Analytical Laboratory of the Faculty of Pharmaceutical Sciences, University of Tokyo for elemental analysis and IR spectra and to the members of Botanical Institute of Tokyo Metropolitan University for the kind suggestion about plant material.

School of Pharmaceutical Sciences,
Showa University,
Hatanodai, Shinagawa-ku, Tokyo

Faculty of Pharmaceutical Sciences,
University of Tokyo,
Hongo, Tokyo

Junzo Shoji (庄司順三)
Sachiko Kawanishi (川西幸子)
Seiichi Sakuma (佐久間聖一)
Shoji Shibata (柴田承二)

Received December 26, 1966

[Chem. Pharm. Bull. 15(5) 723~725 (1967)]

UDC 577.16B : 547.9.04

The Free Radical Reaction of Thiamine

Previously, the pyrolysis of thiamine disulfide and the reaction of S-anion (II) of thiamine (I) with potassium ferricyanide (III) giving thiochrome have been discussed by A. R. Todd, *et al.*[1~3] They have postulated the reaction mechanism involving a radical process for these reactions. However, no coupling products of the S-radical with the other free radicals have been obtained. Generally, an anion $R\text{-}S^-$ reacts with 1-equivalent electron abstracting reagent such as potassium ferricyanide to give a radical R-S., and then the radical dimerizes.[4] This radical would also be able to couple with some other radicals. For the confirmation of the radical mechanism of this reaction, 4-oxo-2,2,6,6-tetramethylpiperidine-1-oxide (V)[5] or 4-methyl-2,6-di-t-butylphenoxy radical[4] was used as stable free radicals to scavenge the S-radical.

In the presence of V, the anion II reacted with III at 5° to give a radical (IV), which reacted immediately with V to afford a new derivative of thiamine (VIII) accompanied by the disulfide (IX). It is considered that the sulfenamide compound VIII would be formed via the coupling intermediate, (VI) or (VII). The reaction mechanism can be illustrated as shown in Chart 1.

VIII showed m.p. 179° (from MeOH). *Anal.* Calcd. for $C_{21}H_{33}O_3N_5S$: C, 57.91; H, 7.64; N, 16.08; S, 7.36. Found : C, 57.78; H, 7.76; N, 16.29; S, 7.39. UV λ_{max}^{EtOH} mμ (log ε) : 235 (4.17); 276 (3.77) (shoulder). IR cm^{-1} ($CHCl_3$ solution, 1.0 m/mNaCl, Grating) : $\nu_{O\text{-}H}$ 3620; $\nu_{asN\text{-}H}$; 3480 $\nu_{sN\text{-}H}$ 3335; $\nu_{C=O}$ 1715. NMR (τ)[6] in d_6-DMSO : 2.08 (singlet); 2.13 (singlet), -N-CHO and -CH=in Pyrimidine. Rf value of paperchromatography : 0.70 (*n*-BuOH, AcOH and H_2O (4:1:5), Toyo-filter paper No. 51). And VIII was negative to the the thiochrome test[7] and turned out to be positive after the reduction with cysteine. The hydrolysis of VIII with hydrochloric acid at 80° gave 2-methyl-4-amino-5-aminomethylpyrimidine dihydrochloride (X) and 4-oxo-2,2,6,6-tetramethylpiperidine hydrochloride (XI). The reduction of VIII with thiophenol in weak acidic aqueous solution at 25°

1) A. R. Todd, P. Sykes : J. Chem. Soc,. **1951**, 534.
2) P. Nesbitt, P. Sykes : *Ibid.*, **1954**, 4584.
3) S. Yurugi : Yakugaku Zasshi, **77**, 259; 264 (1957).
4) R. Stewart : "Oxidation Mechanisms," 85 (1964), W. A. Benjamin, New York.
5) E. G. Rozantzev, M. B. Neiman : Tetrahedron, **20**, 131 (1964).
6) K. Kotera : This Bulletin, **13**, 440 (1965).
7) W. Karrer : Helv. Chim. Acta, **20**, 369 (1937).

