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NEW DIMERIC INDOLE ALKALOIDS FROM *ERVATAMIA HAINANENSIS*

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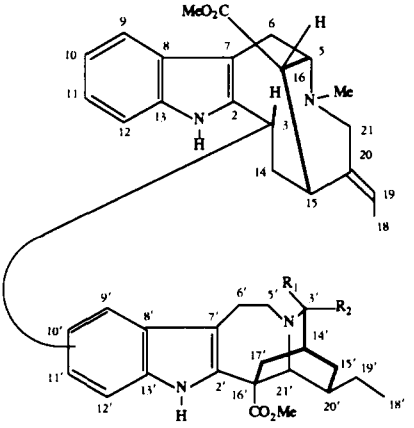
ABSTRACT.—Four new dimeric indole alkaloids, named ervahaimine A [**1**], ervahaimine B [**2**], ervahainamidine A [**3**], and ervahainamidine B [**4**], were isolated from the roots of *Ervatamia hainanensis*. The structures of these new alkaloids have been elucidated by spectral data in correlation with compounds of established structures. The structures of ervahaimine A and ervahaimine B have been confirmed by partial synthesis.

Three new bisindole alkaloids of the voacamine type and eleven monomeric indole alkaloids, which were isolated from the roots of *Ervatamia hainanensis* Tsiang (Apocynaceae), have been reported in previous papers (1,2). As part of our continuing studies on the isolation of active compounds for cardiovascular diseases from this plant, another four new minor bisindole alkaloids of the voacamine type, ervahaimine A [**1**], ervahaimine B [**2**], ervahainamidine A [**3**], and ervahainamidine B [**4**], have been obtained. We now report the structural elucidation of these alkaloids based on analysis of their spectral data by comparing with the spectra of known bisindole alkaloids of the voacamine type. The semisynthesis of two new alkaloids **1** and **2** has been carried out.

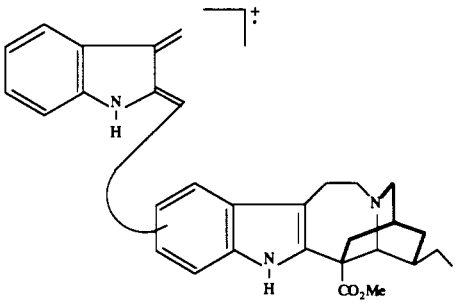
RESULTS AND DISCUSSION

Ervahaimine B [**2**], a white amorphous solid, $C_{42}H_{48}N_4O_5$ (based on hrms), dis-

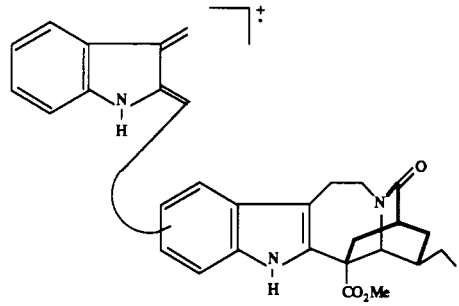


		Connection
1	R_1R_2=O	C-11′
2	R_1R_2=O	C-10′
3	R_1=H, R_2=CH(OH)Me	C-11′
4	R_1=H, R_2=CH(OH)Me	C-10′
5	$R_1=R_2$=H	C-10′
6	$R_1=R_2$=H	C-11′

played a typical indole uv spectrum [λ max (EtOH) 233, 289, 296 nm]. Its ir spectrum showed, in addition to an ester carbonyl absorption at 1730 cm^{-1}, the presence of a second carbonyl function at 1660 cm^{-1}. A molecular ion at *m/z* 688, 14 units higher than that of ervahanine B [**5**], and fragment ions at *m/z* 122, 180, 181, and 194 for the vobasan group in the mass spectrum suggested that ervahaimine B is a bisindole alkaloid of the voacamine type (3) and that the carbonyl function should be located in the iboga moiety. Confirmatory evidence for such an assignment came from the appearance of the abundant ion **8** at *m/z* 493 in the higher mass region of its spectrum, which is the typical ion in the mass spectrum of a bisindole alkaloid of the voacamine type (3). This ion is 14 units higher in comparison with ion **7** at *m/z* 479 obtained from alkaloids without the oxogroup such as ervahanines B [**5**] and A [**6**].



