Chemistry of Natural Compounds, Vol. 36, No. 6, 2000

ALKALOIDS OF *Peganum harmala*

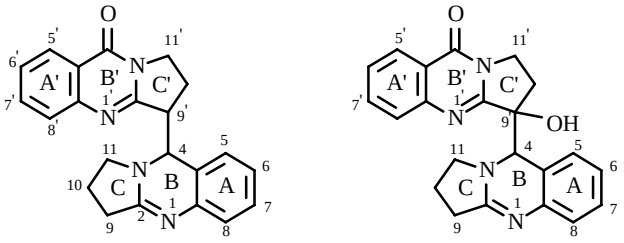
M. F. Faskhutdinov, M. V. Telezhenetskaya, M. G. Levkovich, and N. D. Abdullaev

UDC 547.944/945

Dipegine (1) and dipeginol (2) were isolated from Peganum harmala. *The structures of these alkaloids were established by mass and IR spectra.*

Key words: *Peganum harmala*, alkaloid, dipegine, dipeginol, NMR.

In continuation of research on the alkaloid composition of the aerial part of *Peganum harmala* collected during flowering and at the start of fruiting, we isolated two dimeric quinazolone alkaloids **1** and **2**. According to IR, UV, and mass spectra, **1** was identified as the previously isolated alkaloid dipegine [1]. However, we re-examined its structure on the basis of the mass spectra and ^{1}H and ^{13}C NMR spectra. Alkaloid **2** was new. We named it dipeginol. The structures of **1** and **2** were proven by complete analysis of the ^{1}H and ^{13}C NMR spectra and interpretation of the COSY-45 and DEPT spectra.



1 **2**

Dipegine (1) was isolated as colorless crystals of elemental composition $C_{22}H_{20}N_4O$ according to high-resolution mass spectrometry. The presence in the mass spectrum of peaks for the molecular ion with *m*/*z* 356 and for ions with *m*/*z* 171 and 185 indicates that **1** is a dimeric base that contains the fragments deoxypeganine [2, 3] and deoxyvasicinone [4, 5]. The UV spectrum of **1** is characteristic of quinazolone alkaloids. The IR spectrum has an absorption band for an amide carbonyl at 1660 cm^{-1}. The ^{1}H NMR spectrum (Table 1) exhibits signals for eight aromatic protons coupled to aromatic rings **A** and **A′**, five methylene groups, and two methine protons. The ^{13}C NMR spectrum (Table 2) contains signals for 22 C atoms. The signal characteristic of a carbonyl in the quinazolone heterocycle appears at 163.3 ppm.

Important information about **1** is obtained from the signals in the NMR spectrum for the protons in the 4, 9′, 10′, and 11′ positions, which are related by SSCCs. The coupled protons were identified by performing two-dimensional COSY using COSY-45. This produced pure cross-peaks between protons with geminal and vicinal SSC. A 1-H doublet at 5.70 ppm is assigned to H-4. A 1-H doublet of triplets at 3.47 ppm belongs to the neighboring proton of the bridge on C-9′. Analysis of the COSY spectrum revealed protons 2H-10′ as two 1-H multiplets at 2.06 and 2.12 ppm and signals of well resolved two 1-H multiplets with chemical shifts 3.89 and 4.14 ppm from the methylene at C-11′, which closes the chain of mutually coupled protons. Taking into account the position of the dimeric bridge, the multiplets at 3.89 and 4.14 ppm can be assigned to protons in the 9′ and 11′ positions. A comparison of the ^{1}H NMR spectra of **1** and vasicinone (**3**) (Table 1) showed that both spectra contain this group of signals with the difference that they are shifted to weak field by 0.1-0.2 ppm in **3**. These multiplets at 4.03 and 4.33 ppm in **3** are known to belong to the protons of the C-11 methylene. Therefore, it was concluded that the C-11′ in **1** is not substituted, i.e., the dimeric bridge is located at the C-4–C-9′ position and not C-4–C-11′, as indicated earlier [1].

