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The Alkaloidal Constituents of Goda-Manel (*Crinum zeylanicum* L.), a Sri Lankan Folk Medicine¹⁾

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From the bulbs of an Amaryllidaceous plant, *Crinum zeylanicum* L., a Sri Lankan folk medicine (Sinhalese name, Goda-manel), five alkaloids I—V were isolated and identified as lycorine (I), 3-acetylhamayne (III), 6-hydroxycrinamine (IV), hamayne (V), and a new alkaloid, 6-methoxycrinamine (II).

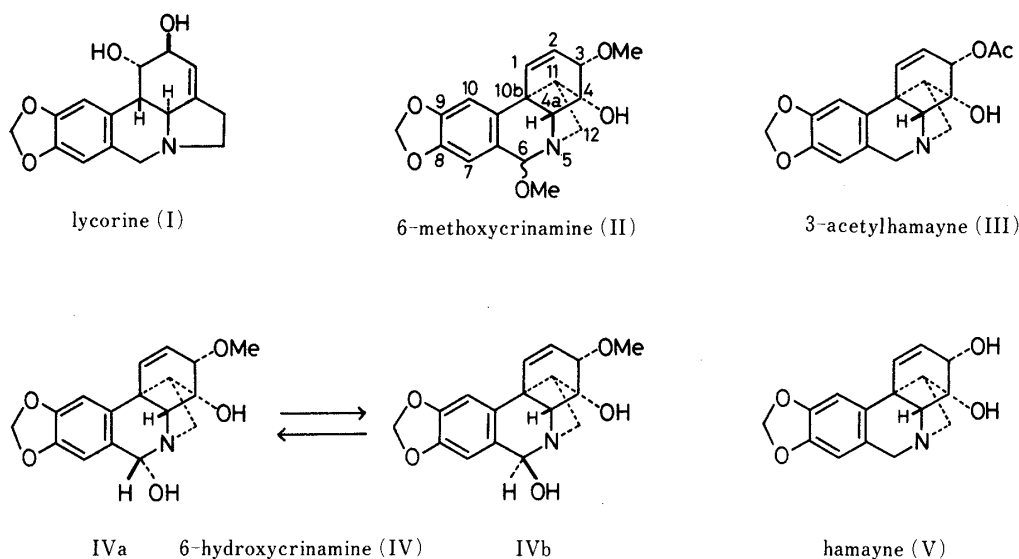
Keywords—*Crinum zeylanicum*; Amaryllidaceae; alkaloid; folk medicine; lycorine; 6-hydroxycrinamine; 6-methoxycrinamine; 3-acetylhamayne; hamayne; acyl migration

A plant with the Sinhalese name Goda-manel is used in Sri Lanka as a folk medicine; the roasted bulbs are used as a rubifacient in rheumatism and the juice of the leaf as a remedy for earache.²⁾ It is applied to abscesses to promote suppuration and is an ingredient in decoctions for fever.³⁾ Some traditional medical practitioners in that country also use the bulbs for the treatment of snake-bite.⁴⁾ Jayaweera²⁾ incorrectly named Goda-manel as the Amaryllidaceous plant, *Crinum bulbispermum* BURM., while correctly describing the plant. *Crinum bulbispermum* is not indigenous to Sri Lanka and Goda-manel is *C. zeylanicum* L.⁵⁾ The only report of work on *C. zeylanicum* is a preliminary communication⁶⁾ in which doubt is expressed as to the correct identification of the species. Crinamine (0.1%), 6-hydroxycrinamine (0.24%), and lycorine (0.05%) were reported to be present, although the parts of the plant used are not disclosed.

Here we report on the alkaloidal constituents of the bulbs of this plant. The methanol extract of the bulbs (900 g) was fractionated as described in the experimental section to afford five alkaloids: compound I (195 mg), compound II (151 mg), compound III (83 mg), compound IV (725 mg), and compound V (236 mg).