7 *m/z* 479



8 *m/z* 493

The position of the carbonyl group at C-3′ of ervahaimine B was revealed by the ^{1}H-nmr spectrum, in which a complex two-proton multiplet appears at δ 4.5 ppm. The multiplet was interpreted as being a singlet of a proton superimposed on multiplet peaks of another proton; these peaks were ascribed to C-14′ and C-21′ as in the case of 3-oxocoronaridine [**9**] (4). The aromatic protons of ervahaimine B can be located at δ 7.32 for H-9′, a doublet at δ 6.98 for H-11′ and a doublet at δ 7.17 for H-12′. Thus, ervahaimine B [**2**] was postulated to have the structure of 3′-oxoervahanine B. Comparison of ^{1}H-nmr data of ervahaimine B with ervahanine B allowed the signals of all protons in **2** to be assigned as listed in Table 1.

Ervahaimine A [**1**] gave a molecular ion at *m/z* 688 and other fragment ions at *m/z* 122, 180, 181, 194, 493 in the mass spectrum; these are the same as for ervahaimine B [**2**]. Its uv spectrum was quite similar to that of ervahaimine B. However, the ^{1}H-nmr spectra of ervahaimines A and B showed significant differences in the signals of aromatic protons. For ervahaimine A, a doublet at δ 7.34 for H-9′, a doublet at δ 6.95 for H-10′, and a singlet at δ 7.04 for H-12′ were observed. This suggested structure **1** for ervahaimine A, in which two moieties are linked at the C-11′ instead of the C-10′ position.

The proposed structures of ervahaimines A [**1**] and B [**2**] were confirmed by partial synthesis utilizing the method of Thomas *et al.* (5). Vobasinol [**10**], which was obtained from vobasine [**11**] by $NaBH_4$ reduction, was condensed with 3-oxocoronaridine [**9**] prepared from coronaridine [**12**] by I_2 oxidation in THF. Under acidic conditions a mixture of **1** and **2** was obtained. The mixture was separated by preparative tlc to two products. One of them was shown to be identical with ervahaimine B [**2**] by means of co-tlc, co-ir, and ms. The other one, present in a very small quantity, gave the same R_f value on tlc as that of ervahaimine A [**1**].

Ervahainamidines A [**3**] and B [**4**] were found to have the same molecular formula, $C_{44}H_{54}N_4O_5$ ($[M]^+$ 718.4074), with the indole chromophore as indicated by their uv

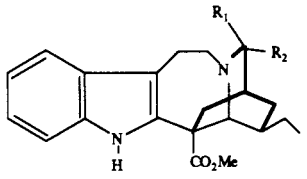
TABLE 1. ^{1}H-nmr data of ervahaimine B [**2**], ervahanine B [**5**], ervahainamidine A [**3**], and 3-hydroxyethylcoronaridine [**14**] ($CDCl_3$, $\delta_{TMS}=0$).

