Chemistry of Natural Compounds, Vol. 34, No. 3, 1998

STRUCTURE OF MUCROLIDIN. ANALYSIS OF THE INFLUENCE OF SOME OXYGEN-CONTAINING SUBSTITUENTS ON THE CHEMICAL SHIFTS OF THE PROTONS OF THE ANGULAR METHYL OF SESQUITERPENOIDS WITH THE EUDESMANE TYPE OF SKELETON

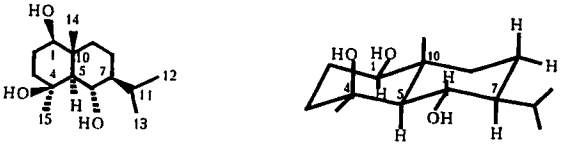
M. B. Izbosarov,[a] I. M. Yusupova,[b] B. Kh. Abduazimov,[a] B. Tashkhodzhaev,[b] A. Vdovin,[b] and N. D. Abdullaev[b]

UDC 547.992:547.37+548.737

A new eudesmanol, mucrolidin has been isolated from the epigeal part of Tanacetopsis mucronata. Its IR, mass, and PMR spectral characteristics have been investigated. Its composition has been established unambiguously by x-ray structural analysis and its spatial structure as 1,4,6-trihydroxy-1α,5α,6β,7α(H)-eudesmane. The interrelationship of the mucrolidin molecules in crystal packing is considered. With this sesquiterpenoid as an example, and using the characteristics of the PMR spectra of related compounds, the influence of some oxygen-containing functional groups at C-1, C-4, and C-6 on the chemical shifts of the protons of the methyl at C-10 has been analyzed.

Continuing an investigation of the plant *Tanacetopsis mucronata* (Regel et Schmalh.) S. Kovalesk. [1], we have isolated a new eudesmanol with mp 214-216°C, which has been called mucrolidin (**1**). The mass spectrum of mucrolidin revealed the peak of the molecular ion with *m/z* 256 (M^+, $C_{15}H_{28}O_3$) and the peaks of ions with *m/z* 238 (M^+-H_2O), 220 (M^+-2H_2O), and 202 (M^+-3H_2O) due to the ejection of water molecules, and also fragments characteristic for the breakdown of eudesmanolides under electron impact (see the Experimental section). In the PMR spectrum we observed doublet signals at 0.82 and 0.89 ppm with the SSCCs $^3J_{11,23} = {}^3J_{11,13} = 7$ Hz, which is characteristic for the methyl protons of an isopropyl group at C-7 in the eudesmane sesquiterpenoids [2-4]. The assignment of the signals of the other protons in the PMR spectrum was made with the aid of the double-resonance method and also by comparing the spectral characteristics of (**1**) with those of known compounds (Table 1).

The resonance signal of the H-6 proton, located geminally to the C-6 hydroxy group, appeared in the form of a doublet of doublets, each with $^3J = 9$ Hz, which is due to its *trans*-diaxial spin-spin interaction with the H-5 and H-7 protons. This indicates a possible α-orientation of H-5 and β-orientation of H-6. The vicinal SSCCs of H-1 with the protons at C-2 are 11 and 4 Hz, which shows the equatorial orientation of the hydroxy group at C-1. Assuming the chair-chair conformation frequently found for eudesmanolides with a β-oriented axial angular methyl group and considering the equatorial arrangement of three hydroxy groups at C-1 and C-6, it is possible to propose structure (**1a**) for mucrolidin.



1a

a) Pharmaceutical Institute, Tashkent-15, fax (2712) 56 45 04. b) Institute of the Chemistry of Plant Substances, Academy of Sciences of the Republic of Uzbekistan, Tashkent, fax (3712) 40 64 75. Translated from Khimiya Prirodnykh Soedinenii, No. 3, pp. 320-326, May-June, 1998. Original article submitted July 7, 1997.

