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ACRIDONE ALKALOIDS FROM *SEVERINIA BUXIFOLIA**

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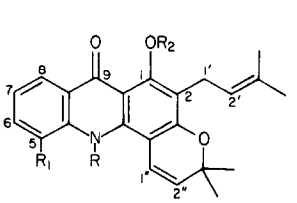
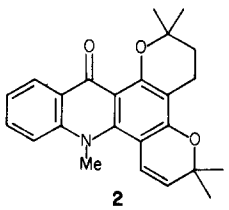
Key Word Index—*Severinia buxifolia*; Rutaceae; acridone alkaloids; severifoline; *N*-methylseverifoline; atalaphyllinine; *N*-methylatalaphylline; 5-hydroxy-*N*-methylseverifoline; ^{1}H NMR.

Abstract—Two new acridone alkaloids, severifoline and *N*-methylseverifoline along with the known alkaloids, *N*-methylatalaphylline, atalaphyllinine and 5-hydroxy-*N*-methylseverifoline, were isolated from the root bark of *Severinia buxifolia*. The structures of severifoline and *N*-methylseverifoline were established by chemical and spectroscopic methods.

INTRODUCTION

Severinia buxifolia is a wild thorny shrub which has been claimed as a useful folk medicine in the treatment of chronic rheumatism, paralysis, snake-bite and malaria[1]. Scora *et al.*[2–4] and Dreyer[5] have reported the isolation of essential oils, coumarins, alkaloids and triterpenoids from the leaves and fruits of this plant. The present report describes the isolation and the structural elucidation of two new acridone alkaloids, severifoline (**1c**) and *N*-methylseverifoline (**1a**), along with three known alkaloids atalaphyllinine (**1d**), *N*-methylatalaphylline (**3**), and **1e** from the root-bark of *S. buxifolia* collected in Formosa.





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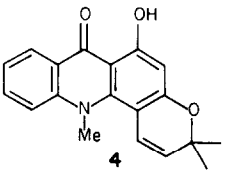
	R	R_1	R_2
1a	Me	H	H
1b	Me	H	Me
1c	H	H	H
1d	H	OH	H
1e	Me	OH	H



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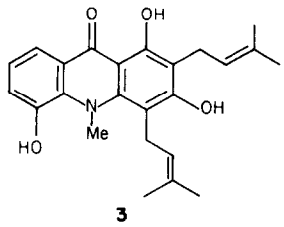

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RESULTS AND DISCUSSION

The ethanolic extracts of the root-bark of *S. buxifolia* was partitioned between H_2O and $CHCl_3$. Subsequent separation of the $CHCl_3$-soluble compounds was achieved by Si gel CC affording five acridone alkaloids.

N-Methylseverifoline (**1a**), orange-yellow needles, $C_{24}H_{25}NO_3$ (M^+ 375), mp 152–154°, showed a deep green color reaction with $FeCl_3$. Its UV spectrum exhibited absorption bands at λ_{max} 228, 254, 288, 295, 343 and 417 nm, characteristic of the 9-acridone system[6–11]. The IR spectrum of **1a** showed bands at 1642, 1605 and 1570 cm^{-1} which were also reminiscent of an acridone nucleus. In the UV spectrum of **1a**, no shift with alkali was observed, but a bathochromic shift with $AlCl_3$ indicated the presence of a chelated phenolic OH group[12]. Both acetylation with Ac_2O–pyridine at room temperature, or methylation with diazomethane of **1a** was unsuccessful. Methylation of **1a** was only achieved by heating with MeI and K_2CO_3 in acetone to yield **1b**. The similar lack of reactivity of a phenolic OH group has been observed in noracronycine (**4**) which has a peri-oriented OH group on the C-9 carbonyl moiety[12]. The ^{1}H NMR spectrum of **1a** is shown in Table 1, along with those of known acridone alkaloids isolated from the same plant at the same time for comparison. In the ^{1}H NMR of **1a**, a lower field sharp one-proton singlet at δ 14.92 which disappeared on exchange with D_2O, was assigned to the 1-QH group intramolecular hydrogen-bonded to the acridone carbonyl. The double doublet centred at δ 8.38 is characteristic of the C-8 proton in acridones [13]. The ^{1}H NMR spectrum of **1a** was in close agreement with that of **1e**, an alkaloid previously isolated by Fraser and Lewis [12], except for signals in the aromatic region (Table 1).