Proton	Compound			
	2[a]	**5**	**3**	**14**
H-3	4.65 dd	4.62 dd	4.67 dd	
H-5	4.08 dd	4.02 dd	4.10 dd	
H_R-6	3.21 dd	3.24 dd	3.27 dd	
H_S-6	3.53 dd	3.50 dd	3.52 dd	
H-9	7.58 m	7.58 dd	7.57 m	
H-10	7.10 m	7.10 m	7.10 m	
H-11	7.10 m	7.10 m	7.10 m	
H-12	7.10 m	7.10 m	7.50 m	
H_R-14	2.54 d	2.71 d	2.55 d	
H_S-14	1.98 m	1.92 ddd	1.89 m	
H-15	3.81 m	3.74 m	3.87 m	
H-16	2.75 dd	2.72 dd	2.70 dd	
H-18	1.65 d	1.63 d	1.68 d	
H-19	5.35 q	5.30 q	5.38 q	
H_R-21	3.78 d	3.73 d	3.76 d	
H_S-21	2.96 d	2.88 d	2.98 d	
N-Me	2.64 s	2.58 s	2.65 s	
OAc	2.48 s	2.45 s	2.47 s	
N-H	7.54 s	7.49 s	7.67 s	
H_R-3′[b]	—	2.88 d	—	—
H_S-3′[b]	—	2.78 d	2.77 br d	2.75 br d
H_R-5′[c]	3.31 m	3.33 m	3.43 m	3.45 m
H_S-5′[c]	3.17 m	3.15 m	3.32 m	3.35 m
H_R-6′[d]	3.14 m	3.09 m	3.15 m	3.2 m
H_S-6′[d]	3.01 m	2.95 m	2.92 m	3.0 m
H-9′	7.32 s	7.31 s	7.38 d	7.48
H-10′	—	—	7.00 d	7.11
H-11′	6.98 d	6.98 d	—	7.17
H-12′	7.17 d	7.16 d	7.03 s	7.26
H-14′	4.46 m	1.84 m	1.75 m	1.73 m
H_R-15′	1.71 dd	1.70 dd	1.55 m	1.58 m
H_S-15′	1.25 m	1.12 m	1.35 m	1.32 m
H_R-17′[e]	1.95 d	1.83 d	1.97 br d	1.92 br d
H_S-17′[e]	2.25 d	2.53 d	2.62 br d	2.67 br d
H-18′	0.95 t	0.88 t	0.91 t	0.91 t
H_R-19′	1.42 dq	1.42 dq	1.45 dq	1.48 dq
H_S-19′	1.52 dq	1.54 dq	1.60 dq	1.60 dq
H-20′	1.33 m	1.29 m	1.30 m	1.36 br dd
H-21′	4.46 s	3.48 s	3.72 s	3.70 nad[f]
OAc	3.71 s	3.64 s	3.73 s	3.75 s
N-H	7.98 s	7.77 s	7.73 s	7.75 s
C*H*OHMe	—	—	3.55 m	3.57 m
CHOH*Me*	—	—	1.10 d	1.17 d

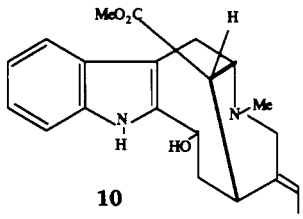
[a]Coupling constants for compound **2**: $J_{3,14R}=12$ Hz, $J_{3,14S}=3$ Hz, $J_{5,6R}=10$ Hz, $J_{5,6S}=2$ Hz, $J_{6R,6S}=14$ Hz, $J_{14R,14S}=14$ Hz, $J_{14R,15}=12$ Hz, $J_{14S,15}=3$ Hz, $J_{15,16}=2$ Hz, $J_{18,19}=7$ Hz, $J_{19,21R}=1$ Hz, $J_{19,21S}<0.2$ Hz, $J_{21R,21S}=12$ Hz, $J_{14',17'R}=3$ Hz, $J_{14',17'S}=3$ Hz, $J_{15'R,15'S}=12.5$ Hz, $J_{15'R,20'}=10$ Hz, $J_{15'S,20'}=5$ Hz, $J_{17'R,17'S}=12$ Hz, $J_{18',19'R}=7$ Hz, $J_{18',19'S}=7$ Hz, $J_{19'R,20'}=7$ Hz, $J_{19'S,20'}=7$ Hz, $J_{20',21'}<0.2$ Hz, $J_{11',12'}=8$ Hz.

[b–e]Assignments may be interchanged.

[f]nad = narrow doublet.



