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NEW ERYTHRINA ALKALOIDS OF *COCCULUS LAURIFOLIA*

D. S. BHAKUNI and H. UPRETY
Central Drug Research Institute, Lucknow, India

and

D. A. WIDDOWSON
Department of Chemistry, Imperial College, London SW7 2AY, England

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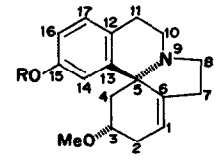
Key Word Index—*Cocculus laurifolia*; Menispermaceae; new *Erythrina* alkaloids.

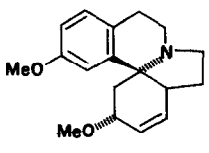
Abstract—Four abnormal *Erythrina* alkaloids have been isolated from the leaves of *Cocculus laurifolia*. Of these, two new bases, isococculidine and coccoline, have been assigned the structures (3*S*,5*S*)-3,15-dimethoxyerythrin-l-ene and (3*R*,5*S*)-15-hydroxy-3-methoxyerythrin-l-en-8-one respectively.

INTRODUCTION

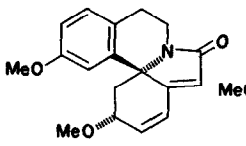
A programme of screening of Indian plants over a wide range of biological activities led to the observation [1] of hypotensive activity in the 50% aqueous ethanolic extract of the leaves of *Cocculus laurifolia*, a shrub from northern India. A search for the active principle(s) from the alkaloidal fraction resulted in the isolation of three new dibenz [d,f] azonine bases, laurifine, laurifonine and laurifinine [2]. In addition, four *Erythrina* alkaloids, cocculidine (**1**), cocculine (**2**), isococculidine (**3**) and coccoline (**4**) were isolated. A number of bisbenzylisoquinoline [3,4], aporphine [4,5] and *Erythrina* alkaloids [6–9] have been reported previously from this and related plants and the structures of the dibenz [d,f] azonine alkaloids have been published [2]. An X-ray study [10] has established the structures of cocculidine (**1**) and cocculine (**2**). We now report the structures of isococculidine (**3**) and coccoline (**4**).



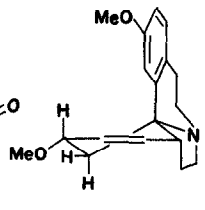
(**1**) R = H
(**2**) R = Me



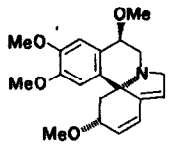
(**3**)



(**4**)



(**5**)

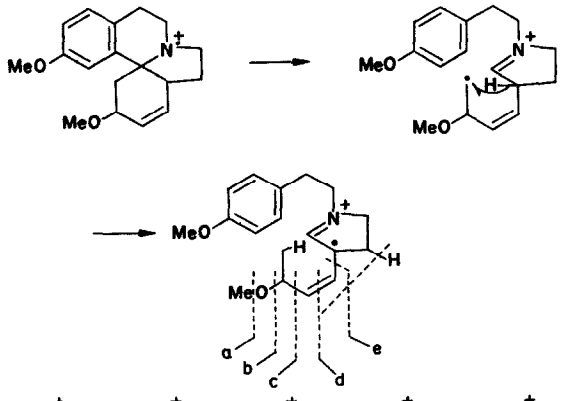


(**6**)

RESULTS AND DISCUSSION

The four *Erythrina* alkaloids (**1**–**4**) were isolated by column chromatography, The ether soluble portion of the total alkaloidal mixture was divided into phenolic and non-phenolic bases. The non-phenolics were chromatographed on neutral alumina. Elution with benzene–hexane (1:1) and benzene–hexane (2:1) gave isococculidine (**3**) mp 95–96°, $[\alpha]_D$ + 124°. The structural assignment as **3** follows from the physical data. The compound analysed for $C_{18}H_{23}NO_2$ and showed λ_{max} 230 and 280 nm, and an IR spectrum which indicated the absence of hydroxyl or ketonic function. The MS had M^+ at *m/e* 285 in agreement with the microanalysis and a fragmentation pattern very similar to that of a 1,6-diene type of *Erythrina* alkaloid [11]. In particular there was an intense peak at *m/e* 254 with only low intensity fragments at *m/e* 226, 212, 200, corresponding to successive fragmentations of a 1(**2**) unsaturated ring D with the accompanying loss in this case of additional hydrogen atoms. A rationalisation, based on established precedent [11], is given in Scheme 1.