Compound I was sparingly soluble in the usual organic solvents and had mp 258—260 °C (dec.). Its infrared (IR) spectrum showed an OH absorption at 3350 cm⁻¹, but no absorption due to a carbonyl group. Acetylation with acetic anhydride and pyridine gave a diacetate, mp 225—227 °C, which was identical with diacetyllycorine⁷⁾ (direct comparison). Therefore compound I was identified as lycorine (I).⁸⁾

Compound IV, the major alkaloid of the bulbs of this plant, crystallized as needles from CH₂Cl₂, mp 146—150 °C, with [α]_D +42° (CHCl₃). The formula C₁₇H₁₉NO₅ was established by the high resolution mass spectrum (MS). Although IV was homogeneous in chromatography, its ¹H-nuclear magnetic resonance (¹H-NMR) spectrum (in CDCl₃) was rather complex, suggesting that it was a 3:1 mixture of two isomers epimeric at a hydroxyl group in a benzylic position. Since we have encountered such a phenomenon in the total synthesis of haemanthidine and 6-hydroxycrinamine,⁹⁾ we immediately recognized this as characteristic of the 6-hydroxycrinane system due to the two stereoisomers of the compound being in equilibrium, as shown in Chart 1. The ¹H-NMR and IR spectra of IV and those of 6-

Chart 1. Alkaloids of Goda-Manel (*Crinum zeylanicum* L.)

hydroxycrinamine¹⁰⁾ were superimposable, confirming that compound IV is 6-hydroxycrinamine (IV).⁶⁾

Compound II was obtained as an oil. It formed a crystalline picrate, mp 205—207°C, which was analyzed as $C_{18}H_{21}NO_5 \cdot C_6H_3N_3O_7$. Comparison of the 1H -NMR spectrum of the free base with those of crinamine (VI) and 6-hydroxycrinamine (IV) suggested that compound II is 6-methoxycrinamine (II), since it contained an extra OCH_3 at δ 3.57 together with a benzylic proton at δ 4.39 as a singlet. This assignment was supported by the ^{13}C -NMR spectrum of the base and confirmed by partial synthesis of the base from 6-hydroxycrinamine (IV) (see Experimental). This is the first report of the isolation of 6-methoxycrinamine (II). The configuration of the 6-methoxyl group was suggested to be β (*trans* to the ethanamine bridge), but was not definitely established, although compound II was stereochemically homogeneous.

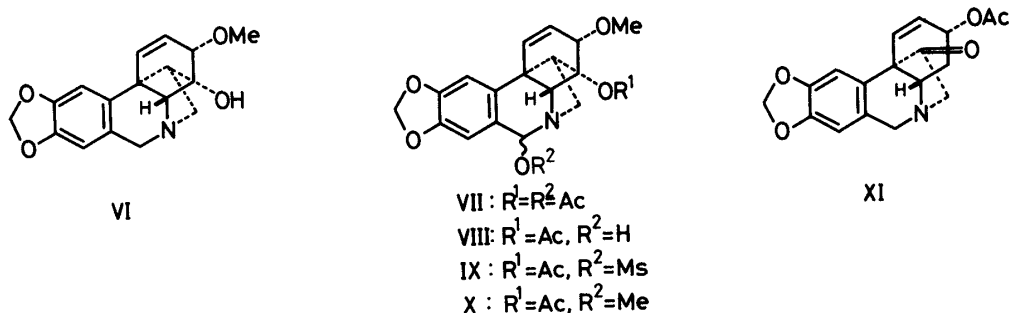


Chart 2

Compound III crystallized as prisms from benzene, mp 115—116°C, and had the formula $C_{18}H_{19}NO_5$. Its IR and 1H -NMR spectra indicated the presence of an acetoxy group (1735 cm^{-1} and δ 2.09). Oxidation of compound III with dimethylsulfoxide-acetic anhydride gave a product exhibiting an IR absorption at 1745 cm^{-1} indicative of a five-membered ring ketone. The above evidence together with the spectral resemblance between compound III and crinamine (VI) suggested III to be 3-acetyldemethylcrinamine (3-acetylhamayne). The

^1H -NMR spectrum of compound III was superimposable on that of 3-acetylhamayne.¹¹⁾