9 R_1R_2=O
12 R_1=R_2=H
14 R_1=H, R_2=CH(OH)Me



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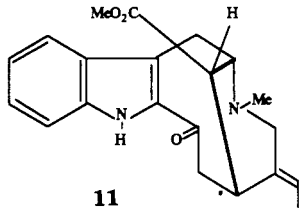

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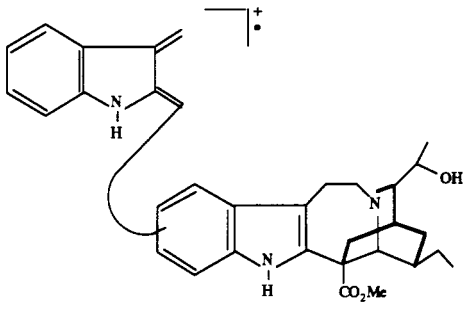

13 *m/z* 523

spectra. The two ir spectra indicated the presence of NH, OH, and ester carbonyl functionalities but absence of a carbonyl peak at 1660 cm^{-1}. Peaks in their mass spectra were almost identical in mass number but different in intensity. Of diagnostic significance were the ions at *m/z* 122, 124, and 136, which are found in coronaridine, and at *m/z* 122, 180, and 194, which are typical of vobasine.

The molecular ions ($[M]^+$ 718) and the typical ion **13** (*m/z* 523) of ervahainamidines A and B are 30 units higher than those of ervahaimine B [**2**] and 44 units higher than those of ervahanine B [**5**], indicating the presence of the same vobasine moiety in these voacamine-type alkaloids but indicating differences in the iboga part. In ^{1}H-nmr spectra the absence of the 2H multiplet at δ 4.50, which has been claimed to be characteristic for iboga-type alkaloids with the 3-carbonyl group, is in accord with the ir evidence. Two extra peaks of a three-proton doublet at δ 1.10 ppm (J = 7 Hz) and a one-proton multiplet at δ 3.55 ppm in ervahainamidine A (δ 1.15 ppm and 3.58 ppm in ervahainamidine B) were observed. The above data led us to postulate the presence of a hydroxyethyl group (-CHOHMe). Therefore, the iboga part of ervahainamidines A or B has an alternative structure: 3′-hydroxyethylcoronaridine or 3′-ethyl-19′-hydroxycoronaridine. Comparison of ^{1}H-nmr data of ervahainamidine A with those of 3-hydroxyethylcoronaridine [**14**] (Table 1) showed that ervahainamidine A has the structure **3**, because the chemical shift of H-19′ should be about δ 4.18 ppm while the location of the OH group is in the 19′ position (2). The different δ values of aromatic protons of ervahainamidines A and B in the ^{1}H-nmr spectra indicated that ervahainamidine B [**4**] is C-10′-connected.

The isolation of 3-hydroxyethylcoronaridine [**14**] and vobasine [**11**] in the monomeric fraction of the total alkaloids from *E. hainanensis* supported the above pro-

posal. The presence of **14** and **11** raises the question whether these dimers could be artifacts. The mild procedures used in the extraction and isolation of these alkaloids renders the above supposition unlikely.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—^{1}H-nmr spectra were recorded with an IEF 400 instrument, ir spectra with a Perkin-Elmer 257 spectrophotometer, and uv spectra with a Bausch & Lomb spectrophotometer. Mass spectra were recorded with AEI MS-50 and ZAB-2F mass spectrometers.

ISOLATION OF ALKALOIDS.—Extraction and isolation of total alkaloids have been reported in previous papers (1,2). The dimeric fractions were further fractionated by cc on Si gel and preparative tlc. Ervahaimines A, B and ervahainamidines A, B were obtained.