The NMR spectrum confirmed this assignment and enabled the relative stereochemistry to be established. The spectrum consisted of 2 methoxyl resonances (τ 6.29, 6.78), olefinic proton multiplets (τ 3.94 and 4.15), three aromatic proton resonances (τ 3.27, 3.31 and 2.90) and the methylene multiplets. Double resonance experiments gave the coupling constants for the ring D protons as



(a) M^+ - 15; (b) M^+ - 31; (c) M^+ - 59; (d) M^+ - 73; (e) M^+ - 85

given in Table 1. Erythristemine (**6**), the X-ray structure of which has been determined [12], is shown for comparison. Assuming the basic erythrinane structure the coupling of the proton α—to the methoxy group (H-3), to an olefinic proton fixed the position of the double bond as 1(**2**) and the magnitude ($J_{2,3} \sim 1.5$ Hz) indicated the 4-axial conformation of H-3. The methoxy group must therefore be ψ-equatorial as in the previously characterised alkaloids [13,14]. The coupling of H-3 to H-4a (10Hz) and H-4e (6Hz) confirmed this assignment and showed the C3-C4 bond to be in a gauche conformation. A value for $J_{1,6}$ of 3.5–4 Hz indicated a ψ-equatorial orientation for H-6. The only conformation for ring D which accommodates these facts is the half chair and the stereostructure must be as in **5**. The position of the aromatic methoxy-group also follows from the NMR spectrum. The proton at lowest field (*m*- to oxygen) appeared as one half of an AB quartet with additional multiplicity arising from benzylic (and *para*-) coupling. This proton therefore is H-17 and the methoxy group must be at 15. The absolute stereochemistry was not determined* but may reasonably be assumed to be as in all other *Erythrina* alkaloids [10,12,15] including those which co-occur in *Cocculus laurifolia*.

Elution of the alumina column with chloroform gave cocculidine (**1**) mp 86–87° $[\alpha]_D$ +260°, spectroscopically identical to the reported material [6,7].

The phenolic bases were chromatographed over neutral alumina and eluted with benzene–ethyl acetate–methanol. Benzene–ethyl acetate (1:1) eluted coccoline, (**4**), mp 245–246°, $[\alpha]_D$ +233°. The alkaloid analysed for $C_{17}H_{17}NO_3$. The MS (M^+ 283) agreed with this. Furthermore, the simplicity of the fragmentation pattern, with M^+ 100% and the loss of methyl and methoxy groups dominant, again suggested a 1,6-diene type [11]. The IR spectrum showed ν_{max} 3300, 1665 cm^{-1} and the UV spectrum, λ_{max} 284 infl. 256 and 230 nm. The 256 peak is not normally present in *Erythrina* alkaloids and taken with the 1665 cm^{-1} band in the IR spectrum suggested a dienone system. The NMR spectrum showed three aromatic protons. Of these a low field doublet (*m*- to oxygen) (τ 3.04) was clear of the others. There were signals due to three olefinic protons: one at low field (τ 3.42) comprising the 'B' component of an ABX system, one at τ 3.85, comprising the 'A' component of the same system, and one at τ 4.16 (singlet). The methoxyl group was at τ 6.74.

Double resonance experiments (Table 1) enabled these data to be interpreted fully. Irradiation at τ 6.50 (α-to-oxygen) caused a collapse of the small (2 Hz) splitting of the olefinic protons at τ 3.42 leaving the AB system (*J* 10 Hz) of the two lower field olefinic protons. This implies a *cis* orientation of the double bond. The irradiation also sharpened the signal of the 'A' part of the system (τ 3.85) indicating 0.5 Hz allylic coupling. These results are accommodated by the 3-methoxy-1,6-diene system of the *Erythrina* alkaloids. However, the lack of fine structure on the remaining olefinic proton the low field of the double doublets as well as the IR and UV spectra require that the carbonyl group be placed at C-8. The coupling constant between H-3 and H-2 (2 Hz) is comparable to that of other *Erythrina* alkaloids (Table 1) and implies a 3-ψ-equatorial conformation for the methoxyl group.

Table 1. NMR coupling constants (Hz) of ring D of Δ^1 [2] *Erythrina* alkaloids

J	Isococulidine (3)	Coccoline (4)	Erythristemine (6)
1,2	10.5	10	10
2,3	1.5	2	2
3,4a	10	nd*	10.5
3,4e	6	nd	5.5
4a,4e	12	nd	10.5
1,3	1.5	0.5	nd
1,6	3.5	—	—
2,6	~1.5	—	—

* nd = not determined.

The remaining ambiguity is the position of the phenolic group. As above, the occurrence of a low field doublet due to an aromatic proton (τ 3.04) requires C-15 or C-16 substitution. Irradiation at τ 7.09 (benzylic region) caused this low field doublet to sharpen and had no effect on the other aromatic protons. Irradiation >10 Hz either side of τ 7.09 had no effect. It follows that the hydroxyl group is at C-15.