Compound V was obtained as an amorphous powder from ethyl acetate. It formed a crystalline diacetate, mp 119–121 °C, $\text{C}_{20}\text{H}_{21}\text{NO}_6$, on acetylation with acetic anhydride and pyridine. The ^1H -NMR spectrum of the diacetate suggested it to be diacetyldemethylcrinamine (XII). Compound V and its diacetate therefore must be identical with hamayne¹²⁾ and diacetylhamayne,¹²⁾ respectively. However, the reported physical data for hamayne and diacetylhamayne differ markedly from those of compound V and its diacetate (Table I). In order to definitively prove the structure, partial hydrolysis of the diacetate was carried out. Treatment of the diacetate with sodium bicarbonate in methanol furnished, in good yield, monoacetate which was identical with 3-acetylhamayne (III). Consequently, compound V was concluded to be demethylcrinamine (V) (hamayne). The discrepancies in the physical data of compound V and the diacetate when compared with those reported for hamayne and

TABLE I. Physical Data for Compound V and Hamayne

Compound V	Hamayne ^{a)}
Free base	
Amorphous from EtOAc	mp 79–80 °C (plates from acetone) [α] _D ²⁵ +43° (EtOH) $\text{C}_{16}\text{H}_{17}\text{NO}_4 \cdot 2/3\text{H}_2\text{O}$ IR ν : 3400 (OH) (KBr)
IR ν : 3350 (OH) (CHCl_3)	
Diacetate	
mp 119–121 °C (needles from MeOH) $\text{C}_{20}\text{H}_{21}\text{NO}_6$ IR ν : 1738 (CO) (KBr) NMR δ : 2.04, 2.09 (each 3H, s) $2 \times \text{OAc}$ 3.69, 4.35 (each 1H, d, $J=16$ Hz) Ar-CH ₂ -N < 4.97 (1H, t, $J=5$ Hz) >CH-OAc 5.45 (1H, m) >CH-OAc 5.87, 6.24 (each 1H, dd, $J=11, 2$ Hz) -CH=CH- 5.90 (2H, s) -OCH ₂ O- 6.47, 6.86 (each 1H, s) Ar-H	mp 77.5–78 °C (needles from MeOH) $\text{C}_{20}\text{H}_{21}\text{NO}_6$ IR ν : 1730 (CO) (Nujol) NMR δ : 2.01, 2.07 (each 3H, s) $2 \times \text{OAc}$ 3.66, 4.32 (each 1H, d, $J=18$ Hz) Ar-CH ₂ -N < 4.95 (1H, t, $J=6$ Hz) >CH-OAc 5.44 (1H, m) >CH-OAc 5.87, 6.21 (each 1H, dd, $J=10, 2$ Hz) -CH=CH- 5.89 (2H, s) -OCH ₂ O- 6.46, 6.84 (each 1H, s) Ar-H

a) See ref. 12.

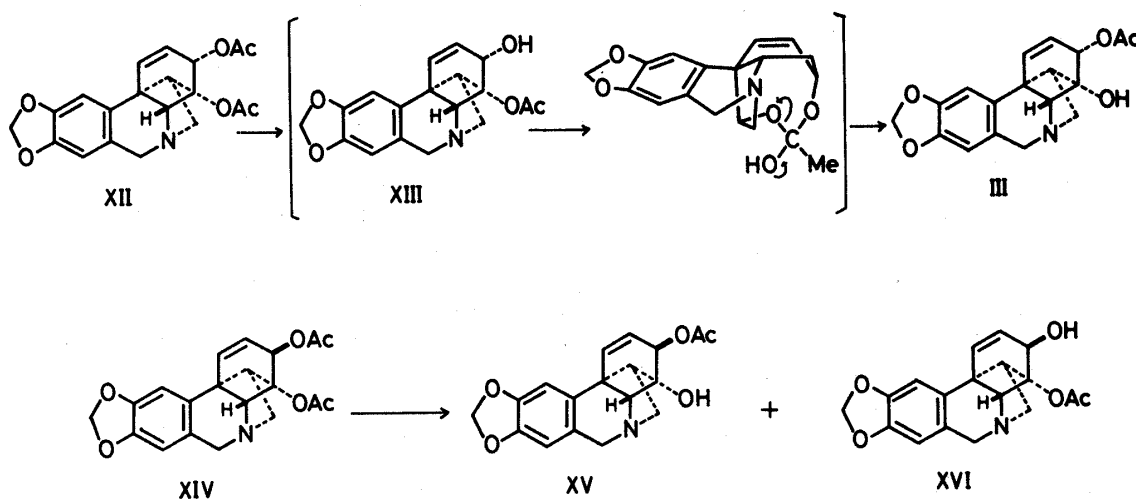


Chart 3

diacetylhamayne, respectively, may be due to polymorphism in these compounds.