0009-3130/98/3403-0289$20.00

TABLE 1. Characteristics of the PMR Spectra of Mucrolidin (**1**) and of Compounds (**2-8**) ($CDCl_3$, δ, ppm, J, Hz)

H	1	2 [13]	2a [14]	2b [14]	2c [14]	3 [15]	4 [16]	5 [17]	6 [15]	7 [15]	8 [5]
1	3.17dd (4,11)	3.25dd (5,11)	3.25 dd (5,11)	3.30 dd (5,11)	3.42dd (5,11)	3.52 ddd (4.5, 4.5,12)	3.41 dd (5,10)	3.48 dd (4.8, 11)	3.59 br.d (12)	3.49 br.d (10)	3.44 dd (4, 11)
5	1.77 br.d (10)				2.10 m	2.21 br.d (11)	1.72 d (11.5)	1.90 br.d (9.8)	2.24 br.d (10.5)	1.91 d (10)	
6	3.87 dd (9, 9)	3.73 dd (10, 10)	4.25 m ($W_{1/2}$=7)	4.32 br.s	4.12 dd (5, 11)	4.11 dd (11, 11)	4.13 dd (10, 11.5)	4.04 br.t (9.8, 10)	5.57 dd (10.5, 10.5)	5.76 dd (10, 10)	5.03 dd (9.3, 11)
7					1.75 m	2.84 dddd (3, 11, 11, 3)	1.5–1.8 m	2.40 ddd (3, 10, 11.5)	2.81 dddd (3, 3, 10.5, 10.5)	2.89 dddd (3, 3, 10, 12)	3.34 dt (4.7, 9.3, 9.4)
11					2.0 m	–	2.30 dq (7, 12.5)	–	–	–	–
12	0.82 d (7)	0.90 d (7)	0.95 d (6)	0.93 br.s	0.92 d (7.5)	–	–	–	–	–	–
13	0.89 d (7)	1.00 d (7)	0.95 d (6)	0.96 br.s	1.10 d (7.5)	6.15 d (3)	1.22 d (7)	5.94 dd (1, 3)	6.11 d (3)	6.09 d (3)	1.60 s
13′	–	–	–	–	–	5.53 d (3)	–	5.88 dd (1, 3)	5.38 d (3)	5.28 d (3)	
14	1.18 s	0.75 s	0.90 s	0.91 s	0.77 s	0.86 s	0.98 s	0.75 s	0.87 s	1.05 s	0.75 s
15	1.41 s	5.03 br.s	5.0 br.s	5.01 d (1.5)	5.0 d (1.5)	5 br.s	1.34 s	4.91 d (1.1)	4.86 br.s	1.27 s	5.05 b
15′	–	4.76 br.s	4.86 br.s	4.90 d (1.5)	4.76 d (1.5)	4.86 br.s	–	4.78 d (1.1)	4.60 br.s	–	4.95 b

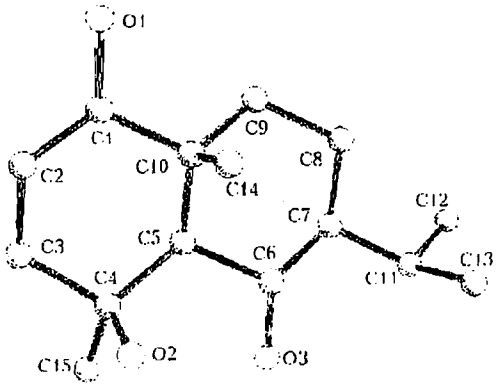
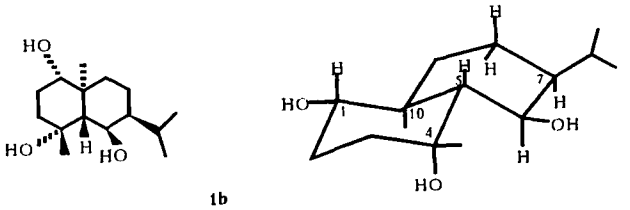


Fig. 1. Spatial structure of mucrolidin.