ERVAHAIMINE B [**2**].—$C_{42}H_{48}N_4O_5$; $[M]^+$ 688.3621, calcd 688.3624; uv λ max (EtOH) nm 233, 289, 296; ir ν ($CHCl_3$) cm^{-1} 3380, 3260, 2960, 2920, 1730, 1660, 1460, 740; ms *m/z* (%) $[M+Me-H]^+$ 702 (13), $[M]^+$ 688 (47), 657 (32), 644 (24), 630 (49), 599 (31), 571 (21), 540 (9), 493 (35), 450 (24), 435 (34), 320 (47), 309 (65), 307 (49), 283 (79), 225 (24), 220 (21), 194 (34), 182 (100), 181 (50), 180 (51), 166 (12), 154 (14), 136 (15), 124 (26), 122 (65), 110 (21); ^{1}H nmr see Table 1.

ERVAHAIMINE A [**1**].—$C_{42}H_{48}N_4O_5$; uv λ max (EtOH) nm 233, 289, 296; ir ν ($CHCl_3$) cm^{-1} 3320, 3220, 2920, 1724, 1652, 1450, 750; ms *m/z* (%) $[M+Me-H]^+$ 702 (10), $[M]^+$ 688 (55), 657 (30), 644 (27), 630 (52), 599 (42), 493 (45), 194 (40), 182 (95), 181 (60), 180 (100), 166 (14), 154 (17), 136 (20), 124 (30), 122 (70), 110 (30); ^{1}H-nmr (400 MHz, $CDCl_3$) δ 0.95 (t, 3H, $J=7$ Hz, H-18′), 1.40 (dq, 1H, $J_{19'R,18'}=J_{19'R,20'}=7$ Hz, H_R-19′), 1.50 (dq, 1H, $J_{19'S,18'}=J_{19'S,20'}=7$ Hz, H_S-19′), 1.65 (d, 3H, $J=7$ Hz, H-18), 2.45 (s, 3H, OAc), 2.64 (s, 3H, N-Me), 3.71 (s, 3H, OAc), 4.08 (dd, 1H, $J_{5,6R}=10$ Hz, $J_{5,6S}=5$ Hz, H-5), 4.49 (m, 1H, H-14′), 4.49 (s, 1H, H-21′), 4.67 (dd, 1H, $J_{3,14R}=12$ Hz, $J_{3,14S}=3$ Hz, H-3), 5.38 (q, 1H, $J_{18,19}=7$ Hz, H-19), 6.95 (d, 1H, $J=8$ Hz, H-10′), 7.04 (s, 1H, H-12′), 7.16 (m, 3H, H-10, H-11, H-12), 7.43 (d, 1H, $J=8$ Hz, H-9′), 7.62 (m, 1H, H-9), 7.51 (br s, 1H, N-H), 7.95 (br s, 1H, N-H).

ERVAHAINAMIDINE A [**3**].—$C_{44}H_{54}N_4O_5$; uv λ max (EtOH) nm 235, 290, 298; ms *m/z* (%) $[M+Me-H]^+$ 732 (43), $[M]^+$ 718 (88), 687 (63), 673 (44), 660 (48), 642 (85), 629 (52), 615 (100), 584 (48), 523 (20), 492 (19), 478 (50), 420 (25), 363 (21), 309 (19), 307 (19), 279 (18), 208 (10), 194 (41), 182 (90), 181 (78), 180 (88), 144 (8), 136 (44), 130 (15), 124 (13), 122 (85), 110 (13); ^{1}H nmr see Table 1.