A comment appears to be necessary regarding the formation of 3-acetylhamayne (III) from diacetylhamayne (XII), since this transformation involves the regio-selective hydrolysis of the more hindered 11-acetoxyl group. We consider this unexpected result as being due to acetyl migration from the initially formed 11-monoacetate (XIII) to the 3-acetate (III) as shown in Chart 3. Similar hydrolysis of the stereoisomeric diacetyl-11-hydroxyvittatine (XIV), in which the 3-acetoxyl group is in a quasi-axial orientation and is therefore less hydrolyzable than in XII, is known to give a mixture of the 3-acetate (XV) and the 11-acetate (XVI);¹³⁾ an intramolecular acyl migration of the 11-acetyl group to the 3-position in the latter compound is impossible.

Experimental

Unless otherwise stated, the following procedures were adopted. Melting points were determined on a Yanagimoto micro hot-stage apparatus, and are uncorrected. IR spectra were taken in KBr disks and are given in cm^{-1} . $^1\text{H-NMR}$ (100 MHz) and $^{13}\text{C-nuclear magnetic resonance}$ ($^{13}\text{C-NMR}$) (25.0 MHz) were measured in CDCl_3 solution with tetramethylsilane as an internal standard on a JEOL FX-100 spectrometer and are given in δ . High-resolution mass spectra were taken with a Hitachi M-80 machine. Wakogel C-200 (silica gel) was used for column chromatography. For thin layer chromatography (TLC), Merck precoated plates GF₂₅₄ were used and spots were observed by spraying Dragendorff reagent or 1% ceric sulfate in 10% H_2SO_4 followed by heating at 100 °C until coloration appeared.

Extraction of the Alkaloids from Goda-Manel—The plant Goda-manel was collected in the suburbs of Polonnaruwa in the North Central Province of Sri Lanka in January 1982. The bulbs were chopped and half-dried, and the total dried material (900 g) was extracted with methanol for 72 h in a Soxhlet apparatus. Concentration of the extract left a syrup (90 g) which was partitioned 3 times between 3% H_2SO_4 (100 ml) and CHCl_3 (100 ml). The water layer was basified with solid K_2CO_3 , then extracted 10 times with $\text{CHCl}_3\text{-MeOH}$ (10:1) (250 ml). The combined organic layer, without washing, was concentrated to give a crude alkaloid fraction (4.5 g) which contained some inorganic salts. This fraction was digested 3 times with hot CHCl_3 (50 ml) to divide it into soluble and insoluble fractions. The CHCl_3 -soluble fraction, on storage at room temperature deposited colorless prisms (compound I) (140 mg). The mother liquor was concentrated to leave a gummy residue (2.5 g), which was chromatographed on silica gel to give the following crude alkaloids: compound II (0.3 g) and compound III (0.3 g) from the $\text{CHCl}_3\text{-MeOH}$ (20:1) eluate, compound IV (1.2 g) from the $\text{CHCl}_3\text{-MeOH}$ (20:1.5) eluate, and compound V (0.5 g) from the $\text{CHCl}_3\text{-MeOH}$ (5:1) eluate. These products were purified by rechromatography on neutral alumina to yield the alkaloids in pure forms: II (151 mg), III (83 mg), IV (725 mg), and V (236 mg).

The CHCl_3 -insoluble fraction (2.0 g) was digested with hot $\text{CHCl}_3\text{-MeOH}$ (1:1) (3×50 ml) and the extract, without concentration, was passed through a short column of silica gel, then concentrated to yield a further crop of compound I (55 mg). The $\text{CHCl}_3\text{-MeOH}$ insoluble residue was acetylated with Ac_2O and pyridine (18 h, r.t.) and the product was chromatographed on silica gel to give a neutral compound (oil, 1.5 g) from the $\text{CHCl}_3\text{-MeOH}$ (20:1) eluate and diacetyllycorine (30 mg) from the $\text{CHCl}_3\text{-MeOH}$ (5:1) eluate.