On the basis of the above data the structure **4** was assigned to coccoline. Again the absolute stereochemistry was not determined but is assigned on the basis of that determined for cocculidine [10].

The presence of the carbonyl function at C-8 has not previously been recorded in the natural alkaloids. Also it is known [16] that autoxidation of the 1,6-dienes gives rise to such functionality and it is possible that coccoline may be an artefact of the drying process.

EXPERIMENTAL

Extraction. Air dried leaves of *Cocculus laurifolia* DC (29 kg) which were collected in September 1971 from Dehra Dun, India, were extracted with EtOH. The solvent was removed *in vacuo* and the dark green viscous mass was extracted with 5% HCl. The aq. acidic soln was defatted with petrol and then basified with aq. $NaHCO_3$. The liberated bases were extracted with $CHCl_3$. The solvent was evap under red press to give the alkaloidal mixture which was divided into Et_2O soluble and Et_2O insoluble fractions. The Et_2O soluble material was further separated into phenolic and non-phenolic bases.

Isococculidine (**3**). The mixture of non-phenolic bases (5 g) were chromatographed on neutral alumina (200 g). The column was eluted with hexane, hexane–C_6H_6 and $CHCl_3$ mixtures. Fractions of 25 ml were taken and the elution monitored by TLC. Elution with C_6H_6–hexane (1:1) and (1:2) afforded isococculidine (**3**), (1 g), mp 95–96° (from C_6H_6–hexane) $[\alpha]_D$ + 124° (*c*, 1.2 in MeOH), methiodide mp 198–199° (from Et_2O–MeOH). The free base had ν_{max}^{KBr} cm^{-1}: 2904, 2784, 1603, 1470, 1241, 1104 and 882; λ_{max}^{MeOH} nm: 230 and 280; NMR ($CDCl_3$): τ 6.78 (3H, *s*, 3-OMe), 6.29 (3H, *s*, 15-OMe), 3.94 (1H, *m*, H-1), 4.15 (1H, *m*, H-2), 3.31 (1H, *dd*, J_1 8Hz, J_2 2.5Hz, H-16), 3.27 (1H, *brs*, H-14), 2.90 (1H, *dd*, J_1 8 Hz, J_2 + $J_{benzylic}$ 3 Hz, H-17), 6.45 (1H, *m*, H-3), 7.23 (1H, *m*, H-6), 8.06 (1H, *dd*, J_1 12 Hz, J_2 6Hz, H-4e), 8.32 (1H, *dd*, J_1 12 Hz, J_2 10 Hz, H-4a); MS: M^+ at *m/e* 285 with significant peaks at *m/e* 270, 251, 241, 240, 226, 212, 200. (Found: C, 75.80; H, 7.92; N, 4.90. $C_{18}H_{23}NO_2$ requires C, 75.79; H, 8.07; N, 4.91%). Elution of the column with $CHCl_3$ gave cocculidine (**1**), (18 g), mp 86–87°, $[\alpha]_D$ +260° (lit. [3] mp 86–87°, $[\alpha]_D$ + 250.9°), spectroscopically identical in all respects with the reported material.

*A correlation between cocculidine and isococculidine via the possible identity of their dihydro-derivatives was not possible because of the resistance of cocculidine to hydrogenation.

Coccoline (**4**). The mixture of phenolic bases (3 g) was chromatographed on neutral alumina (250 g). Elution with C_6H_6–EtAc (1:1) and EtAc furnished coccoline (**4**), (200 mg), mp 245–246° (from EtAc) $[\alpha]_D$ + 233° (*c*, 1.08 in MeOH), λ_{max} nm: 231 and 258, ν_{max}^{KBr} cm^{-1}: 2900, 1665, 1500, 1455, 1270, and 1235, NMR ($CDCl_3$): τ 3.04 (1H, *dd*, *J* 8 Hz, H-17), 3.34 (2H, *m*, H-14 and H-16), 3.44 (1H, *dd*, J_1 10 Hz, J_2 2 Hz, H-2), 3.86 (1H, *dd*, J_1 10 Hz, J_2 0.5 Hz, H-1) 4.14, (1H, *s*, H-7), 6.76 (3H, *s*, OMe). The MS had M^+ at *m/e* 283 with significant peaks at *m/e* 268, 252, 240, 222, 210 and 181. (Found: C, 72.20; H, 6.35; N, 4.91; $C_{17}H_{17}NO_3$ requires C, 72.07; H, 6.05; N, 4.94%). Elution with EtAc–MeOH (19:1) gave cocculine (**2**), (60 mg) mp 220–221°, (from EtAc), $[\alpha]_D$ + 252° (*c*, 1.32 in MeOH), (lit. [3] mp 217–218°, $[\alpha]_D$ + 271.7°). This material was spectroscopically identical in all respects with the reported compound.

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