A New Irregular Trihydroxy Sesquiterpene from *Teucrium mascatense*

by **Liaqat Ali**[a]), **Imtiaz Hussain**[b]), **Tania Shamim Rizvi**[a]), **Abdul Latif Khan**[a]), **Shaukat Shujah**[a]), **Jawahar Mohammed Salim Al-Sahi**[c]), **Ahmed Al-Harrasi***[a])[c]), and **Javid Hussain***[a])[c])

[a]) UoN Chair of Oman's Medicinal Plants and Marine Natural Products, University of Nizwa, Birkat Al-Mouz, Nizwa-616, Oman

[b]) Department of Chemistry, Kohat University of Science and Technology, Kohat, Pakistan

[c]) Department of Biological Sciences and Chemistry, College of Arts and Sciences, University of Nizwa, Birkat Al-Mouz, Nizwa-616, Oman

(phone: +968-254-46705; fax: +968-254-46612; e-mail: javidhej@gmail.com; aharrasi@unizwa.edu.on)

A new trihydroxy sesquiterpene, *rel*-(1*R*,4a*R*,5*S*,6*S*,7*S*,8a*R*)-decahydro-6,8a-dimethyl-5-(propan-2-yl)naphthalene-1,6,7-triol (**1**), has been isolated as a result of the phytochemical investigation on the CH_2Cl_2 extract of *Teucrium mascatense*. The structure elucidation of the new constituent was carried out by the combined use of 1D- (^{1}H- and ^{13}C-NMR) and 2D-NMR (HMBC and HSQC) spectroscopic analysis, along with mass spectrometric techniques. In addition to the new constituent **1**, the known metabolite **2**, previously isolated from *Crataegus pinnatifida*, was also identified.

Introduction. – *Teucrium*, belonging to the family lamiaceae, is a medicinally important genus containing more than 300 species with a wide range of biological activities, including antioxidant, antimicrobial, anti-inflammatory, antispasmodic, and analgesic properties, *etc.* [1]. In folk medicine, *Teucrium* is reported for the treatment of diarrhea, cough, jaundice, and abdominal pain. The essential oil of various species is also reported to possess antimicrobial, antioxidant, and anticancer activities, in addition to be used as preservative ingredients in food and pharmaceutical industries [2][3]. *Teucrium mascatense* BOISS. is a perennial plant and is commonly used for the preparation of traditional medicines to treat several ailments, including diabetes, stomach ache, gastro-intestinal ailments, and inflammatory conditions [4]. It is an endemic plant to the Northern parts of Oman and found in the rocky hills above 1000 m high altitude. The leaves are boiled in water in combination with *Rhazya stricta*, *Fagonia indica*, myrrh, sea salt, and black salt, and the resulting concoction is used to treat abdominal colic, fever, and diabetes [4]. In continuation of our search for secondary metabolites from medicinal plants [5][6], the present study was conducted on the title plant, and the isolation and structure elucidation of one new and one known sesquiterpene is reported herein. Previously, a number of monoterpenes, sesquiterpenes, diterpenes, and triterpenes has been reported from various species of the genus [7]. However, to the best of our knowledge, this is the first report on the chemical constituents of *T. mascatense*.

Results and Discussion. – The dried and powdered material of *T. mascatense* was extracted with MeOH and then subjected to phytochemical investigations. The CH_2Cl_2-

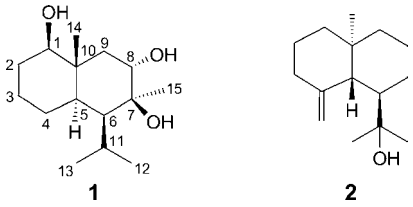

Fig. 1. *Structures of compounds* **1** *and* **2**

soluble fraction was chromatographed on a SiO_2 column to furnish sub-fractions F_1–F_{12}. *Fr.* F_5 was further purified through repeated CC to afford compounds **1** and **2** (*Fig. 1*). The chemical structure of the known metabolite **2** was confirmed by comparison of its spectroscopic data with published literature values [8].

Compound **1** was isolated from the CH_2Cl_2-soluble fraction of the crude MeOH extract in the form of a gummy colorless solid, and was identified as a terpenoid by displaying a pink colored spot on TLC when sprayed with ceric sulfate reagent followed by heating. The presence of OH groups was indicated by an IR band at 3475 cm^{-1}. The molecular formula was established as $C_{15}H_{28}O_3$ (two degrees of unsaturation) on the basis of a *pseudo*-molecular ion peak ($[M+H]^+$) in the HR-ESI-MS at *m*/*z* 257.2119 ($C_{15}H_{29}O_3^+$; calc. 257.2117).