Lycorine (Compound I) (I)—Colorless prisms, mp 258—260 °C (dec.). [lit. mp 266—267 °C (dec.).]⁸⁾ IR: 3350 (OH). Acetylation of this with Ac_2O -pyridine (overnight, r.t.) gave the diacetate, mp 225—227 °C, as leaflets from MeOH. IR: 1725 (CO). $^1\text{H-NMR}$: 1.90, 2.03 (each 3H, s, $2 \times \text{OAc}$), 3.49, 4.14 (each 1H, d, $J=15$ Hz, $\text{ArCH}_2\text{N}<$), 5.30, 5.32 (each 1H, m, $2 \times >\text{CHOAc}$), 5.50 (1H, m, $-\text{CH}=\text{C}<$), 5.90 (2H, s, OCH_2O), 6.55, 6.74 (each 1H, s, ArH). This was identical with an authentic sample of diacetyllycorine (lit. mp 215—216 °C)⁷⁾ on the basis of $^1\text{H-NMR}$, IR, and TLC comparisons.

6-Methoxycrinamine (Compound II) (II)—Oil, IR (CHCl_3): 3450 (OH). $^1\text{H-NMR}$: 3.40, 3.57 (each 3H, s, $2 \times \text{OMe}$), 4.39 (1H, s, $>\text{NCHOMe}$), 5.89 (2H, ABq, $\Delta\delta=0.05$ ppm, $J=1.2$ Hz, OCH_2O), 6.18 (2H, s, $-\text{CH}=\text{CH}-$), 6.72, 6.73 (each 1H, s, ArH). $^{13}\text{C-NMR}$: 146.2 s, 136.9 s, 136.0 s, 126.9 s, 109.5 d, 102.7 d (aromatic ring), 136.0 d, 123.3 d (C1, C2), 101.0 t (OCH_2O), 78.5 d (C3), 28.9 t (C4), 50.3 s (C10b), 76.2 d (C11), 95.9 d (C6), 60.0 d (C4a), 58.5 t (C12), 55.8 q, 56.3 q (OMe). The picrate crystallized as yellow needles from EtOAc, mp 205—207 °C. Anal. Calcd for $\text{C}_{18}\text{H}_{21}\text{NO}_5 \cdot \text{C}_6\text{H}_3\text{N}_3\text{O}_7$, C, 51.43; H, 4.32; N, 10.00. Found: C, 51.49; H, 4.17; N, 10.29.

3-Acetylhamayne (Compound III) (III)—Colorless prisms from benzene, mp 115—116 °C, $[\alpha]_D^{20} = +126^\circ$ ($c=0.5$, CHCl_3). High-resolution MS: Calcd for $\text{C}_{18}\text{H}_{19}\text{NO}_5$: 329.1262. Found: 329.1266. IR: 3250 (OH), 1735 (CO). $^1\text{H-NMR}$: 2.09 (3H, s, OAc), 2.36 (1H, s, OH, disappeared on addition of D_2O), 3.68, 4.33 (each 1H, d, $J=17$ Hz, $\text{ArCH}_2\text{N}<$), 4.00 (1H, t, $J=5$ Hz, $>\text{CHOH}$), 5.45 (1H, m, $>\text{CHOAc}$), 5.90 (2H, s, OCH_2O), 6.07 (1H, dd, $J=11$ and 1.5 Hz) and 6.33 (1H, dd, $J=11$ and 2 Hz) ($-\text{CH}=\text{CH}-$), 6.48, 6.79 (each 1H, s, ArH). The $^1\text{H-NMR}$ spectrum was superimposable on that of 3-acetylhamayne.¹¹⁾

6-Hydroxycrinamine (Compound IV) (IV)—Colorless needles from CH_2Cl_2 , mp 146—150 °C, $[\alpha]_D^{20} = +42^\circ$