A high value of the SSCC between the H-5, H-6, and H-7 protons does not always determine their relative orientation unambiguously. With a different configuration of the C-5, C-6, and C-10 asymmetric centers and the realization of a boat conformation for ring ***B*** (**1b**), these protons may also interact with the appearance of a ^3J value of ~10 Hz [5]. Therefore, it is possible to propose as an alternative for mucrolidin the structure (**1b**) with a chair-boat conformation.



1b

The choice between the two probable structures was made on the basis of the results of an x-ray structural study. The analysis performed permitted the structure of mucrolidin to be determined unambiguously as (**1a**). An analysis of the intracyclic torsion angles and calculation of the asymmetry parameters showed that ring ***A*** approximates to an ideal chair form, $C_s(\Delta C_s(1.4) = 0.55°)$ [6], the C-1 and C-4 atoms departing from the plane of the other four atoms by −0.70 and 0.56 Å, respectively. Ring ***B*** also has a chair form but with a slight distortion of the ideal form ($\Delta C_s(9.6) = 4.47°$), with the C-9 and C-6 atoms departing from the plane of the other four atoms by 0.65 and 0.66 Å, respectively.

The linkage of rings ***A/B*** is *trans*, which is characterized by the torsion angle C-14−C-10−C-5−H-5 = 175.1°. No anomalous deviations are observed in the values of the valence angles: they agree well with the adopted standard values [7]. The presence of three hydroxy groups in the molecule favors the formation of intermolecular hydrogen bonds in the crystal. An analysis of intermolecular contacts shows that all the hydroxy groups participate in hydrogen bonds of the O−H···O type, as is witnessed by the distances O1···O2 (2.90 Å), O3···O2 (2.77 Å), and O3···O1 (2.73 Å) (Fig. 2). In the crystal packing, the molecules of mucrolidin form two-dimensional networks about the y, z axes transformed by 2_1 screw axes.

Thus, in mucrolidin rings ***A*** and ***B*** are *trans*-linked, the methyl at C-10 is oriented β-axially, and the hydroxy groups at C-4 and C-6 are oriented *cis*-diaxially and *trans*-axially-equatorially, respectively, relative to the C-10 methyl.

In the PMR spectra of eudesmanolides having no carbonyl groups or double bonds in the β- or γ-positions relative to C-14 but having hydroxy groups at C-1 and C-6, the signals of the angular methyl at C-10 fall between 0.7 and 0.9 ppm [1, 2, 8, 9]. In the case of mucrolidin, which contains three hydroxy groups, at C-1, C-4, and C-6, the protons of this methyl group resonate at 1.18 ppm.

To explain the downfield shift, we considered it necessary to make an analysis of the PMR characteristics of mucrolidin and related compounds in order to evaluate the influence of some oxygen-containing functional groups located in the β- and γ-positions to C-14 on the chemical shifts of the protons of the angular methyl group. In this case, the downfield shift of the signal of the protons of the angular methyl group is obviously due to the identical orientations of the hydroxy group at C-4 and the methyl group at C-10, i.e., their spatial propinquity, which leads to an appreciable descreening of the protons of the latter [10, 11]. The descreening influence of the β-equatorial hydroxy group at C-1 is considerably smaller [2, 5, 10-12].

When methyl and hydroxy groups are present at C-4 in place of the exomethylene group, in the PMR spectrum a downfield shift of the signal of the protons of the angular methyl group by 0.05-0.2 ppm is observed (compare compounds (**2**) and (**1**), (**3**) and (**4**), and (**6**) and (**7**), $\Delta\delta$ = 0.43, 0.12, and 0.18, respectively, see Table 1). In the case of the mutual *cis*-di-