The ^{1}H-NMR data (*Table*) of **1** indicated the presence of two tertiary Me (δ(H) 0.94 (*s*, Me(14)) and δ(H) 1.33 (*s*, Me(15))) and two secondary Me signals (δ(H) 0.93 (*d*, $J=6.6$, Me(12)) and δ(H) 1.09 (*d*, $J=6.6$, Me(13))). The O-bearing CH groups (H–C(1) and H–C(8)) appeared at δ(H) 3.33–3.34 (*m*) and δ(H) 4.35 (*dd*, $J=4.7$, 11.4), whereas sp^3 CH groups (H–C(5), H–C(6), and H–C(11)) were observed at δ(H) 1.58–1.60 (*m*), δ(H) 1.72–1.74 (*m*), and δ(H) 2.02–2.06 (*m*), respectively. The ^{13}C-NMR data and the DEPT experiments, coupled with the 2D-HMQC correlations, were in agreement with the above assignments and revealed the presence of four Me, four CH_2, five CH groups, as well as two quaternary C-atom signals (*Table*). These signals were resolved into two O-bearing CH groups ((C(1) at δ(C) 79.5) and (C(8) at δ(C) 73.8)), two sp^3 CH groups ((C(5) at δ(C) 47.5 and (C(6) at δ(C) 50.3)), two tertiary Me groups (δ(C) 13.9 (C(14))) and (δ(C) 23.9 (C(15))), and one ^{i}Pr group ((C(11) at δ(C) 25.2), (C(12) at δ(C) 22.4), and (C(13) at δ(C) 24.4)). The high field signal at δ(C) 45.5 (*Table*) was assigned to the quaternary C-atom (C(10)), whereas the O-bearing quaternary C-atom (C(7)) was observed at δ(C) 72.7. The CH_2 functionalities in the molecule were observed at δ(C) 35.6 (C(2)), δ(C) 22.9 (C(3)), δ(C) 28.1 (C(4)), and δ(C) 40.1 (C(9)). These assignments were also supported by the analysis of ^{1}H-NMR data (*Table*). The position of different functionalities was assigned on the basis of long-range interactions observed in HMBC experiments (*Fig. 2*). The HMBC cross-peaks of H–C(6) (δ(H) 1.72–1.74 (*m*)) with C(7) (δ(C) 72.7), C(5) (47.5), and C(11) (25.2) revealed the neighbourhood of H–C(6). The HMBC experiment also showed correlations between H–C(5) (δ(H) 1.58–1.60 (*m*)) and C(10) (δ(C) 45.5), C(6) (50.3), and C(4) (28.1). The Me(12) (δ(H) 0.93 (*d*, $J=6.6$)) and Me(13) (δ(H) 1.09 (*d*, $J=6.6$)) showed long-range interactions with C(6) (δ(C) 50.3) and C(11) (δ(C) 25.2)), thus confirming the position of the ^{i}Pr group. The H-atom geminal to the OH group (H–C(8) at δ(H) 4.35 (*dd*, $J=4.7$, 11.4)) showed cross-peaks with the quaternary

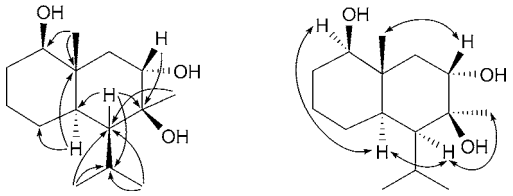

Fig. 2. *Key HMBC* (H → C) *and NOESY* (H ↔ H) *correlations of compound* **1**

Table. *^{13}C- and ^{1}H-NMR Data* (at 150 and 600 MHz, resp.; in $CDCl_3$) *and HMBCs of Compound* **1**. δ in ppm, J in Hz.

Position	δ(H)	δ(C)	HMBC
1	3.33 – 3.34 (*m*, 1 H)	79.5	C(2), C(10)
2	1.21 – 1.22 (*m*, 1 H), 1.47 – 1.48 (*m*, 1 H)	35.6	
3	1.52 – 1.53 (*m*, 1 H), 1.70 – 1.71 (*m*, 1 H)	22.9	
4	1.57 – 1.58 (*m*, 1 H), 1.67 – 1.68 (*m*, 1 H)	28.1	
5	1.58 – 1.60 (*m*, 1 H)	47.5	C(4), C(10)
6	1.72 – 1.74 (*m*, 1 H)	50.3	C(5), C(7), C(11)
7		72.7	
8	4.35 (*dd*, J = 4.7, 11.4)	73.8	C(7), C(9)
9	1.56 – 1.57 (*m*, 1 H), 1.74 – 1.75 (*m*, 1 H)	40.1	
10		45.5	
11	2.02 – 2.06 (*m*, 1 H)	25.2	C(5), C(6), C(12)
12	0.93 (*d*, J = 6.6)	22.4	C(6), C(11)
13	1.09 (*d*, J = 6.6)	24.4	C(6), C(11)
14	0.94 (*s*, 3 H)	13.9	C(1), C(5), C(9), C(10)
15	1.33 (*s*, 3 H)	23.9	C(6), C(7), C(8)

C(7) (δ(C) 72.7) and CH_2(9) (δ(C) 40.1), whereas the second H-atom geminal to a OH group (H–C(1) at δ(H) 3.33 – 3.34 (*m*)) showed interactions with the quaternary C(10) (δ(C) 45.5) and CH_2(2) (δ(C) 35.6).