S.Yu. Yunusov Institute of the Chemistry of Plant Substances, Academy of Sciences of the Republic of Uzbekistan, Tashkent, fax (99871) 120 64 75. Translated from Khimiya Prirodnykh Soedinenii, No. 6, pp. 481-484, November-December, 2000. Original article submitted November 13, 2000.

 0009-3130/00/3606-0602$25.00

TABLE 1. ^{1}H NMR Spectra of Vasicinone, Dipegine, Dipeginol, and Dipeginol Acetate ($CDCl_3$, TMS = 0)

Protons	Vasicinone		Protons	Dipegine		Dipeginol		Dipeginol acetate	
	d, ppm	J, Hz		δ, ppm	J, Hz	δ, ppm	J, Hz	δ, ppm	J, Hz
			H-4	5.70	2.1	5.40	s	5.64	s
			H-5	7.02	8.0, 1.5	6.92	7.5; 1.5	7.30	d
			H-6	7.18	8.0, 6.9, 1.8	7.30	7.5; 7.2; 1.8	7.38	t
			H-7	7.04	7.9, 6.9, 1.5	7.24	7.2; 7.2; 1.5	7.31	t
			H-8	7.09	7.9, 1.8	7.46	7.2; 1.8	7.49	d
			H-9a	2.64	16.5, 9.5, 9.5	3.04	m	4.02	9.8; 7.8; 7.8
			H-9e	2.56	16.5, 6.5, 6.5	2.87	m	3.74	8.5: 8.5
			H-10a	1.82	2H, m	2.13	m	2.31	m
			H-10e	-	-	2.37	m	2.16	m
			H-11a	2.79	10.1, 7.8, 7.8	2.90	12.4; 8.6; 8.0	3.35	m
			H-11e	3.14	10.1, 5.9, 5.9	3.89	12.4; 9.0; 3.2	3.19	m
H-5	8.28	8.1, 1.3, 1.2	H-5′	8.25	8.0, 1.4, 0.6	8.20	8.1; 1.5; 0.5	8.25	8.0; 1.2
H-6	7.51	8.1, 6.8, 1.3	H-6′	7.44	8.0, 6.7, 1.6	7.50	8.1; 6.8; 1.5	7.52	8.0; 7.9
H-7	7.78	8.2, 6.8, 1.3	H-7′	7.72	8.2, 6.7, 1.4	7.80	8.2; 6.8; 1.5	7.79	7.9; 7.9; 1.2
H-8	7.71	8.2, 1.3, 1.2	H-8′	7.67	8.2, 1.6, 0.6	7.73	8.2; 1.5; 0.5	7.67	7.9
H-9	5.16	7.3, 6.8	H-9′	3.47	8.9, 8.9, 2.1	-	-	-	-
H-10a	2.26	13.4, 8.4, 7.2, 6.8	H-10′a	2.06	13.4, 9.0, 8.9, 8.2	2.25	14.0; 9.0; 8.0	2.56	14.9; 10.0; 8.0
H-10e	2.63	13.4, 7.6, 7.3, 4.7	H-10′e	2.12	13.4, 8.9, 8.2, 4.3	2.66	14.0; 7.8; 3.4	2.72	14.9; 12.2; 2.7
H-11a	4.33	12.3, 7.6, 7.2	H-11′a	4.14	12.4, 8.2, 8.2	4.52	10.9; 8.0; 7.8	4.11	12.2; 12.2, 8.0
H-11e	4.03	12.3, 8.4, 4.7	H-11′e	3.89	12.4, 9.0, 4.3	3.78	10.9; 9.0; 3.4	3.14	12.2, 10.0; 2.7
			OH-9′	-	-	-	-	-	-
			Me	-	-	-	-	1.98	s

