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Shojiro Uyeo,*1 Katsumi Kotera,*2 Tetsuo Okada,*2 Shuzo Takagi,*3 and Yoshisuke Tsuda*4 : Occurrence of the Alkaloids Vittatine and Haemanthamine in *Lycoris radiata* HERB.*5

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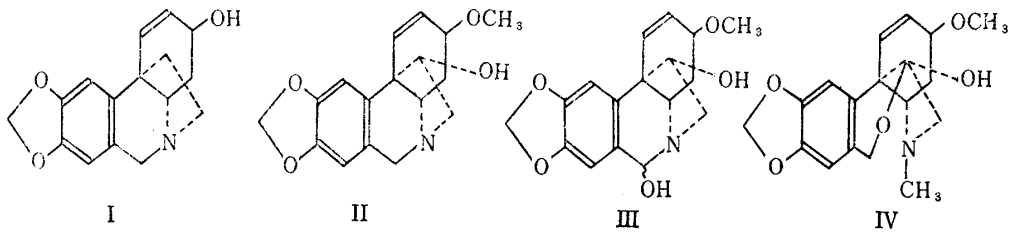
In the course of our systematic investigations on the minor alkaloids of *Lycoris radiata* HERB. (Amaryllidaceae)[1] we have further isolated two known alkaloids vittatine (I), $C_{16}H_{17}O_3N$, m.p. 206~208°, $[\alpha]_D$ +36.6° (EtOH), and haemanthamine (II), $C_{17}H_{19}O_4N$, m. p. 203~205°, $[\alpha]_D$ +43° ($CHCl_3$). The former was obtained by chromatography of the strongly basic fraction of the total bulb-extracts and the latter from the moderately basic fraction.

Vittatine is an alkaloid first isolated by Boit[2] and was shown to be the optical antipode of crinine,[3] whose structure has been firmly established by Wildman.[4] Comparison of our first alkaloid with crinine which was kindly provided by Dr. Wildman was carried out by one of the authors (S. U.) in 1957 who stayed at that time at the National Institute of Health, Bethesda, Maryland. They had the same melting point, the equal absolute value of the optical rotation in opposite directions, and exhibited the superimposable infrared spectra.

Haemanthamine is an alkaloid whose structure (II) was established by Fales and Wildman.[5] The identity of our second alkaloid with authentic haemanthamine, kindly furnished by Dr. Fales, was proved by mixture melting point determination, and by comparison of infrared spectra and optical rotations.



I II III IV

Since vittatine and haemanthamine have been shown to belong to the same series of alkaloids as those of haemanthidine (III) and tazettine (IV) in absolute stereochemistry[6] and haemanthamine has been established to be a biological precursor of tazettine,[7]

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*5 A part of this paper (vittatine) was presented at the Annual Meeting of the Pharmaceutical Society of Japan, Nagoya, April 1958.

1) The most recent paper on this series appeared in : S. Uyeo, Y. Yamato : Yakugaku Zasshi, **85**, 615 (1965).
2) H.-G. Boit : Chem. Ber., **89**, 1129 (1956).
3) H.-G. Boit, H. Ehmke : *Ibid.*, **90**, 369 (1957).
4) W. C. Wildman : J. Am. Chem. Soc., **80**, 2567 (1958).
5) H. M. Fales, W. S. Wildman : *Ibid.*, **82**, 197 (1960).
6) *Idem* : *Ibid.*, **82**, 3368 (1960).
7) *Idem* : *Ibid.*, **86**, 294 (1964).

co-occurrence of these four alkaloids in the same plant implies that vittatine is possibly converted into haemanthamine on the pathway of tazettine biosynthesis.