The relative configuration of **1** was deduced by an analysis of the NOESY spectrum. When Me(14) is assummed to be in a β-orientation, the OH group at C(8) must be α-oriented because of a strong NOESY corelation between Me(14) and H–C(8), whereas the OH groups at C(1), C(7), and the ^{i}Pr group at C(6) were determined to be β-oriented due to the absence of correlations (*Fig. 2*) between Me(14) and H–C(1), H–C(6), and Me(15). The observed correlation between H–C(5) and HO–C(8) also confirmed them to be α-oriented, whereas the absence of a correlation with Me(14) suggested the opposite configuration at the ring junction in the decalin system. NOESY correlations were also observed between H–C(1) and H–C(5), H–C(6), Me(15), and thus indicated them to be on the same face of the molecule with α-orientation. Thus, on the basis of all of the above evidence, the structure of compound **1** was unambiguously established (*Fig. 1*) and named as *rel*-(1*R*,4a*R*,5*S*,6*S*,7*S*,8a*R*)-decahydro-6,8a-dimethyl-5-(propan-2-yl)naphthalene-1,6,7-triol (**1**). However, it has to be noted, that the absolute configuration of (+)-**1** remains to be established.

Experimental Part

General. Column chromatography (CC): silica gel *60* (SiO_2, 0.063 – 0.200 mm, 70 – 230 mesh ASTM; *Merck*). TLC: precoated silica gel plates (*G60* F_{254}; *Merck*), resp. IR Spectra: *Bruker-ATR* spectrophotometer; $\tilde{\nu}$ in cm^{-1}. ^1H- and ^{13}C-NMR spectra: *Bruker Avance 600* (600 (^1H) and 150 (^{13}C) MHz) spectrometer; δ in ppm rel. to residual $CDCl_3/CHCl_3$ (δ(H) 7.24, δ(C) 77.0) as internal standard, *J* in Hz. HR-ESI-MS: *QSTAR XL* mass spectrometer (*Applied Biosystem*); in *m*/*z*.

Plant Material. The whole plant of *T. mascatense* BOISS. was collected from the mountains of Al-Jabel Al-Akhdar, Oman, in 2013, and was identified by a plant taxonomist at the Department of Biological Sciences and Chemistry, University of Nizwa, Nizwa, Oman. The voucher specimen has been deposited with the Herbarium of the above Department.

Extraction and Isolation. The dried and powdered plant material of *T. mascatense* (3.5 kg) was extracted with MeOH at r.t. The crude MeOH extract (104.8 g) was partitioned into hexane, CH_2Cl_2, AcOEt, and BuOH fractions. The CH_2Cl_2 fraction (21.4 g) was subjected to SiO_2 CC (hexane, hexane/CH_2Cl_2, and CH_2Cl_2/MeOH) by gradual increase of the mobile phase polarity. Twelve sub-fractions (*Frs.* $F_1 - F_{12}$) were obtained. *Fr.* F_5 was obtained at 40% CH_2Cl_2/hexane and was then subjected to further purifications. Compound **1** (4.4 mg) was obtained as a gummy solid after purification through repeated chromatographic separations. The semi-pure compounds were subjected to further purification through prep. TLC plates, and the known compound **2** (3.8 mg) was obtained pure by elution with 55% CH_2Cl_2/hexane.

rel-*(1*R*,4a*R*,5*S*,6*S*,7*S*,8a*R*)-Decahydro-6,8a-dimethyl-5-(propan-2-yl)naphthalene-1,6,7-triol* (**1**). Gummy colorless solid. $[\alpha]_D^{25} = +14.5$ ($c = 0.03$, MeOH). IR: (MeOH): 3475. ^1H-NMR (600 MHz, $CDCl_3$): see *Table*. ^{13}C-NMR (150 MHz, $CDCl_3$): see *Table*. HR-ESI-MS: 257.2119 ($C_{15}H_{29}O_3^+$, $[M+H]^+$; calc. 257.2117).

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REFERENCES

[1] H. Henchiri, B. Bodo, A. Deville, L. Dubost, L. Zourgui, A. Raies, P. Grellier, L. Mambu, *Phytochemistry* **2009**, *70*, 1435.
[2] S. M. M. Shah, F. Ullah, S. M. H. Shah, M. Zahoor, A. Sadiq, *BMC Comp. Alt. Med.* **2012**, *12*, 244.
[3] H. Amiri, *Iran. J. Pharm. Res.* **2010**, *9*, 417.
[4] S. A. Ghazanfar, *J. Biogeogr.* **1991**, *18*, 299.
[5] L. Ali, S. Ali, J. Hussain, A. Al-Harrasi, A. L. Khan, S. T. H. Shah, T. S. Rizvi, *Helv. Chim. Acta* **2014**, *97*, 556.
[6] L. Ali, J. Hussain, A. Al-Rawahi, A. Al-Harrasi, *Rec. Nat. Prod.* **2014**, *8*, 407.
[7] F. Piozzi, M. Bruno, S. Rosselli, A. Maggio, *Heterocycles* **2005**, *65*, 1221.
[8] C.-C. Zhou, X.-X. Huang, P.-Y. Gao, F.-F. Li, D.-M. Li, L.-Z. Li, S.-J. Song, *J. Asian. Nat. Prod. Res.* **2014**, *16*, 169.

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