Phytochemistry, Vol. 25, No. 8, pp. 2005–2007, 1986.
Printed in Great Britain.

0031–9422/86 $3.00 + 0.00
Pergamon Journals Ltd.

DIHYDROPERFAMINE, AN ALKALOID FROM *HAPLOPHYLLUM GLABRINUM*

Zs. RÓZSA, M. RÁBIK, K. SZENDREI, A. KÁLMÁN*, Gy. ARGAY*, I. PELCZER†, M. AYNECHI‡, I. MESTER§ and J. REISCH§

Department of Pharmacognosy, University Medical School, Szeged, Hungary; *Central Research Institute for Chemistry, The Hungarian Academy of Sciences, Budapest, Hungary; †EGIS Pharmacochemical Works, Spectroscopy Department, Budapest, Hungary; ‡Department of Pharmacy, Tehran University, Tehran, Iran; §Institute of Pharmaceutical Chemistry, Westfalian Wilhelms University, Münster, West Germany

(*Received* 6 *November* 1985)

Key Word Index—*Haplophyllum glabrinum*; Rutaceae; roots; alkaloids; coumarins; lignan.

Abstract—A new tetrahydrofuroquinoline alkaloid has been characterized from the roots of *Haplophyllum glabrinum* besides known alkaloids, coumarin and lignan derivatives. Its molecular structure was established by X-ray analysis.

INTRODUCTION

The genus *Haplophyllum* A. Juss. (Rutaceae) consists of about 70 perennial herbs distributed from the Mediterranean to eastern Siberia. Data about the secondary metabolites of 25 *Haplophyllum* species are available with alkaloids, amines, amides, coumarins and lignans reported from them. No chemical investigation has previously been made on *H. glabrinum* native to Iran. The present communication reports on the isolation and stereochemistry of a new alkaloid, named dihydroperfamine (**2**) besides known alkaloids, coumarin and lignan derivatives.

RESULTS AND DISCUSSION

The known compounds, perfamine (**1**), γ-fagarine, haplofidine, skimmianine, 7-isopentenyloxy-γ-fagarine, evoxine, flindersine, methylevoxine, evodine, herniarin, marmesin and justicidin B were identified by comparison of their spectra with published data or by co-TLC with authentic samples. The new alkaloid, designated as HG/1 had UV absorption characteristics for tetrahydrofuroquinoline alkaloids [1]. Its ^{1}H NMR spectrum and the mass spectral fragmentation pattern is very similar to those of **1** (Tables 1 and 2) [2–4]. Both compounds have the same substituents. X-ray analysis of the title compound* substantiated structure **2** depicted in Fig. 1. The saturated bond between the C-5 and C-6 atoms is demonstrated by the bond distance of 1.525 (7) Å and the C–C–C bond angles at these atoms 112.4 (7) and 109.7 (8)°. As shown by the puckering parameters [5]: o = 0.454 (7) Å, θ = 126.3 (8)°, ϕ = 9.5 (1.0)° and the lowest asymmetry factor [6] fC_s (C-6) = 5.0×10^{-2} Å the cyclohexenone ring assumes a slightly distorted envelope (sofa) conformation with C-6 on the flap. In accordance with the interpretation of the IR spectrum the carbonyl group belongs to C-7. The methoxy moiety (δ3.03) is bound to C-8 pseudo*equatorially* (the relevant torsion angles: O-12–C-8–C-8a–C-5a L = 128.6 (8) and O-12–C-8–C-7–C-

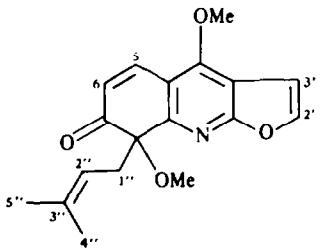
Table 1. ^{1}H NMR spectral data of compounds **1** and **2** [250 MHz, $CDCl_3$, TMS as internal standard; multiplicities and coupling constants (J, Hz) are given in parentheses]

H	1	2
2′	7.66 (*d*, *J* = 2.74)	7.63 (*d*, *J* = 2.74)
3′	7.05 (*d*, *J* = 2.74)	7.01 (*d*, *J* = 2.76)
1″	2.79 (*d*, *J* = 7.69)	2.92 (*d*, *J* = 6.50)
2″	4.81 (*t*)	4.89 (*t*)
4″, 5″	1.53 (*s*), 1.32 (*s*)	1.59 (*s*), 1.47 (*s*)
5	7.93 (*d*, *J* = 10.26)	2.5–3.3 (*m*)
6	6.18 (*d*, *J* = 10.26)	
4-OMe	4.38 (*s*)	4.33 (*s*)
8-OMe	3.10 (*s*)	3.02 (*s*)