($c=0.5$, CHCl_3). High-resolution MS: Calcd for $\text{C}_{17}\text{H}_{19}\text{NO}_5$: 317.1262. Found: 317.1268. IR: 3450 (OH). $^1\text{H-NMR}$ 3.39 (3H, s, OMe), 5.02 (0.75H, s, $>\text{CHOH}$), 5.59 (0.25H, s, $>\text{CHOH}$), 5.92 (2H, s, OCH_2O), 6.20 (1.5H, s, $-\text{CH}=\text{CH}-$), 6.18 (0.5H, s, $-\text{CH}=\text{CH}-$), 6.76, 6.82 (each 0.75H, s, ArH), 6.73, 6.97 (each 0.25H, s, ArH). The IR and $^1\text{H-NMR}$ spectra were superimposable on those of 6-hydroxycrinamine¹⁰⁾ [lit. mp 135–140 °C, $[\alpha]_D^{25} = +46^\circ$ (CHCl_3)].⁶⁾

Demethylcrinamine (Compound V) (Hamayne) (V)—Physical data, see Table I. Hamayne (30 mg) was acetylated with Ac_2O (1 ml) and pyridine (2 ml) for 18 h at room temperature to yield diacetylhamayne (XII) (31 mg), mp 119–121 °C. *Anal.* Calcd for $\text{C}_{20}\text{H}_{21}\text{NO}_6$: C, 64.68; H, 5.70; N, 3.77. Found: C, 64.44; H, 5.67; N, 3.99. See also Table I.

Transformation of 6-Hydroxycrinamine (IV) to 6-Methoxycrinamine (II)—Compound IV (30 mg) was acetylated with Ac_2O (1 ml) and pyridine (2 ml) for 18 h at room temperature and worked up as usual to give the diacetate (VII) (36 mg) which was a 3 : 1 mixture of two compounds as shown by the $^1\text{H-NMR}$ spectrum [δ 1.97 (3H, s, OAc), 2.08, 2.15 (each s, OAc, 3 : 1)]. This mixture was treated with 2.5% NaHCO_3 -MeOH (6 ml) for 5 h at 40 °C. Dilution of the mixture with water, extraction with CHCl_3 , and chromatography of the concentrate on silica gel with EtOAc gave the monoacetate (VIII) (15 mg), which was a 2 : 1 mixture of epimers of the hydroxyl group at C-6 [δ 2.00 (3H, s, OAc), 4.93, 5.53 (each s, $>\text{CHOH}$, 2 : 1)]. Compound VIII was mesylated with methanesulfonyl chloride (5 drops) and pyridine (1 ml) for 3 h at room temperature and the product (IX) obtained after usual work-up was stirred in MeOH at room temperature. After 3 d, the mixture gave two spots on TLC corresponding to the mesylate (IX) and a more mobile one (X). The latter spot was collected by preparative TLC and hydrolyzed with 5% K_2CO_3 -MeOH for 1 h at 60 °C. The resulting compound gave a spot on TLC identical with that of compound II. This gave the picrate, mp and mixed mp 205–207 °C.

DMSO- Ac_2O Oxidation of 3-Acetylhamayne (III)—Compound III (5 mg) in DMSO (1 ml) and Ac_2O (0.5 ml) was stirred for 18 h at room temperature. The mixture was poured into water, and extracted with CHCl_3 . The extract was washed with water, dried, and concentrated to give a residue (XI) (2 mg), which exhibited an IR absorption band (in CHCl_3) at 1745 and a shoulder at 1720 cm^{-1} , but no OH group absorption.

Partial Hydrolysis of Diacetylhamayne (XII) to 3-Acetylhamayne (III)—Diacetylhamayne (XII) (31 mg) was treated with 2.5% NaHCO_3 -MeOH (6 ml) for 6 h at 40 °C. Chromatography of the product on silica gel gave, from the CHCl_3 -MeOH (30 : 1) eluate, 3-acetylhamayne (III) (15 mg). The identification was corroborated by $^1\text{H-NMR}$, IR, and TLC comparisons.

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References and Notes

- 1) a) This paper constitutes a part of the series "Studies on Medicinal Resources in Sri Lanka"; b) A part of this work was presented at the 56th Hokuriku Branch Meeting of the Pharmaceutical Society of Japan, June 1982.
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