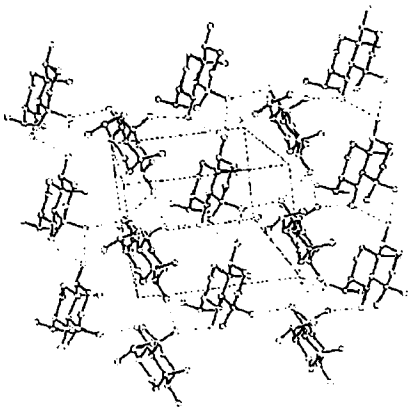
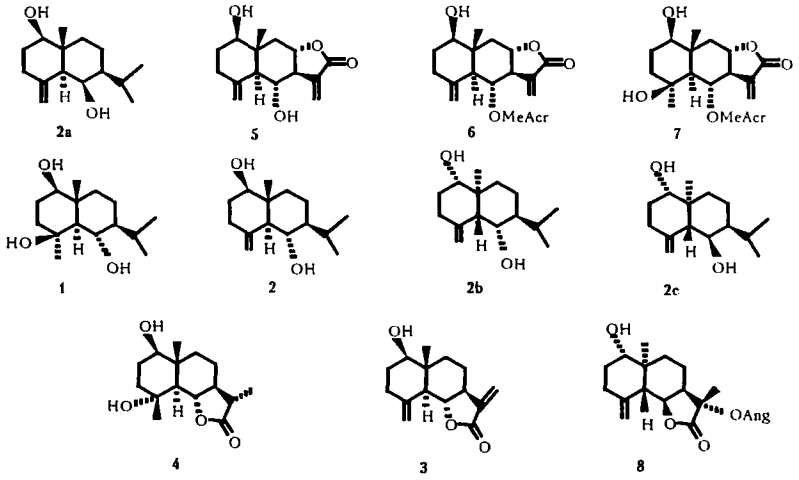


Fig. 2. Molecular packing of mucrolidin.

axial orientation of the hydroxyl at C-4 and the angular methyl group at C-10, the signal of the latter is found in a weaker field. In the case of *cis*-diaxial orientation of the angular methyl and the C-6 hydroxyl, the corresponding protons resonate in a weaker field than when they have the *trans*-axial-equatorial orientation (compare compounds (**2a**) and (**2**), and also (**2b**) and (**2c**), see Table 1).



When an acyl residue or a lactone ring is present in place of the α-oriented equatorial hydroxy group at C-6, a downfield shift of the signal of the protons of the methyl group is observed in the PMR spectrum (compare compounds (**2**) and (**3**), and (**5**) and (**6**), $\Delta\delta = 0.11$ and 0.12, respectively), and, in the case of a β-equatorially orientated hydroxy group, a small downfield shift of the H_3C-14 signal (see Table 1; compare (**2c**) and (**8**)). Consequently, the downfield shift of the signal of the angular methyl in mucrolidin can be explained by the combined influence of the three hydroxy groups: β-axial at C-4, β-equatorial at C-1, and α-equatorial at C-6.

Thus, it may be concluded that if in the structure of a eudesmolide the orientation of hydroxy groups at C-4, C-6, and C-8 has been unambiguously established, then from the chemical shift of its signal it is possible to determine the relative orientation of the angular methyl.

EXPERIMENTAL

IR spectra were taken on a UR-20 instrument (KBr), the mass spectrum on an MKh-1310 mass spectrometer, and PMR spectra on a TESLA BS-567A (100 MHz) spectrometer with $CDCl_3$ as solvent, 0-HMDS, δ scale. Type KSK silica gel was used for column chromatography. Systems: hexane–chloroform (1:1); chloroform; chloroform–alcohol (1:7). R_f values were

TABLE 2. Coordinates ($\times 10^4$) of the Nonhydrogen Atoms in the Mucrolidin Molecule

Atom	x	y	z	U
O1	-6858(7)	6938(5)	2107(3)	74(2)
O2	-445(6)	7369(4)	2675(2)	55(2)
O3	997(6)	4838(4)	1821(2)	52(2)
C1	-5226(10)	6373(7)	2355(4)	55(3)
C2	-4609(10)	7102(8)	2961(4)	67(3)
C3	-2978(11)	6432(8)	3266(4)	65(3)
C4	-1390(10)	6160(7)	2793(4)	49(3)
C5	-2072(9)	5579(6)	2129(3)	43(2)
C6	-635(9)	5412(7)	1584(4)	48(3)
C7	-1312(10)	4649(8)	999(4)	59(3)
C8	-3029(11)	5269(9)	712(4)	68(3)
C9	-4441(10)	5504(8)	1237(4)	61(3)
C10	-3751(10)	6316(6)	1825(4)	49(2)
C11	-120(11)	5264(7)	3178(4)	63(3)
C12	-3318(10)	7687(7)	1582(4)	65(3)
C13	159(15)	4381(11)	484(4)	93(4)
C14	-390(17)	3329(12)	-3(5)	176(9)
C15	764(16)	5562(13)	114(5)	157(7)