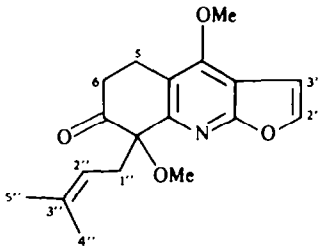
Table 2. Some characteristic mass spectral fragments (% relative intensity in parentheses) from **1** and **2**

	1	2
$[M]^+$	313 (1)	315 (2)
$[M-15]^+$	298 (0.5)	300 (0.5)
$[M-31]^+$	282 (1)	284 (5)
$[M-47]^+$	266 (3)	268 (22)
$[M-69]^+$	244 (12)	246 (60)

*A list of atomic parameters, bond distances and angles, torsion angles are deposited at the Cambridge Crystallographic Data Centre. Structure Factor Table can be obtained from the author (Professor A. Kálmán) on request.



1



2

6 = 159.5 (9)°. The terminal methyl group is in a synclinal [7] position with both C-7 and C-8a about the C-8–O-12 bond. The second substituent of C-8, i.e. the bulky prenyl group, is pseudo*axially* orientated to the carbocyclic ring C-14–C-8–C-7–C-6 = −85.9(8) and C-14–C-8–C-8a–C-5a = 115.1 (8)° which may account for its vigorous thermal motion. The mean value of the isotropic vibrational parameter of the the C-15 . . . C-18 carbon atoms (13.0 $Å^2$) is almost three times greater than the mean $\bar{B}_{eq}$ for the other 19 non-hydrogen atoms (4.7 $Å^2$). The second methoxy group bound to C-4 is somewhat bent out of the least-squares plane of the planar central hetero ring. The adjoining five-membered hetero ring is also planar. The double bond between C-2′ and C-3′ atoms is shown by the short bond length of 1.329 (10) A. ^{13}C NMR data of **1** and **2** are summarized in Table 3.

The occurrence of dihydro- and tetrahydrofuroquinoline alkaloids such as **1** and **2** seems to be restricted to the genus *Haplophyllum*. Representatives of these alkaloid-types have not been isolated from other Rutaceae species so far [8].

Among the known compounds herniarine and marmesine are reported for the first time from *Haplophyllum* species.

EXPERIMENTAL

Roots of *H. glabrinum* were collected in Iran. Voucher specimen is deposited in the herbarium of the Department of Pharmacy, Tehran University, Iran.

Extraction and isolation. Dried, powdered roots (1.48 kg) were extracted in a Soxhlet with petrol, then with $CHCl_3$. The residue of the $CHCl_3$ extract was evapd (18.3 g) and fractionated on a

Table 3. ^{13}C NMR spectral data of compounds **1** and **2** (22.63 MHz, $CDCl_3$, TMS as internal standard)

C	1	2
2	162.10	164.09
3	115.55	117.98
4	157.97	157.53
5	137.19	38.74
5a	105.58	105.38
6	124.37	19.92
7	201.39	209.27
8	86.32	84.94
8a	157.61	152.05
2′	143.52	143.23
3′	105.29	104.62
1″	42.26	38.80
2″	113.64	117.57
3″	136.16	135.43
4″	17.57	17.72
5″	25.75	25.76
4-OMe	59.05	58.62
8-OMe	53.92	52.78

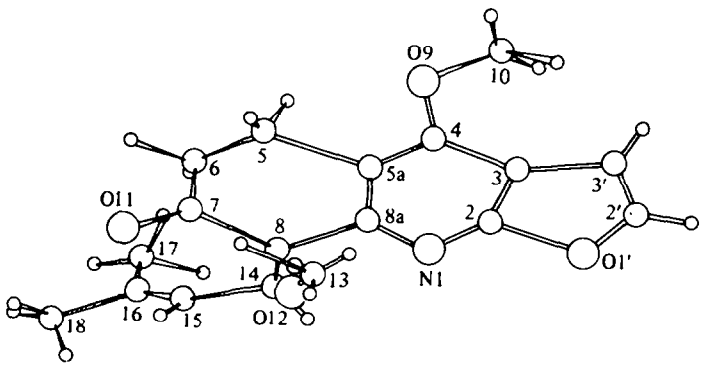


Fig. 1. Perspective view of the molecular structure of dihydroperfamine **2** with atomic numbering. The bare numbers are for carbon atoms unless indicated otherwise. The H atoms are unlabelled.

polyamide column (195 g) with MeOH–H_2O (2:3, 3:2 and 4:1). Fractions eluted with MeOH–H_2O (2:3) were refractionated on a silica gel column with *n*-hexane, *n*-hexane–Me_2CO (9:1 and 7:3), silica gel prep. layers in *n*-hexane–EtOAc (7:3, 3:2) C_6H_6–EtOAc (4:1, 7:3 and 3:2) and polyamide prep. layers in MeOH–H_2O (3:1 and 1:1). By this method 20 mg 7-isopentenyloxy-γ-fagarine, 19 mg flindersine, 10 mg γ-fagarine, 10 mg skimmianine, 4 mg haplofidine, 4.5 mg evoxine, 850 mg perfamine, 400 mg dihydroperfamine, 7.9 mg methylevoxine, 11.4 mg evodine, 7 mg marmesin, 1 mg herniarin and 20 mg justicidin B were obtained.