*Part XII in the series "Constituents of Formosan Folk Medicine—The Constituents of the Root Bark of *Severinia buxifolia*. I." For Part XI, see Wu T.-S., Kuoh C.-S., Ho S.-H., Yang M.-S. and Lee K.-K. (1981) *Phytochemistry* **20**, 527.

Table 1. ^{1}H NMR spectra of *Severinia* alkaloids

	1a	1b	1c*	1d†	1e†	2	3
8-H	8.38 (*dd*, 8)	8.40 (*dd*, 2.5, 8)	8.22 (*d*, 8)	7.70 (*dd*, 2, 8)	7.83 (*dd*, 2, 8)	8.34 (*dd*, 2, 8)	7.78 (*br, d*)
7-H	7.69 (*br, t*, 8)‡	7.60 (*br, t*, 8)‡		6.98 (*t*, 8)	7.12 (*t*, 8)	7.54 (*br, t*, 8)‡	7.07 (*t*, 8)
6-H	7.27 (*t*, 8)‡	7.28 (*t*, 8)‡	7.20 (1H, *m*)	7.10 (*dd*, 2, 8)	7.24 (*dd*, 2, 8)	7.14 (*br, t*, 8)‡	7.16 (*m*)
5-R_1 (1H)	7.40 (*d*, 8)	7.18 (*d*, 8)	7.53 (2H, *m*)	10.05 (*s*)	9.49 (*br, s*)	7.26 (*d*, 8)	9.32 (*br, s*)
N-R	3.87 (3H, *s*)	3.79 (CH, *s*)	9.94 (1H, *br s*)	8.68 (1H, *br s*)	3.80 (3H, *s*)	3.76 (3H, *s*)	3.61 (3H, *s*)
1-OR_2	14.92 (1H, *s*)	3.91 (3H, *s*)	14.70 (1H, *s*)	14.62 (1H, *s*)	14.63 (1H, *s*)	—	14.43 (1H, *s*)
1′-H (2H)	3.40 (*d*, 7)	3.41 (*d*, 7)	3.33 (*d*, 7)	3.32 (*d*, 7)	3.38 (*d*, 7)	2.66 (*t*, 7)	3.48 (*m*)
2′-H	5.32 (*t*, 7)	5.22 (*t*, 7)	5.22 (*t*, 7)	5.24 (*t*, 7)	5.32 (*t*, 7)	1.82 (*t*, 7)	5.28 (*m*)‡
3′-$(CH_3)_2$	1.69, 1.83	1.68, 1.82	1.65, 1.81	1.67, 1.81	1.71, 1.85	1.53 (6H)‡	1.72,§ 1.77§
1″-H	6.56 (*d*, 10)	6.54 (*d*, 10)	6.83 (*d*, 10)	6.59 (*d*, 10)	6.72 (*d*, 10)	6.50 (*d*, 10)	3.48 (*m*)
2″-H	5.49 (*d*, 10)	5.54 (*d*, 10)	5.54 (*d*, 10)	5.59 (*d*, 10)	5.55 (*d*, 10)	5.45 (*d*, 10)	5.36 (m)‡
3″-$(CH_3)_2$	1.54 (6H)	1.55 (6H)	1.47 (6H)	1.48 (6H)	1.55 (6H)	1.47 (6H)‡	1.82 (6H)§

Values are in ppm. Figures in parentheses are coupling constants in Hz.
*Recorded in $CDCl_3 + (CD_3)_2CO$.
†Recorded in $CDCl_3$ + 10% $(CD_3)_2SO$.
‡, §Values with same superscript can be interchanged.

The appearance of four aromatic protons signals at δ 7.69 (*br t*, $J = 8$ Hz), 7.27 (*t*, $J = 8$ Hz), and δ 7.40 (*d*, $J = 8$ Hz), along with C-8 H at δ 8.38 (*dd*, $J = 2.5, 8$ Hz) in the ^{1}H NMR of **1a**, indicated that ring A was unsubstituted.