TABLE 2. ^{13}C NMR Spectra of Dipegine (**1**), Dipeginol (**2**), and Dipeginol Acetate ($CDCl_3$, TMS = 0)

C atoms	Dipegine (**1**), δ, ppm	Dipeginol (**2**), δ, ppm	Dipeginol acetate, δ, ppm	C atoms	Dipegine (**1**), δ, ppm	Dipeginol (**2**), δ, ppm	Dipeginol acetate, δ, ppm
C-10	20.49	20.01	20.88	*C-6′	127.29	127.93	128.04
C-10′	20.55	30.91	28.70	*C-8′	127.38	128.16	128.53
C-9	32.05	32.41	30.79	*C-6	127.76	129.35	129.26
C-11	45.57	43.63	44.02	*C-5′	129.64	131.14	132.03
C-11′	51.92	55.78	55.81	C-7′	135.08	135.42	133.96
C-9′	53.99	83.19	87.58	C-4a	144.03	135.42	135.55
C-4	58.34	63.68	62.83	C-4a′	149.69	149.19	148.84
C-2	121.31	121.49	121.61	C-8a	159.19	158.61	155.12
C-2′	121.53	127.18	127.51	C-8a′	161.57	161.09	160.65
*C-5	124.82	115.39	113.34	C-4′	163.28	165.45	166.73
*C-7	125.32	118.82	119.60	CO	-	-	169.37
*C-8	126.34	127.18	127.91	Me	-	-	22.25

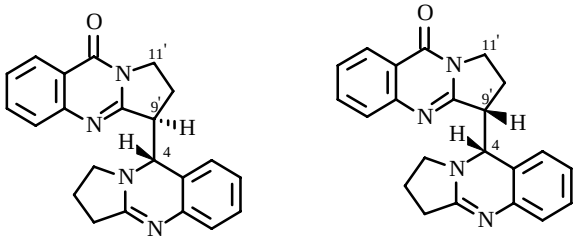
*Signals may be interchanged.

Two asymmetric centers at C-4 and C-9′ occur in dipegine. However, the absolute configurations could not be determined owing to the small amount of compound. Nevertheless, we attempted to determine their relative configurations. Neither molecular modeling using energy criteria nor NMR in achiral solvents could resolve the enantiomers. Therefore, only two diastereomers of the four possible were modeled. These are designated as dipegine S (DS) and dipegine R (DR). The two optical centers in the former have opposite configurations; in the latter, identical.

Barriers to rotation of the dimer halves around the bridging bond were calculated by molecular mechanics. These dependences of the steric strain energy on the dihedral angle H(9)′–C(9)′–C(4)–H(4), as expected, showed that both

TABLE 3. Dihedral Angles (degrees), Strain Energies (kcal/mol), Populations, Calculated Vicinal SSCC ^{3}J(H(9)′—H(4)) of Ground-State Conformations of Dipegine

Conformation	Dihedral angle	E, strain energy	Population	SSCC ^{3}J, Hz	Average ^{3}J
DR1	59.3	46.1	0.99	3.4	
DR2	-162.7	52.1	$4 \cdot 10^{-5}$	15.9	3.4
DR3	-64.6	50.9	$2.6 \cdot 10^{-4}$	2.2	
DS1	-60.9	48.5	0.83	3.0	
DS2	73.6	49.5	0.16	1.5	2.7
DS3	-154.3	51.5	$5.1 \cdot 10^{-3}$	14.8	
Experiment					2.1



DS DR

diastereomers have three relatively stable conformations. All (six) conformations were optimized. The calculated values of the dihedral angles and their strain energies are listed in Table 3.