TABLE 3. Bond Lengths r (Å) and Valence Angles ω (degrees) in the Mucrolidin Molecule

Bond	r	Angle	ω	Angle	ω
C1–C2	1.50(1)	C2–C1–C10	112.1(6)	C8–C7–C13	113.7(7)
C1–C10	1.52(1)	C2–C1–O1	109.6(6)	C9–C8–C7	111.6(6)
C2–C3	1.51(1)	C10–C1–O1	111.4(6)	C10–C9–C8	113.9(6)
C3–C4	1.53(1)	C3–C2–C1	110.0(6)	C1–C10–C5	107.4(6)
C4–C5	1.55(1)	C2–C3–C4	115.5(7)	C1–C10–C12	109.7(6)
C4–C11	1.52(1)	C3–C4–C5	111.3(6)	C9–C10–C1	109.3(6)
C5–C6	1.53(1)	C3–C4–C11	104.9(6)	C9–C10–C5	107.3(5)
C5–C10	1.57(1)	O2–C4–C5	110.2(5)	C9–C10–C12	108.9(6)
C6–C7	1.50(1)	O2–C4–C3	108.0(6)	C5–C10–C12	114.1(6)
C7–C8	1.53(1)	O2–C4–C11	108.2(6)	C7–C13–C14	112.5(9)
C7–C13	1.52(1)	C11–C4–C5	113.9(6)	C7–C13–C15	113.8(9)
C8–C9	1.50(1)	C4–C5–C6	116.2(5)	C14–C13–C15	109.7(8)
C9–C10	1.54(1)	C4–C5–C10	113.9(6)		
C10–C12	1.53(1)	C6–C5–C10	108.2(6)		
C13–C14	1.52(2)	C5–C6–C7	113.3(6)		
C13–C15	1.50(2)	O3–C6–C7	109.1(6)		
O1–C1	1.423(9)	O3–C6–C5	112.5(6)		
O2–C4	1.446(8)	C6–C7–C13	113.3(7)		
O3–C6	1.419(8)	C8–C7–C6	110.6(7)		

obtained on Silufol UV-254 plates in the systems: hexane–ethyl acetate (3:2) and benzene–alcohol (4:1), with a 1% solution of vanillin in concentrated sulfuric acid as the revealing agent.

Isolation of Mucrolidin. Fractions containing mainly tavulin and several unknown substances were rechromatographed on KSK silica gel in a ratio of 50:1 [1]. Elution was conducted first with chloroform–hexane (1:1), and was then continued with chloroform (fractions 3-70). After this, the eluent was changed by the addition of alcohol (1:7). The eluates were collected in 150-ml fractions. By recrystallization, fractions 71-72 yielded a substance with mp 214-216°C (acetone–hexane), R_f 0.31 (benzene–alcohol (4:1)), mass spectrum, m/z, (I_{rel}, %): 256 (M^+) (0.94), 241 (41.8), 238 ($M^+ - H_2O$) (3.7), 224 (8.2), 223 (60.1), 220 ($M^+ - 2H_2O$) (9.4), 205 (20.6), 202 ($M^+ - 3H_2O$) (1.9), 187 (11.3), 181 (18), 180 (36), 177 (14.4), 167 (51), 149 (30), 138 (52), 137 (33), 123 (31), 101 (100), 95 (28).