Perfamine (**1**). Colourless prisms from *n*-hexane–Me_2CO, mp 175–178°; $[\alpha]_{546}^{26.5}$ −20 ($CHCl_3$; *c* 1.00); (lit. mp 164–165°; $[\alpha]$ + 53.4 [2]). UV λ_{max}^{MeOH} nm: 265, 272, 343. ^{1}H NMR, ^{13}C NMR and MS data: Tables 1–3.

Dihydroperfamine (**2**). Colourless prisms from *n*-hexane–Me_2CO, mp 179–183°; $[\alpha]_{546}^{26.5}$ −640 ($CHCl_3$; *c* 1.00) UV λ_{max}^{MeOH} nm: 266, 278 sh. ^{1}H NMR, ^{13}C NMR and MS data: Tables 1–3.

X-ray structure determination of dihydroperfamine (**2**). $C_{18}H_{21}NO_4$ crystallized in the rhombohedral space group R3, $Z = 3$ with $a = 10.981$ (3) Å, $\alpha = 97.62$ (2)°, $D_c = 1.222$ g/cm^3, V = 1285.5 (1.3) Å^3. Intensities of 1987 reflections were measured in the range $1.5 < \theta > 27.0°$ on a computer controlled CAD-4 diffractometer using graphite monochromated Mo-K_α radiation ($\lambda = 0.71073$ Å) and $\omega/2\theta$ technique. The structure was solved by MULTAN [9] and refined using 1545 observed reflections with $I > 3.0\sigma(I)$. At the end of the isotropic refinement an empirical absorption correction of the intensities was performed by the program DIFABS [10]. This improved *R* from 0.127 to 0.120. At this stage the hydrogens were entered in calculated positions since they were all bonded to carbon atoms of well defined geometry. Due to the vigorous thermal motion of the atoms in the prenyl group positional disorder of the terminal C-17 and C-18 methyl groups had to be presumed. However, the additional peaks assigned from a difference Fourier map at $R = 0.075$ could not be refined properly as the alternative positions of these carbon atoms. The anisotropic refinement was terminated at $R = 0.069$ ($R_w = 0.059$). No hydrogen positions were refined. Atomic scattering factors were taken from standard tables [11].

Acknowledgement—The X-ray investigation of dihydroperfamine was sponsored by the Ministry of Health, Budapest, Hungary.

REFERENCES

1. Faizutdinova, Z. Sh., Bessonova, I. A. and Yunusov, S. Yu. (1969) *Khim. Prir. Soedin.* 360. [*Chem. Abstr.* (1969) **71**, 3517a.]
2. Razakova, D. M., Bessonova, I. A. and Yunusov, S. Yu. (1975) *Khim. Prir Soedin.* 812. [*Chem. Abstr.* (1976) **84**, 180443n.]
3. Razakova, D. M., Bessonova, I. A. and Yunusov, S. Yu. (1979) *Khim. Prir. Soedin.* [*Chem. Abstr.* (1981) **94**, 175337h.]
4. Grina, J. A., Ratcliff, M. R. and Stermitz, F. R. (1982) *J. Org. Chem.* **47**, 2648.
5. Cremer, D. and Pople, J. A. (1975) *J. Am. Chem. Soc.* **97**, 1354.
6. Kálmán, A., Czugler, M. and Simon, K. (1982) in *Molecular Structure and Biological Activity* (Griffin, J. F. and Duax, W. L., eds) p. 367. Elsevier Biomedical, New York.
7. Klyne, W. and Prelog, V. (1960) *Experientia* **16**, 521.
8. Mester, I. (1983) *Chemistry and Chemical Taxonomy of the Rutales* (Waterman, P. G. and Grundon, M. F., eds) p. 31. Academic Press, London.
9. Main, P., Hull, S. E., Lessinger, L., Germain, G., Declercq, J.-P. and Woolfson, M. M. (1978) *MULTAN 78. A System of Computer Programs for the Automatic Solution of Crystal Structures from X-ray Diffraction Data.* Universities of York, England and Louvain, Belgium.
10. Walker, N. and Stuart, D. (1983) *Acta Crystallogr.* **A39**, 158.
11. *International Tables for X-ray Crystallography* (1962) Vol. III. Kynoch Press, Birmingham.