Rutaceous Constitutents. Part II.[1] Two Acridone Alkaloids from *Atalantia ceylanica* (Rutaceae)

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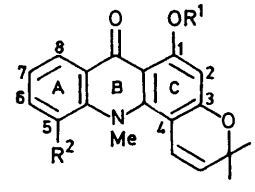
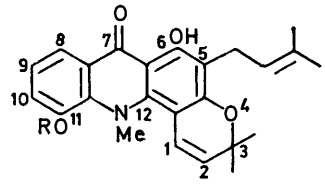
Two acronycine analogues, 3,12-dihydro-6,11-dihydroxy-3,3,12-trimethylpyrano[2,3-*c*]acridin-7-one and its 5-(3-methylbut-2-enyl) derivative, have been isolated from *Atalantia ceylanica* (Rutaceae).

In 1956, Acheson reviewed the 13 alkaloids derived from acridine.[2] Since then, acronycine has been evaluated for its antitumour properties *in vitro*,[3] and *in vivo* and clinical trials are now proceeding.[4] About 13 more alkaloids have been reviewed,[5] and recently 5-hydroxy-9-acridones have been isolated;[6,7] we now report the isolation of two more such alkaloids from *Atalantia ceylanica*.

From the wood of *A. ceylanica*, two major alkaloids (A) and (B) were isolated in 0·04 and 0·003% yield, respectively. Both showed the typical u.v. spectra associated with a 9-acridone nucleus, and in the presence of sodium methoxide or aluminium chloride bathochromic shifts were observed indicating the presence of hydroxy-groups.[8] The i.r. absorption of (A), at 3250 (OH) and 1630 cm^{-1} (CO), indicated that at least one of the hydroxy-groups was hydrogen bonded. Accurate mass measurement gave the molecular formula as $C_{19}H_{17}NO_4$ and the n.m.r. spectrum showed an aromatic proton arrangement similar to that reported for acronycine;[9] the chemical shifts for the remaining protons could be assigned to a dimethylchromen system. The chemical shift for the aromatic proton associated with ring C more closely resembled that for H-2 † in noracronycine and the dimethylchromen ring was consequently assigned an angular orientation. Spin decoupling on the protons of ring A indicated that the remaining hydroxy-group was at C-5, and the alkaloid



(I) a; R^1 = H, R^2 = OH
b; R^1 = Me, R^2 = OMe
c; R^1 = H R^2 = OMe



(II) a; R = H
b; R = Me

was assigned the structure (Ia). The ring protons in the dimethyl ether (Ib) showed an even closer resemblance to those in acronycine, thus substantiating the assignment.

† Acridine numbering is used throughout the Discussion section [see formula (I)]. Systematic numbering is shown in formula (II).

[1] Part I, J. R. Lewis and A. W. Fraser, *Phytochemistry*, 1973, in the press.

[2] R. H. Acheson in 'The Chemistry of Heterocyclic Compounds,' ed. A. Weissberger, vol. 9, Interscience, New York, 1956.

[3] G. H. Svodoba, G. A. Poore, P. J. Simpson, and G. B. Boder, *J. Pharm. Sci.*, 1966, **55**, 758.

[4] J. R. Hartwell, National Cancer Institute, personal communication.

[5] Some of these are described in S. Johne and D. Gröger, *Die Pharmazie*, 1972, **4**, 195.

[6] K. H. Pogel and W. G. Wright, *J. Chem. Soc.* (*C*), 1969, 2327.

[7] T. R. Govindachari, N. Viswanthan, B. R. Pai, V. N. Ramachandran, and P. S. Subramaniam, *Tetrahedron*, 1970, **26**, 2905.

[8] J. Reisch, K. Szendrei, E. Minker, and I. Novak, *Die Pharmazie*, 1972, **4**, 208.

[9] C. S. Oh and C. V. Greco, *J. Heterocyclic Chem.*, 1970, **7**, 261.