X-Ray Structural Analysis. Crystals grown from solution (acetone–hexane) were investigated by the photo method. The unit cell parameters and the space group were determined from precession x-ray photographs and were refined on a Syntex $P2_1$ diffractometer; a = 7.337(3), b = 10.305(4), c = 20.189(6)Å, d_{calc} = 1.502 g/cm^3, space group $P2_12_12_1$, Z = 4. A three-dimensional set of intensities was obtained on the same diffractometer: $\theta/2\theta$ method of scanning using CuK_α radiation (graphite monochromator), $\sin\theta/\lambda < 0.55$, rate of scanning 11.72 deg/min, number of independent and nonzero reflections with $I > 2\sigma(I)$ 1285. Search for the structure was made by the SHELXS-86 program [18] (PC DOS version), where it was possible to find a model of the molecule in the automatic regime. The structure was refined by the method of least squares

successively in the isotropic–anisotropic approximation by the SHELXS-76 program [19]. The coordinates of the nonhydrogen atoms and bond lengths and valence angles are given in Tables 2 and 3.

REFERENCES

1. B. Kh. Abduazimov, B. Tashkhodzhaev, S. Nasirov, I. D. Sham'yanov, M. R. Yagudaev, V. M. Malikov, and S. N. Aminov, Khim. Prir. Soedin., 19 (1991).
2. A. Gareta-Granados, A. Martinez, F. Rivas, M. E. Onorato, and J. M. Arias, J. Nat. Prod., **53**, 436 (1990).
3. F. Bohlmann, M. Grenz, R. K. Gupta, A. K. Dhor, M. Ahmed, R. M. King, and H. Robinson, Phytochemistry, **19**, 2391 (1980).
4. F. Bohlmann, K.-H. Knoll, C. Zdero, P. K. Mahanta, M. Grenz, A. Suwita, D. Ehlers, N. Le Van, W.-R. Abraham, and A. A. Natu, Phytochemistry, **16**, 965 (1977).
5. M. Holub, M. Budesinsky, Z. Smitalova, D. Saman, and V. Rychlewska, Coll. Czech. Chem. Commun., **51**, 903 (1986).
6. W. L. Duax, C. Weers, and D. C. Robrer, in: Stereochemistry, N. L. Allinger (ed.), J. Wiley, New York, Vol. 9 (1976), p. 200.
7. F. N. Allen, O. Kennard, and D. G. Watson, J. Chem. Soc., Perkin Trans. II, No. 12, S1-S19 (1987).
8. Z. Samek, M. Holub, H. Grabarczyk, B. Drozda, and V. Herout, Coll. Czech. Chem. Commun., **38**, 1804 (1973).
9. A. G. Gonzalez, A. Galimdo, H. Mansilla, V. H. Kesternicn, J. A. Palenzuela, and M. L. Rodriguez, J. Nat. Prod., **53**, 461 (1990).
10. F. Bohlmann, G. S. Hirschmann, J. Jakupovic, R. M. King, and H. Robinson, Phytochemistry, **29**, 1989 (1984).
11. R. Mata, G. Delgado, and A. Romo de Vivar, Phytochemistry, **23**, 1665 (1984).
12. P. Sing, J. Jakupovic, F. Bohlmann, R. M. King, and H. Robinson, Phytochemistry, **24**, 2110 (1985).
13. F. Bohlmann, N. Ates (Goren), R. M. King, and H. Robinson, Phytochemistry, **22**, 1675 (1983).
14. A. Kamel, J. Nat. Prod., **58**, 428 (1995).
15. F. Bohlmann, J. Jakupovic, M. Ahmed, and A. Schuster, Phytochemistry, **22**, 1623 (1983).
16. J. F. Sanz, E. Falco, and J. A. Marco, J. Nat. Prod., **53**, 940 (1990).
17. M. Luz Cardona, I. Fernandes, B. Garcia, and J. R. Pedro, J. Nat. Prod., **53**, 1042 (1990).
18. G. M. Sheldrick, Acta Crystallogr., **A46**, 467 (1990).
19. G. M. Sheldrick, SHELX-76 Program for Crystal Structure Determination, Cambridge, England.