The key parameter for deducing the three-dimensional structure is the vicinal SSCC between H-9′ and H-4. This vicinal constant is calculated using the formula:

$^3J_{(HH')} = A + B\cos\theta + C\cos 2\theta + \cos\theta[(\Delta S_1 + \Delta S_4)\cos(\theta - 120) + (\Delta S_2 + \Delta S_3)\cos(\theta + 120)]$, where A, B, and C are constants (A = 8.17, B = 1.96, C = 6.54), ΔS_1, ΔS_2, ΔS_3, and ΔS_4 are table values for the substituent increments [6].

The vicinal constants for both diastereomers were averaged taking into account the populations of their conformations. The populations of the conformations were estimated from the Boltzmann distribution. The final data for these conformations (Table 3) suggest that the studied dimeric quinazolone alkaloid **1** most probably has opposite configurations for C-4 and C-9′ relative to each other.

Thus, the NMR results for the dimeric quinazolone alkaloid pegine (**1**) suggest the new structure with the bridge located between C-4 and C-9′. The molecular-mechanics calculations and SSCC suggest the most probable relative configurations for the asymmetric C atoms C-4 and C-9′.

Dipeginol (2) is a new dimeric quinazolone alkaloid of composition $C_{22}H_{20}N_4O_2$, mp 243°C (dec.). The UV spectrum of **2** (λ_{max}/CCl_4 276, 302, 315 nm) is analogous to that of dipegine. The mass spectrum has strong peaks with *m*/*z* 201 and 171 and weak ions corresponding to decomposition of vasicinone [5, 7] and deoxypeganine. The IR spectrum has an absorption band for amide carbonyl at 1680 cm^{-1}. Thus, **2** is proposed to have a structure similar to dipegine and is a dimeric base consisting of vasicinone and deoxypeganine moieties.

The ^{1}H NMR spectrum of **2** (Table 1) has signals from eight protons of aromatic rings **A** and **A**′ in the aromatic region. These are very similar to those of **1**. Of these, four protons correspond with the vasicinone part of the dimer; the remaining four, with the deoxypeganine part. The presence in the ^{1}H NMR spectrum of a 1-H singlet at 5.40 ppm that is assigned to H-4 indicates that the dimeric bridge is joined on one hand to C-4 of the deoxypeganine part of the dimer. The lack of splitting of the signal for H-4 due to vicinal SSCC indicates that the dimeric bridge on the other hand is joined to a quaternary (without a proton) C atom. Recalling that the ^{1}H NMR spectrum contains signals for four protons coupled to each other via geminal and vicinal SSCC at 2.25, 2.66, 4.52, and 3.78 ppm, which are characteristic of two neighboring methylene groups, it can be proposed that the dimeric bridge is joined to C-9′, which also has an OH group on it.

Recording the ^{1}H NMR spectrum of **2** in DMSO, which decreases the rate of proton exchange, confirms that the dimer contains on OH, which appears at 6.90 ppm. Acetylation of **2** by acetic anhydride in pyridine gave the monoacetyl derivative.

The ^{13}C NMR spectrum of **2** (Table 2) exhibits signals for 22 C atoms. A singlet at 83.19 ppm is assigned to C-9′, which shifts upon acetylation by 4.3 ppm to weak field (87.58 ppm). The signal for C-10′ also shifts by 2.2 ppm, which

confirms the structure of **2** that we proposed.

The ^{1}H and ^{13}C NMR spectra of **2** and a comparison of the resulting properties with those of vasicinone and **1** suggest that the dimeric bridge is located between C-4 and C-9′. This conclusion agrees well with the genesis of this series of alkaloids.

Thus, the comprehensive study of the ^{1}H and ^{13}C NMR spectra of **2** and its acetyl derivative suggest that the structure of the new dimeric quinazolone alkaloid is 9′-hydroxydipegine.