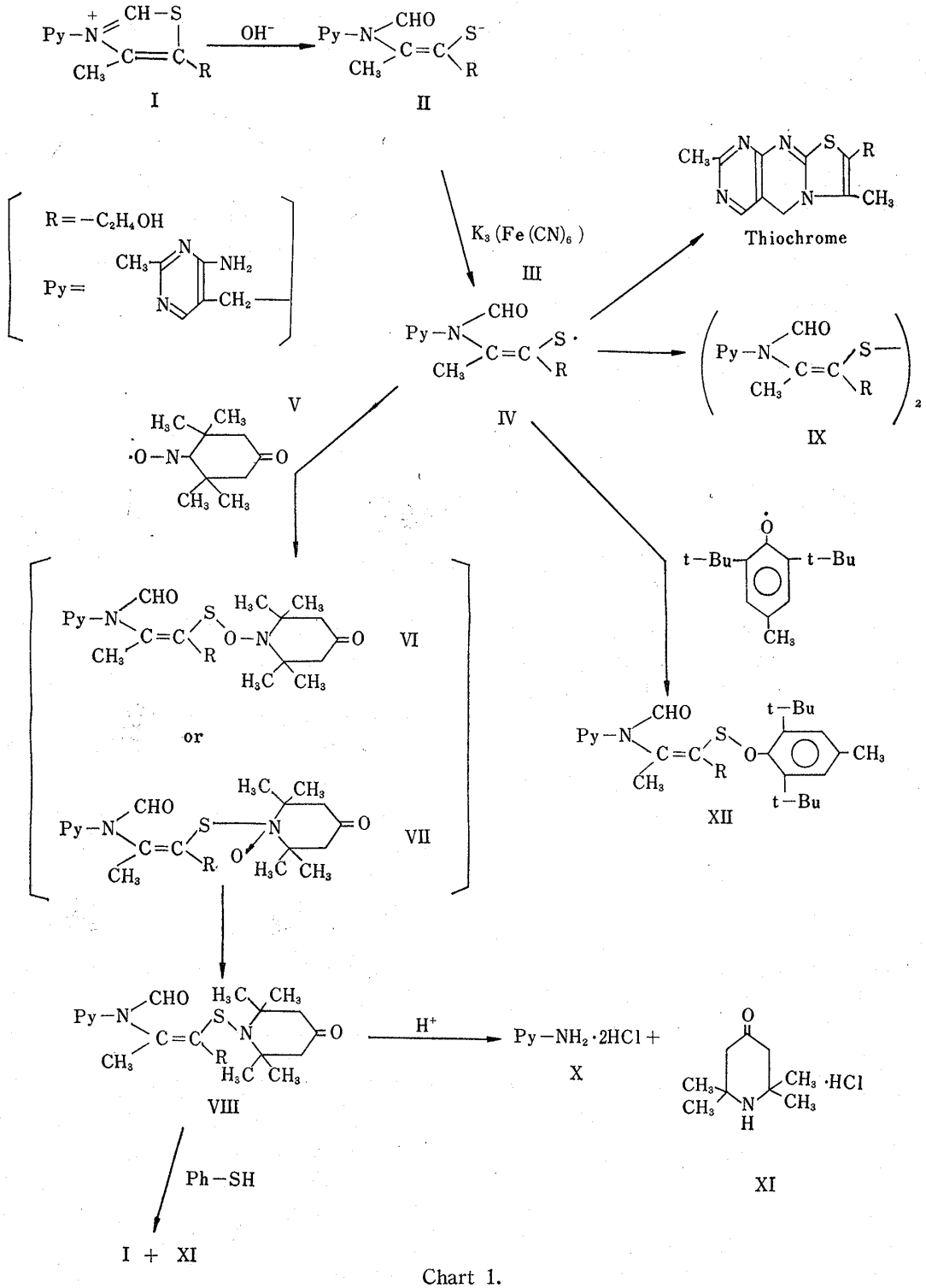
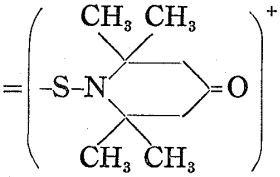
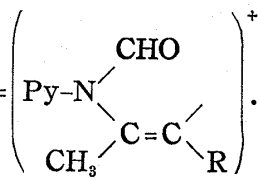


Chart 1.

gave thiamine hydrochloride I and XI quantitatively. Furthermore, high resolution doublefocusing mass spectra were obtained using a JEOL-JMS-OlS mass spectrometer with ionizing voltage of 70 V and 40 V. The mass spectra of VIII contained the expected molecular ion peak at m/e 435.230, and large fragment peak at m/e 122.186=$C_6H_8N_3$=$(Py)^+$; 186.098=$C_9H_{16}ONS$=  and 249.136=$C_{12}H_{17}O_2N_4$=  . The

structure Ⅷ is supported from these facts.