The hydroxy-groups in (Ia) behaved differently towards methylating agents: diazomethane gave a monomethyl ether (Ic), the remaining hydroxy-group being still hydrogen bonded to the carbonyl group. The latter could however be methylated by dimethyl sulphate. A similar lack of reactivity of the *peri*-hydroxy-group has been observed with noracronycine.[9]

The mass spectrum of (A) showed a typical 9-acridone breakdown: loss of CH_3 from the dimethylchromen ring [10] followed by loss of CO from ring B; an alternative fragmentation involved the loss of HCO directly from ring B and was related to the presence of an *N*-methyl substituent.[11]

The isolation of compound (Ia) as a plant product is particularly interesting since it has been tentatively reported as one of the metabolites [12] isolated from rat bile after dosage with acronycine. Biogenetically, the introduction of the 5-hydroxy-group probably occurs at a late stage, perhaps *via* an arene oxide intermediate.[13]

Alkaloid (B) had the molecular formula $C_{24}H_{25}NO_4$, and the formation of a monomethyl ether and a diacetate indicated the presence of two hydroxy-groups, in environments similar to those in (Ia). Its n.m.r. spectrum showed the absence of a proton corresponding to H-2, and an additional series of resonances compatible with a dimethylallyl side chain. On this evidence structure (IIa) was formulated and the mass spectrum supported this assignment. Typical loss of CH_2 and CHO occurred but the dimethylallyl substituent at C-2 gave rise to two types of fragmentation, analogous to those reported for 2-methoxy-9-acridones,[11] involving loss of an isopentenyl radical, an isobutenyl radical, and an isopropyl radical. All these fragmentations are compatible with the presence of a dimethylallyl substituent attached directly to the aromatic ring C, and are analogous to published data for 2-substituted 9-acridones [11] and for the two related 5-hydroxy-acridone alkaloids, atalaphylline, and its *N*-methyl ether.[7]

EXPERIMENTAL

I.r. spectra were recorded for KBr discs. N.m.r. spectra were recorded on a Varian HA 100 spectrophotometer. All chromatography was carried out on silica gel (Merck Kieselgel PF).

Isolation.—The bark of *Atalantia ceylanica* (800 g) was extracted at room temperature with ethanol (4 l) for 3 weeks. Filtration and evaporation of the solution left a brown syrup which was partitioned between chloroform and water. The chloroform solution was separated, dried ($MgSO_4$), filtered, and evaporated to give a black syrup (15·5 g). T.l.c. showed the presence of four compounds, two with relatively high R_F and two with low R_F (on silica gel developed with chloroform). The two pairs were separated by preparative t.l.c. (same system). The material having high R_F was further separated by preparative t.l.c., first on silica gel developed with diethyl ether–light petroleum (b.p. 30—40°) (2 : 3) and then on silica gel developed with benzene–ethyl acetate (9 : 1), to give two alkaloids, (A) and (B), R_F 0·34 and 0·40.

Alkaloid (A), 3,12-Dihydro-6,11-dihydroxy-3,3,12-trimethylpyrano[2,3-c]acrid-7-one (Ia).—Crystallisation of alkaloid (A) from diethyl ether–light petroleum (b.p. 60—80°) gave red *needles* (319 mg, 0·04%), m.p. 252—254°, R_F (benzene–ethyl acetate, 9 : 1) 0·34, R_F [diethyl ether–light petroleum (b.p. 30—40°)] 0·35, $\lambda_{max.}$ (EtOH) 238 (log ε 4·24), 267 (4·62), 284 (4·61), 293sh (4·57), 325 (4·27), 341sh (4·07), and 424 nm (3·73), $\lambda_{max.}$ (EtOH–NaOMe) 237, 281, 293sh, 343, and 456 nm, $\lambda_{max.}$ (EtOH–$AlCl_3$) 272sh, 276, 293, 304, 330sh, 365, and 480 nm, $\nu_{max.}$ 3250 (OH), and 1630 cm^{-1} (CO), δ($CDCl_3$–5% CD_3OD) 1·51 (s), 3·80 (s), 5·45 (d), 6·13 (s), 6·69 (d), 7·12 (s), 7·17 (s), and 7·82 (t), δ [$(CD_3)_2CO$] 1·49 (s), 3·83 (s), 5·68 (d), 6·11 (s), 6·72 (d), 7·22—7·32 (complex), 7·78 (dd), and 14·20 (s) (Found: C, 70·3; H, 5·4. $C_{19}H_{17}NO_4$ requires C, 70·6; H, 5·3%) *m/e* 324 (11%), 323 (43), 322 (10), 309 (26), 308 (100), 294 (12), 293 (60), 280 (7), 268 (3), 267 (3), 265 (6), 264 (6), 250 (4), 236 (6), 194 (3), 180 (2), 178 (2), 161·5 (4), 154 (10), 146·5 (12), 132·5 (6), and 118·5 (9), *m** 278·7 (308 ⟶ 293).

