with benzoylchloride in an aqueous sodium hydroxide solution. Moreover, VII was converted into a dimeric oxazine derivative IX, m.p. 232° (d.), as monohydrate, $(C_{13}H_{19}N_4O_3S_2)_2 \cdot H_2O$ [IR ν_{max}^{Nujol} cm^{-1}: 1660 (CO), NMR (60 M.c.p.s., d_6-DMSO): doublet at 8.72 τ (3H, J = 6 c.p.s., -CH-CH$_3$) and a pair of doublets at 5.29 τ and 6.04 τ (each 1H, J = 15 c.p.s., -N(CO-CH$_2$-O-)] when stirred for two days at room temperature in the presence of two equimolar amounts of aqueous sodium hydroxide. An analogous compound has been also obtained from hydroxyethylthiamine (7), therefore it provides a further support for the structure of VII.

The potent catalytic activity of thiamine for the acyloin forming reaction of aldehydes has been found first by Ugai and coworkers (8). It is interesting however that the reaction of phenylglyoxal with thiamine

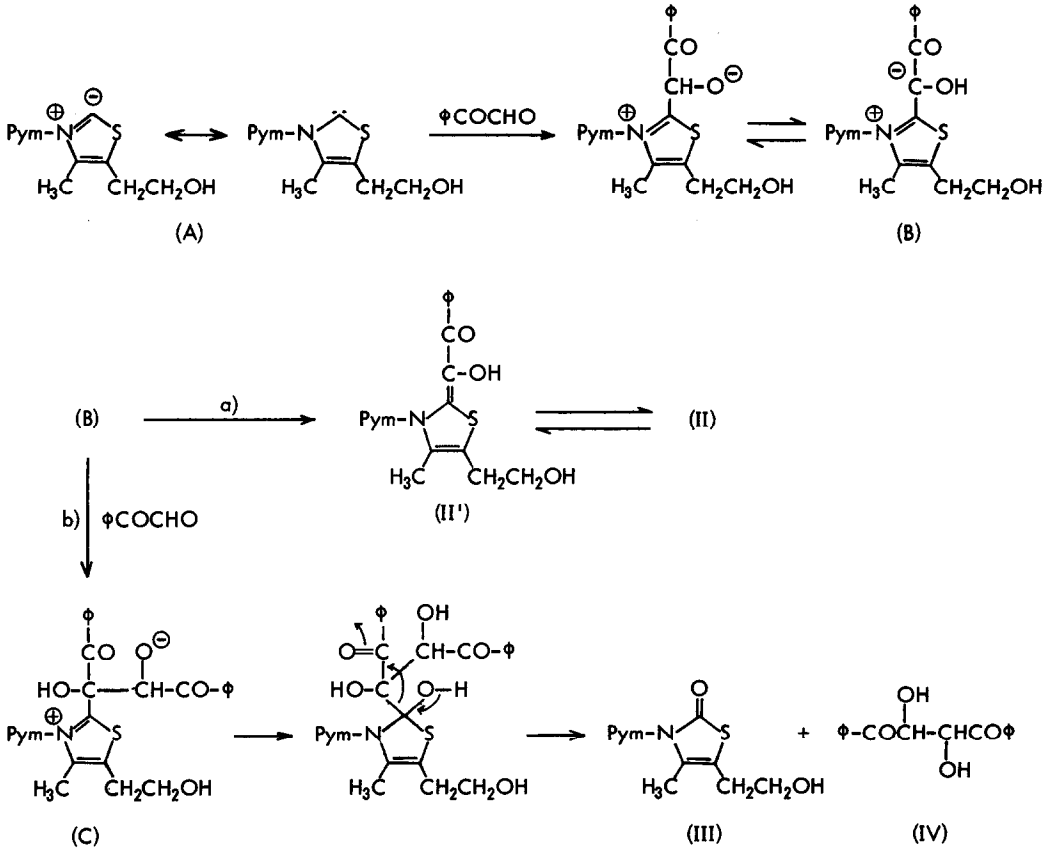

Chart - 3

unexpectedly yielded IV, a dihydroderivative of benzoylformoin which is the expected acyloin product, though it was in poor yield. The reaction may probably be rationalized by the formulation illustrated in Chart-3. The thiazoium-zwitterion (nucleophilic carbene) (1, 9) (A) initially formed from thiamine (sodium salt or hydrochloride) would first add to phenylglyoxal to form carbanion (B) from which the major product II' would be produced by a simple isomerization via path a). While the carbanion (B) might possibly undergo further condensation with remaining phenylglyoxal to give (C) via path b), and hydrolytic cleavage of (C) would lead to the minor products, III and IV.

The present results provide an example that thiamine is readily condensed with α-ketoaldehyde under mild condition and the product undergoes facil air oxidation to generate α-keto acid, which is notable from the biochemical point of view. The reaction has been confirmed also for a number of α-ketoaldehydes including some heterocyclic glyoxals. Detailed investigation of the reaction will be described in another paper.

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