These spectral data suggested the structure of *N*-methylseverifoline as formula **1a**.

Furthermore, treatment of **1a** with 85% formic acid at 80–90° for 3 hr gave **2** as pale yellow plates. In the ^{1}H NMR of **2**, new appearances of triplets at δ 2.66 (2H) and δ 1.82 (2H), and a singlet at δ 1.47 (6H), instead of the disappearance of a low-field singlet at δ 14.92 and the signals ascribed the protons of the prenyl moiety in **1a**, were reminiscent of the formation of a dimethylpyran ring in **2**. On the basis of these results, *N*-methylseverifoline should be represented by the formula **1a**, corresponding to 2-prenylated noracronycine.

Severifoline (**1c**) was obtained as pale yellow needles, mp 253–254°, $C_{23}H_{23}NO_3$ (M^+ 361). Its UV and IR spectra are characteristic of a 9-acridone [6–11] (see Experimental). The ^{1}H NMR spectrum (Table 1) of **1c** showed signals at δ 14.70 (1H, *s*) which disappeared on deuteration. This low-field signal is clearly due to the chelated OH at C-1. A one-proton singlet at δ 9.96 (*br*, also exchangeable on deuteration), instead of the N–Me signal at δ 3.87 in **1a** was attributed to an N–H proton. Four aromatic protons appeared at δ 8.22 (1H, *d*, $J = 8$ Hz), 7.20 (1H, *m*), and δ 7.53 (2H, *m*), and the signal pattern of the other protons was similar to the spectrum of **1a**. The above data were in accordance with the structure **1c** for severifoline. In agreement with this proposition, prolonged methylation of **1c** with MeI and K_2CO_3 furnished an N, O-Me derivative, $C_{25}H_{27}NO_3$ (M^+ 389), which was identical to **1b** described above by comparisons of their IR, PMR, MS, and mmp.

The other three alkaloids isolated from the same plant, were characterized as atalaphyllinine (**1d**) [14], *N*-methylatalaphylline (**3**) [15], and **1e*** [12] by comparison with the authentic samples or their derivatives (mmp, IR, ^{1}H NMR, MS).

EXPERIMENTAL

Mps are uncorr. ^{1}H NMR (100 MHz) were recorded in $CDCl_3$ except where noted. Chemical shifts are shown in ppm (δ) with TMS as int. standard. MS were recorded using a direct inlet system. UV were determined in MeOH and IR recorded in KBr except where noted.

Plant material. Severinia buxifolia (Poir.) Tenore (*Atalantia buxifolia*) was collected from Tainan, Taiwan, and identified by Prof. C.-S. Kuoh. The specimen is deposited in the Herbarum of Chia-Nan Junior College of Pharmacy, Tainan, Taiwan.

Extraction and separation. The EtOH extract of the root-bark (4.2 kg) was treated with $CHCl_3$ and H_2O. The $CHCl_3$ layer was evapd to dryness and subjected to Si gel CC by eluting successively with *n*-hexane, C_6H_6, C_6H_6–Me_2CO (1 : 1) and EtOAc. The C_6H_6 fraction was rechromatographed on Si gel and eluted with the same solvent to afford **1a** (80 mg), **1c** (50 mg), **3** (40 mg), **1d** (2.1 g), and **1e** (2.4 g).

N-*Methylseverifoline* (**1a**). Orange-yellow crystals from Me_2CO, mp 152–154°, calc. for $C_{24}H_{25}NO_3$, C: 76.77, H: 6.71, N: 3.73. Found: C: 76.61, H: 6.79, N: 3.49. UV λ_{max} nm: 228, 254, 288, 295, 343, 417. λ_{max} nm (+ $AlCl_3$): 236, 258, 298, 352, 428. IR ν_{max} cm^{-1}: 1642, 1605, 1570. MS *m/z* (%): 375 (M^+, 100), 360 (83), 332 (61), 320 (64), 306 (19).