Methylation of (A) (38 mg) with diazomethane in methanol–diethyl ether gave the 11-O-*methyl derivative* (Ic) (38 mg), which crystallised, after t.l.c. purification, from acetone–hexane as orange needles, m.p. 155—157°, $\lambda_{max.}$ (EtOH) 240 (log ε 4·13), 272sh (4·38), 280sh (4·40), 296 (4·46), 320sh (3·96), 346 (3·88), and 428 nm (3·46), $\lambda_{max.}$ (EtOH–NaOMe) 231sh, 244, 297, 323sh, 344sh, and 434nm, $\lambda_{max.}$ (EtOH–$AlCl_3$) 226, 236, 272sh, 300, 344sh, 362sh, and 434 nm, $\nu_{max.}$ 3400 (OH) and 1630 cm^{-1} (CO), δ($CDCl_3$) 1·52 (6H, s, CMe_2), 3·74 (3H, s, NMe), 4·02 (3H, s, OMe), 5·54 (1H, d, *J* 10 Hz, $-CH{=}CH-$), 6·15 (1H, s, H-5), 6·66 (1H, d, *J* 10 Hz, $-CH{=}CH-$), 7·11—7·30 (2H, complex, H-9 and H-10), 7·95 (1H, dd, *J* 7 and 3 Hz, H-8), and 14·81 (1H, s, exchanging with D_2O, OH), M^+ 337·1312 ($C_{20}H_{19}NO_4$ requires *M*, 337·1314).

Alkaloid (A) (20 mg), anhydrous K_2CO_3 (300 mg), methyl iodide (0·5 ml), and anhydrous acetone (5 ml) were stirred at room temperature for 4 days; methylation was then complete (t.l.c.). The solution was filtered and evaporated to give a solid (20 mg), which crystallised from diethyl ether–light petroleum (b.p. 60—80°) as yellow needles of the 6,11-*di*-O-*methyl derivative* (Ib), m.p. 97—98°, $\lambda_{max.}$ (EtOH) 234sh (log ε 3·98), 263 (4·32), 281 (4·31), 292sh (4·23), 315 (3·97), 335sh (3·61), and 396 nm (3·50) (no change with NaOMe or $AlCl_3$), $\nu_{max.}$ 1630 cm^{-1} (CO), δ($CDCl_3$) 1·51 (6H, s, CMe_2), 3·60 (3H, s, NMe), 3·95 (3H, s, OMe), 3·97 (3H, s, OMe), 5·54 (1H, d, *J* 10 Hz, $-CH{=}CH-$), 6·29 (1H, s, H-5), 6·66 (1H, d, *J* 10 Hz, $-CH{=}CH-$), 7·02—7·24 (2H, complex H-9 and H-10), and 7·90 (1H, q, *J* 7 and 2 Hz, H-8), M^+ 351·1449 ($C_{21}H_{21}NO_4$ requires *M*, 351·1470).

Alkaloid (A) (48 mg), acetic anhydride (1 ml), and pyridine (1 ml) were heated on a steam-bath for 2 h. The mixture was poured into water and the product extracted with chloroform. The extract was washed with water, dried, and evaporated to yield a solid (48 mg) which was purified on silica gel by elution with diethyl ether–light petroleum (b.p. 30—40°) (1 : 1), giving the 6,11-*diacetate* (37 mg, 62%), as the main product. This crystallised from diethyl

[10] J. R. Lewis and J. B. Reary, *J. Chem. Soc.* (*C*), 1970, 1662.
[11] H. J. Bowie, R. G. Cooks, R. H. Prager, and H. M. Thredgold, *Austral. J. Chem.*, 1967, **20**, 1179.
[12] H. R. Sullivan, R. E. Billings, J. L. Occolowitz, H. E. Boaz, F. J. Marshall, and R. E. McMahon, *J. Medicin. Chem.*, 1970, **13**, 904.
[13] G. Guroff, J. W. Daly, D. Jerina, J. Renson, B. Witkop, and S. Udenfriend, *Science*, 1967, **157**, 1524.