ERVAHAINAMIDINE B [**4**].—$C_{44}H_{54}N_4O_5$; $[M]^+$ 718.4074, calcd 718.4094; uv λ max (EtOH) nm 235, 290, 298; ir ν ($CHCl_3$) cm^{-1} 3430, 3360, 3280, 2930, 1710, 1620, 1460, 750; ms *m/z* (%) $[M+Me-H]^+$ 732 (27), $[M]^+$ 718 (78), 687 (65), 673 (100), 660 (51), 642 (78), 615 (92), 584 (20), 523 (49), 492 (35), 478 (71), 420 (29), 363 (37), 309 (31), 307 (29), 279 (18), 208 (18), 194 (73), 182 (67), 181 (67), 180 (69), 156 (18), 144 (17), 136 (59), 130 (22), 124 (22), 122 (69), 110 (24); ^{1}H nmr (400 MHz, $CDCl_3$) δ 0.89 (t, 3H, $J=7$ Hz, H-18′), 1.15 (d, 3H, $J=7$ Hz, -CHOHMe), 1.70 (d, 3H, $J=7$ Hz, H-18), 2.46 (s, 3H, OAc), 2.70 (s, 3H, N-Me), 3.58 (m, 1H, -C*H*OHMe), 3.71 (s, 3H, OAc), 5.52 (q, 1H, $J=7$ Hz, H-19), 7.04 (d, 1H, $J=8$ Hz, H-11′), 7.11 (m, 2H, H-10, H-11), 7.21 (d, 1H, $J=8$ Hz, H-12′), 7.40 (s, 1H, H-9′), 7.53 (m, 1H, H-12), 7.60 (m, 1H, H-9), 7.77 (br s, 1H, N-H), 7.84 (br s, 1H, N-H).

SEMISYNTHESIS OF ERVAHAIMINES A AND B.—*Oxidation of coronaridine.*—A solution of I_2 (1053 mg) in THF (18 ml) was added dropwise to a stirred mixture of a solution of coronaridine (780 mg) in THF (20 ml) and $NaHCO_3$ (1145 mg) in H_2O (18 ml). After stirring 2 h, H_2O (50 ml) and CH_2Cl_2 (250 ml) were added. The organic phase was washed successively with sodium thiosulfate solution and H_2O, dried with anhydrous Na_2SO_4, and evaporated in vacuum. The residue was chromatographed on a Si gel column. 3-Oxocoronaridine [**9**] (260 mg) was obtained, which was identified by ir, ms, and tlc comparison with an authentic sample (2).

Reduction of vobasine HCl.—Excessive $NaHB_4$ was added to a solution of vobasine HCl (197 mg) in MeOH and reacted for 8 h at room temperature. H_2O and Et_2O were added. The Et_2O layer was dried with anhydrous Na_2SO_4 and evaporated in vacuum. The residue was chromatographed on a Si gel column. Vobasinol (60 mg) was isolated and identified by ms, ir, and tlc comparison with an authentic sample (2).

Reaction of 3-oxocoronaridine with vobasinol.—A mixture of 3-oxocoronaridine (30 mg) and vobasinol (30 mg) in 2% HCl (5 ml) was heated under reflux for 8 h below 75°. The reaction mixture was then diluted with H_2O and neutralized with K_2CO_3 giving a white precipitate which was extracted with $CHCl_3$. The residue obtained on evaporation was separated by cc on Sephadex LH-20 and preparative tlc to give er-

vahaimine B [**2**] (3 mg) which was identified by ms, ir, and tlc comparison with natural ervahaimine B and ervahaimine A (minor) which has the same R_f value on tlc as that of natural ervahaimine A.

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LITERATURE CITED

1. X.Z. Feng, C. Kan, H.-P. Husson, P. Potier, S.-K. Kan, and M. Lounasmaa, *J. Nat. Prod.*, **44**, 670 (1981).
2. X.Z. Feng, C. Kan, P. Potier, S.-K. Kan, and M. Lounasmaa, *Planta Med.*, **44**, 212 (1982).
3. H. Budzikiewiez, C. Djerassi, F. Puisieux, F. Percheron, and J. Poisson, *Bull. Soc. Chim. Fr.*, 1899 (1963).
4. J.P. Kutney and Ileana Perez, *Helv. Chim. Acta*, **65**, 2242 (1982).
5. D.W. Thomas, H. Achenbach, and K. Biemann, *J. Am. Chem. Soc.*, **88**, 1537 (1966).

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