A similar experiment has been carried out on a phenoxy radical instead of V in order to prove the free radical process more definitely. In this case, the normal coupling product (Ⅻ) was obtained at 0~5° accompanied by the disulfide Ⅸ. Ⅻ showed m.p. 197° (from MeOH). *Anal.* Calcd. for $C_{27}H_{40}O_3N_4S$: C, 64.80; H, 8.00; N, 11.20; S, 6.40. Found: C, 64.52; H, 8.04; N, 10.98; S, 6.26. UV λ_{max}^{EtOH} mμ (log ε): 234.5 (4.22); 276.5 (4.03). Further Ⅻ was negative to the thiochrome test and turned out to be positive after the reduction with cysteine. From these facts, Ⅻ is confirmed to have normal coupling structure and S-alkyl structure should be excluded for Ⅻ.

Therefore, it has been shown that the reaction of thiamine in thiol form with $K_3(Fe(CN)_6)$ involves a free radical intermediate, since the typical coupling products have been isolated, and that syntheses of new types of thiamine derivatives are accomplished by these methods.

Central Research Laboratories,
Sankyo Co., Ltd.
1-2 Hiromachi, Shinagawaku, Tokyo

Keisuke Murayama (村山圭介)
Takao Yoshioka (吉岡孝雄)

Received January 19, 1966

[Chem. Pharm. Bull. 15(5) 725~726 (1967)]

UDC 547.92.02 : 582.938

Components of *Boucerosia aucheriana* Decne

Boucerosia aucheriana Decne (Asclepiadaceae) is a plant indigenous to Pakistan and known to have a very bitter taste. A dried collection of whole plants from the vicinity of Peshawar, Pakistan, furnished a mixture of glycosides after usual isolation procedure.[1] The mixture consists of various glycosides and, after mild acid hydrolysis, cymarose, sarmentose, oleandrose and digitoxose were detected as the sugar components.

The aglycone part is also a noncrystalline mixture of diversified esters and resistent to further purification. After alkaline hydrolysis, however, it was partitioned into a crystalline neutral fraction and various acids, *i.e.* benzoic acid, acetic acid, propionic acid, *n*-butyric acid, isovaleric acid and *n*-valeric acid.

The neutral fraction holds two closely related components; boucerin, $C_{21}H_{34}O_4$*[1] (Ⅰ), m.p. 239~241°, $[\alpha]_D$ −3.7° (c=0.26, MeOH), IR ν_{max}^{Nujol} cm^{-1}: 3356, 1063, 848, NMR τ (in pyridine): 8.93 (singlet, 3H), 8.53 (doublet, 3H, J=6 c.p.s.), 8.33 (singlet, 3H), 4.51 (multiplet, 1H), and dihydroboucerin, $C_{21}H_{36}O_4$ (Ⅱ), m.p. 143°/205°, $[\alpha]_D$ +3.9° (c=0.29, MeOH). IR ν_{max}^{Nujol} cm^{-1}: 3380, 3320, 1043, NMR τ (in pyridine): 8.81 (singlet, 3H), 8.57 (doublet, 3H, J=6c.p.s.), 8.36 (singlet, 3H). The difference between both compounds lies only in one double bond, thus Ⅰ was transformed to Ⅱ by catalytic hydrogenation. The absence of a carbonyl function is evident from the IR spectra and a normal C_{21} steroidal structure with a hydroxyl function at C-20 was anticipated from the NMR data.

Acetylation of Ⅰ and Ⅱ with pyridine-acetic anhydride gave corresponding triacetates: (Ⅲ), $C_{27}H_{40}O_7$, m.p. 147~149°, IR ν_{max}^{Nujol} cm^{-1}: 3520, 3430, 1737, 1723, 1260, 1242, NMR τ (in $CDCl_3$): 8.98 (6H), 8.81 (doublet, 3H, J=6 c.p.s.), 7.96 (6H), 7.87 (3H), 4.60

*1 Satisfactory analytical results were obtained for all compounds described in this communication.

1) J. v. Euw, H. Hess, P. Speiser, T. Reichstein: Helv. Chim. Acta, **34**, 1821 (1951).