Experimental

Isolation of Vittatine—— The mother liquors (33 g.), after separation of most of lycorine, lycorenine, homolycorine, tazettine, lycoramine, galanthamine, and phenolic alkaloids from the total alkaloids of *Lycoris radiata* Herb., was dissolved in 5% HCl (1L.), and washed with $CHCl_3$. A small amount of K_2CO_3 was added to the solution to adjust the pH to 6.6 and the aqueous layer was extracted with EtOAc to give fraction 1. This operation was repeated 20 times, increasing the pH value of the aqueous phase stepwise by ca. 0.2 for each extraction, and total extracts consisting of 20 fractions were examined. Fraction 1~14 (pH 6.6~9.2, 5L. in total) gave a further crop of tazettine, hippeastrine, haemanthidine, galanthamine, lycor amine, lycorenine, and pluviine. Fraction15 (pH 9.4, 1L.), on evaporation after drying over K_2CO_3, left a gum (6 g.), which gave lycorenine (1 g.) on trituration with acetone. The mother liquor from the crystallization of lycorenine was dissolved in benzene (200 ml.), filtered to remove insoluble amorphous material (0.2 g.), chromatographed on an alumina column (32×1.5 cm.), and the column was eluted successively with the following solvents : (a) benzene (400 ml.); (b) EtOAc-benzene (1:19) (300 ml.); (c) EtOAc-benzene (1:9) (300 ml.); (d) EtOAc-benzene (1:9) (600 ml.); (e) EtOAc-benzene (1:1) (600 ml.); (f) EtOAc-benzene (1:1) (300 ml.); (g) EtOAc (200ml.); and (h) EtOAc (300 ml.). The eluting solvent (a), (b), and (c) gave mainly lycoramine (3.5 g.); solvent (d) and (e) gave lycorenine (1 g.); solvent (f) gave a mixture of alkaloids; solvent (g) gave vittatine (0.2 g.); and solvent (h) eluted a mixture of vattatine and lycorine. Vittatine was crystallized from benzene and then from acetone to give prismatic needles, m.p. 206~208°, $[\alpha]_D^{21}$ +36.6°(c=0.46,EtOH). UV λ_{max}^{EtOH} mμ (log ε) : 240 (3.50), 295 (3.69). An analytical sample was dried over P_2O_5 for 10 hr. at 15°. *Anal.* Calcd. for $C_{16}H_{17}O_3N$: C, 70.83; H, 6.32; N, 5.16. Found : C, 70.24; H, 6.10; N, 5.31. The IR spectrum of this alkaloid in Nujol mull was superimposable on that of crinine. The picrate formed yellow prisms, m.p. 235° (decomp.) (from MeOH).

Isolation of Haemanthamine—— The total alkaloids extracted from *Lycoris radiata* was dissolved in acetic acid, filtered, basified with K_2CO_3, and the resulting product was extracted with $CHCl_3$. Crystals of lycorine, which is very slightly soluble in $CHCl_3$, formed a third layer between $CHCl_3$ and H_2O and were collected on a filter. The $CHCl_3$ layer was washed with aqueous NaOH to remove phenolic alkaloids and then extracted with dilute HCl. The acidic aqueous layer was brought to pH 6.6 with Na_2CO_3 and the solution was extracted with EtOAc to give fraction 1 which gave on concentration tazettine as the main product. The pH value of the aqueous layer was then increased by 0.4 and the solution was again extracted with the same solvent to give fraction 2, from which hippeastrine, haemanthidine and a further crop of tazettine were isolated after chromatography. Fraction 3 involved the EtOAc extract from the aqueous layer adjusted to pH 7.4 and afforded on concentration lycorenine. The mother liquor from the crystallization of lycorenine was chromatographed in benzene on alumina. Benzene and benzene-$CHCl_3$ eluates gave galanthamine, lycoramine, and lycorenine. Further elution with $CHCl_3$ yielded haemanthamine as prisms, m.p. 203~205° after crystallization from acetone, $[\alpha]_D^{24}$ +43°(c=1.1, $CHCl_3$). *Anal.* Calcd. for $C_{17}H_{19}O_4N$: C, 67.76; H, 6.36; N, 4.65; O, 21.24; OCH_3, 10.32. Found : C, 67.97; H, 6.36; N, 5.07; O, 21.52; OCH_3, 9.91. The base did not depress the melting point of an authentic sample of haemanthamine, m.p. 203~205°, $[\alpha]_D$ +42° ($CHCl_3$) and the IR spectra of the two samples in Nujol mull were superimposable.

The picrate formed yellow scales on crystallization from acetone m.p. 224~226° (decomp.). *Anal.* Calcd. for $C_{17}H_{19}O_4N \cdot C_6H_3O_7N$: C, 52.09; H, 4.19; N, 10.57. Found : C, 52.22; H, 4.33; N, 10.66.

The methiodide crystallized from acetone-MeOH in needles, m.p. 191~193°. *Anal.* Calcd. for $C_{17}H_{19}O_4N \cdot CH_3I \cdot 1/2H_2O$: C, 47.80; H, 5.13; N, 3.10. Found : C, 47.69; H, 5.22; N, 3.48.

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