ether–petroleum as prisms, m.p. 178—181°, $\nu_{max.}$ 1765 (Ac) and 1645 cm^{-1} (CO), δ($CDCl_3$) 1·51 (6H, s, CMe_2), 2·90 (3H, s, Ac), 2·93 (3H, s, Ac), 3·58 (3H, s, NMe), 5·65 (1H, d, *J* 9 Hz, $-CH{=}CH-$), 6·46 (1H, s, H-5), 6·55 (1H, d, *J* 9 Hz, $-CH{=}CH-$), 7·20—7·42 (2H, complex, H-9 and H-10), and 8·18 (1H, dd, *J* 7 and 2 Hz, H-8), M^+ 407·1373 ($C_{23}H_{21}NO_6$ requires *M*, 407·1369).

Alkaloid (*B*), 3,12-*Dihydro*-6,11-*dihydroxy*-3,3,12-*dimethyl*-5-(3-*methylbut*-2-*enyl*)*pyrano*[2,3-c]*acridin*-7-*one* (IIa). Alkaloid (B) crystallised from diethyl ether–light petroleum (b.p. 60—80°) as red *needles* (24 mg, 0·003%), m.p. 190—191·5, R_F (benzene–ethyl acetate, 9 : 1) 0·40, R_F [diethyl ether–light petroleum (b.p. 30—40°), 1 : 1] 0·46, $\lambda_{max.}$ (EtOH) 239 (log ε 4·29), 272 (4·59), 296 (4·63), 323sh (4·22), 347 (4·07), and 436 nm (3·83), $\lambda_{max.}$ (EtOH–NaOMe) 244, 283, 299sh, 308sh, 341sh, and 460 nm, $\lambda_{max.}$ ($EtOH–AlCl_3$) 270, 308, 367, and 482 nm, $\nu_{max.}$ 3300 (OH) and 1650 cm^{-1} (CO), δ($CDCl_3$) 1·52 (s), 1·68 (s), 1·82 (s), 3·37 (d), 3·78 (s), 5·30 (m), 5·51 (d), 6·63 (d), 7·00—7·18 (m), 7·80—7·95 (m), and 14·32 (s), M^+ 391·1736 ($C_{24}H_{25}NO_4$ requires *M*, 391·1752), *m/e* 392 (31%), 391 (95), 390 (11), 377 (30), 376 (100), 360 (17), 349 (15), 348 (67), 346 (7), 337 (16), 336 (17), 334 (6), 322 (34), 320 (15), 319 (7), 318 (25), 308 (14), 306 (20), 295 (7), 294 (9), 293 (9), 281 (11), 280 (16), 269 (6), 243 (7), 219 (11), 167 (25), 160 (8), 153 (7), 119 (35), 100 (8), and 93 (4).

Alkaloid (B) (12 mg) in methanol was treated with diazomethane in diethyl ether at room temperature for 18 h. Evaporation of the solvent left a solid which crystallised from diethyl ether–light petroleum (b.p. 60—80°) as yellow needles (8 mg) of the 11-O-*methyl derivative*, m.p. 126·5°, $\lambda_{max.}$ (EtOH) 241 (log ε 4·13), 272sh (4·39), 296 (4·50), 319sh (4·10), 347 (3·91), and 430 nm (3·53), $\lambda_{max.}$ (EtOH–NaOMe) 242, 273sh, 284sh, 295, 320sh, 344, and 430 nm, $\lambda_{max.}$ ($EtOH–AlCl_3$), 266, 308, 364, and 486 nm, $\nu_{max.}$ 3420 (OH) and 1650 cm^{-1} (CO), δ($CDCl_3$), 1·52 (6H, s, CMe_2), 1·71 (3H, s, MeC=C), 1·83 (3H, s, MeC=C), 3·38 (2H, d, *J* 7 Hz, CH_2), 3·69 (3H, s, NMe), 3·98 (3H, s, OMe), 5·11 (1H, m, HC=C), 5·54 (1H, d, *J* 10 Hz, HC=C), 7·06—7·32 (2H, complex, H-9 and H-10), 7·94 (1H, dd, *J* 7 and 3 Hz, H-8), and 14·38 (1H, s, OH) (Found: C, 73·8; H, 7·0%; M^+, 405·1833. $C_{25}H_{27}NO_4$ requires C, 74·05; H, 6·7%; *M*, 405·1940).

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