Cyclization of N-*methylseverifoline* (**1a**). **1a** 30 mg was heated at 80–90° for 3 hr with HCO_2H (85%, 2 ml) and then left at room temp. overnight. H_2O was added and the soln extracted with $CHCl_3$. The $CHCl_3$ layer was washed with $NaHCO_3$, H_2O, dried and evapd. The residue was purified by prep. TLC (Si gel, C_6H_6–Me_2CO, 9:1) to afford pale yellow plates of **2**; mp 189–191° (Et_2O). UV λ_{max} nm: 210, 226, 243 (*sh*), 279, 296 (*sh*), 340 (*sh*), 400. IR ν_{max} cm^{-1}: 1640, 1575, 1557. MS *m/z* (%): 375 (M^+, 74), 360 (100), 312 (5), 320 (51), 304 (15), 290 (12).

N,O-*Dimethylseverifoline* (**1b**). (1) **1a** 20 mg in Me_2CO (3 ml) was refluxed with MeI (2 ml) and dry K_2CO_3 (2 g) for 36 hr. The soln was filtered evapd and the residue chromatographed on Si gel in C_6H_6–Me_2CO, (9:1) to yield **1b** as an amorphous gum. IR ν_{max} ($CHCl_3$) cm^{-1}: 1615, 1595, 1565. UV λ_{max} nm: 209, 230 (sh), 270 (sh), 280, 295 (sh), 340, 402. MS *m/z* (%): 389 (M^+, 70), 374 (100), 358 (35), 356 (33), 346 (36), 320 (94), 290 (30). (2) Treatment of **1c** (20 mg) in a similar way to (1) afforded an uncrystallizable gum which was identical to **1b** by comparison of IR, TLC, ^{1}H NMR and MS.

Severifoline (**1c**). Yellow needles (Me_2CO), mp 253–254°. UV λ_{max} nm: 224, 240, 253, 278, 296, 338, 410. IR ν_{max} cm^{-1}: 3330, 3240, 1640, 1610, 1540. MS *m/z* (%): 361 (M^+, 100), 346 (90), 318 (63), 306 (52), 291 (30), 290 (54), 278 (25), 262 (15).

Identification of the known compounds **1d**, **1e** and **3** involved direct comparison with authentic samples or their derivatives [12, 15].

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REFERENCES

1. Sasaki S. (1924) *Khoyo Taiwan Minkan Yakuyo Shokubutsu Shi*, p. 36 (khobunkan), Taipei.
2. Scora R. W. (1966) *Phytochemistry* **5**, 823.
3. Tin-Wa M., Scora R. W. and Kumanoto J. (1972) *Lloydia* **35**, 183.
4. Tin-Wa M., Bonomo S. and Scora R. W. (1979) *Planta Med.* **37**, 379.
5. Dreyer D. L. (1967) *Tetrahedron* **23**, 4613.
6. Sangster A. W. and Stuart K. L. (1965) *Chem. Rev.* **65**, 69.
7. Reisch J., Szendri K., Minker E. and Novak I. (1972) *Pharmazie* **27**, 208.
8. Brown R. D. and Lahey F. N. (1950) *Aust. J. Sci. Res.* **A3**, 593.
9. Brockmann H., Muxfekdt H. and Haese G. (1956) *Chem. Ber.* **89**, 2174.
10. Orgel L. E. (1965) *The Chemistry of Heterocyclic Compounds. Acridines* (Weissberger A., ed.), p. 289. Interscience, New York.
11. Price J. R. and Willis J. B. (1959) *Aust. J. Chem.* **12**, 589.
12. Fraser A. W. and Lewis J. R. (1973) *J. Chem. Soc. Perkin Trans.* 1, 1173; Rastogi K., Kapil R. S. and Popli S. P. (1980) *Phytochemistry* **19**, 945.
13. Oh C. S. and Greco C. V. (1970) *J. Heterocycl. Chem.* **7**, 261.
14. Basa S. C. (1975) *Phytochemistry* **14**, 835.
15. Govindachari T. R. and Viswanathan N. (1970) *Tetrahedron* **26**, 2905.

*The structure of this alkaloid corresponds to 5-hydroxy-*N*-methylseverifoline.