EXPERIMENTAL

UV spectra were recorded in CCl_4 solution on a Perkin—Elmer Lambda-16 instrument. IR spectra were taken on a Perkin—Elmer FT-IR 2000 spectrometer (in KBr pellets). NMR spectra were recorded on a Varian UNITYplus 400 spectrometer [^{1}H (400 MHz) and ^{13}C (100 MHz)]. Chemical shifts are given in ppm relative to TMS. DEPT and 2D-experiments were carried out using standard microprograms of Varian. Mass spectra were measured on an MX-1310 instrument. Molecular mechanics calculations used the program set HyperChem 6.0 Evaluation, which was supplied by Hypercube Inc., for which the authors are grateful.

General comments on the calculational material and the extraction of the total alkaloids have been published [1, 8, 9].

Isolation of Dipeginol (2). Total alkaloids were treated with acetone. The part of the alkaloid mixture that was insoluble in acetone (100 g) gave deoxypeganine perchlorate upon treatment with perchloric acid. The mother liquor was evaporated, basicified with NH_4OH, and treated with ether. The solid left after evaporation of the ether was ground with acetone. The part that was insoluble in acetone yielded peganine [10]. The part that was soluble in acetone was evaporated and ground several times with acetone—ethylacetate (1:1). The remaining solid was crystallized from CH_3OH. Yield 0.036 g of dipeginol as elongated prisms.

Dipegine (1): $C_{22}H_{20}N_4O$, mp 221-223°C (acetone). UV spectrum [CCl_4, λ_{max}, nm (log ε)]: 226 (4.48), 277 (4.14), 305 (4.05), 317 (3.89) nm. IR spectrum (KBr, ν_{max}, cm^{-1}): 1590, 1620, 1660. ^{1}H NMR ($CDCl_3$, 400 MHz) and ^{13}C NMR ($CDCl_3$, 100 MHz) (Tables 1 and 2). EIMS, *m/z* (I_{rel}): [M^+] 356, 185 (3), 171 (100).

Dipeginol (2): $C_{22}H_{20}N_4O_2$, mp 243°C. UV spectrum [CCl_4, λ_{max}, nm (log ε)]: 225 (4.46), 276 (4.11), 303 (4.04), 317 (3.82) nm. IR spectrum (KBr, ν_{max}, cm^{-1}): 1580, 1630, 1680 cm^{-1}. ^{1}H NMR ($CDCl_3$, 400 MHz) and ^{13}C NMR ($CDCl_3$, 100 MHz) (Tables 1 and 2). EIMS, *m/z*: 201, 171.

REFERENCES

1. B. Kh. Zharekeev, Kh. N. Khashimov, M. V. Telezhenetskaya, and S. Yu. Yunusov, *Khim. Prir. Soedin.*, 264 (1974).
2. Kh. N. Khashimov, M. V. Telezhenetskaya, and S. Yu. Yunusov, *Khim. Prir. Soedin.*, 456 (1969).
3. V. N. Plugar′, Ya. V. Rashkes, A. Karimov, M. V. Telezhenetskaya, and S. Yu. Yunusov, *Khim. Prir. Soedin.*, 68 (1983).
4. R. L. Vittorg and F. Gatta, *Gazz. Chim. Ital.*, **99**, 59 (1969).
5. S. Johne, B. Jung, and D. Groger, *J. Prakt. Chem.*, **319**, 919 (1977).
6. J. C. William, J. J. Steven, and D. G. Richard, *Magn. Res. Chem.*, **23**, 335 (1985).
7. K. Pandita, M. S. Bhatia, R. K. Thappa, S. G. Agarwal, K. L. Dhar, and C. K. Atal, *Planta Med.*, **48**, 81 (1983).
8. Kh. N. Khashimov, M. V. Telezhenetskaya, Ya. V. Rashkes, and S. Yu. Yunusov, *Khim. Prir. Soedin.*, 453 (1970).
9. B. Kh. Zharekeev, M. V. Telezhenetskaya, and S. Yu. Yunusov, *Khim. Prir. Soedin.*, 279 (1973).
10. R. R. Arndt, S. H. Eggers, and A. Jordan, *Tetrahedron*, **23**, 3521 (1967).