(1970).
11) F.M. Shafik and F.S.G. Soliman, **Pharmazie**, 29, 247 (1974).
12) F.M. Shafik and F.S.G. Soliman, ***ibid.***, 29, 290 (1974).
13) H.G. Garg, **J. Med. Chem.**, 15, 446 (1972).

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Phytochemistry of *Eryngium creticum*

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Abstract ❑ The isolation and identification of 9 compounds from *Eryngium creticum*, which grows wildly in Jordan, are described. The isolated compounds are deltion, marmesin, quercitol, 3-(β-D-glucopyranosyloxymethyl)-2,4,4-trimethyl-2,5-cyclohexadien-1-one, 1-β-D-glucopyranosyloxy-3-methoxy-5-hydroxybenzene, β-sitosterol, β-sitosterol-β-D-glucopyranose, mannitol and dulcitol.

Keyphrases ❑ *Eryngium creticum*, Umbelliferae, coumarin, sitosterol, glycoside.

Eryngium creticum Lam. (Umbelliferae) is a perennial or biennial, glaucous, globrous herb with a height of up to 20-50 cm (1), and the plant is widely distributed in Jordan (2). There is a current scientific interest in the plant because of its traditional use as diuretic and laxative (1), and as a remedy for scorpion and snake bites (3,4). Upon testing the efficiency of ***E. creticum*** aqueous extract in combination with the ***Leiurus quinquestriatus*** scorpion venom, the extract was found to prolong the life saving period in the tested animals from 20 minutes to 8 hours (3), and in combination with ***Cerastus*** snake venom from 12 to 72 hours (4). The aqueous extract of ***E. creticum*** caused a dose-related inhibition of tracheal and jejunal contraction induced by the ***L. quinquestriatus*** scorpion venom and inhibited the spontaneous movements of the jejunum (5).

The genus ***Eryngium*** is known to contain terpene aldehyde esters (6), acetylenes (7), flavonoids (8,9), coumarin derivatives (10,11) and monoterpene glycosides (12).

Experimental:

Melting points were determined on Stuart Scientific melting point apparatus and are uncorrected. IR spectra (KBr) were determined on a JASCO IR-810 spectrometer. UV spectra were determined on a Unicam 810 Kontron Spectrophotometer and PMR spectra on JEOL GX-270 spectrometer using TMS as internal standard. Low resolution MS spectra were recorded on a quadruple instrument Finnigan MAT 112, 70eV. Sephadex LH-20-100 (Fluka) was used for gel chromatography. Silica gel (Kieselgel 60-Merck) was used for CC, while silica gel (Kieselgel 60-F_{254}, Merck) was used for TLC. Nonsugar compounds were analysed by TLC using $(Me)_2CO$- $CHCl_3$-MeOH (7:9:1) and sprayed with 1% vanillin-HCl reagent. Sugars were analyzed by PC (Whatmann #1) with EtOAc-n.BuOH-H_2O-AcOH (6:8:8:5) and visualized by Tollen's reagent. D-Mannitol and dulcitol (Fluka) were used for direct comparison with the isolated compounds. Anhyd. Na_2SO_4 was routinely used for drying solvents, and all solvents were evaporated under reduced pressure at 40°C.

Plant material:

Plant material used in this study was collected from Mafraq area, 45 kilometers north of Amman, in June 1990 and identified by professor Dr. Al-Eisawi, Plant Taxonomist, Department of Biological Sciences, University of Jordan. A herbarium specimen was deposited at the Faculty of Pharmacy, University of Jordan, Amman, Jordan.

Extraction and fractionation:

Powdered dried roots of ***E. creticum*** (12.1 kg) were extracted by percolation with cold MeOH (35.6 L) and the solvent was evaporated to leave a residue (516.3 g). The residue was suspended in H_2O (1.75 L) and first extracted with Et_2O (3 X 2 L; 74.94 g; Fraction A); then with $CHCl_3$ (3 X 1.5 L; 1.64 g; Fraction B). Fractions A and B were subsequently combined because of TLC similarity and named (Fraction A). Percolation with n-BuOH (3 X 3 L) gave 140.5 g (Fraction C). The water-soluble fraction was lyophilized (163.4 g; Fraction D).

Chromatography of fraction A:

Fraction A was dissolved in light petr.-EtOAc (7:3) (35ml) and chromatographed over a column of silica gel (335 g) (column A) in the same solvent. Elution with light petr.-EtOAc mixtures followed by $CHCl_3$ and $CHCl_3$-MeOH

mixtures afforded various fractions which were collected (200ml) and combined according to TLC analysis. Column chromatographic fractionation resulted in the isolation of deltoin **(I)** (12 mg) (0.00001% yield), mp 104-105°C; $[\alpha]_D^{23}$-51° ($CHCl_3$) (11,13); marmesin **(II)** (9 mg) (0.00007% yield), mp 188-189°C; $[\alpha]_D^{23}$+ 26.4°C ($CHCl_3$) (11,13) and quercitol **(III)** (25.4 mg) (0.0002% yield), mp 234°C (14).

Chromatography of fraction C:

Fraction C was mixed with silica gel (30g) and chromatographed over a column of silica gel (430g) (column B) in $CHCl_3$. Elution with $CHCl_3$ followed by $CHCl_3$-MeOH mixtures and $CHCl_3$-MeOH-H_2O mixtures afforded various fractions which were collected (200ml) and combind according to TLC analysis.

3-(β-D-Glucopyranosyloxymethyl)-2,4,4-trimethyl-2, 5-cyclohexadien-1-one (IV): Elution of the column (column B) with $CHCl_3$-MeOH (95:5) (1.2 L) and with $CHCl_3$-MeOH (92:8) (0.8 L) afforded an amorphous residue **IV** (22.7 mg) (0.00019% yield), after freeze-drying. IR $\sqrt{}$ (cm^{-1}): 3385, 1665, 1620, 835. PMR ($CDCl_3$), δ 1.95 (s, 3H, Me at C-2), 1.28 and 1.31 (2s, 2 X 3H, 2 X gem. Me at C-4), 6.94 (d, 1H, J = 9.7 Hz, H-5), 6.14 (d, 1H, J = 9.7 Hz, H-6), 4.32 and 4.75 (2d, 2H, J gem= 10.9 Hz, CH_2 at C-3), 4.37 (d, 1H, J = 7.6 Hz, H-1') 3.89-3.16 (m, H-sugar). Eims m/z: (rel. int. %): M^+ 328 (6), 166 (100) 149 (40), 121 (19.8), 109 (34.2) (12).

1-β-D-Glucopyranosyloxy-3-methoxy-5-hydroxybenzene (V): Further elution of the column (column B) with $CHCl_3$-MeOH (92:2) (0.6 L) and $CHCl_3$-MeOH (88:12) (1.2 L) yielded compound V as a white substance (14.3 mg, 0.00012% yield) after freeze-drying. $[\alpha]^{23}$ - 66.4° (MeOH); UV λmax (MeOH) (log∈): 224 (2.79), 269 (2.21), 271 (sh) (2.16) nm. PMR ($CDCl_3$) δ 6.16 (t, 1H, J = 2.12 Hz, H-2), 6.14 (t, 1H, J = 2.12 Hz, H-6), 6.02 (t, 1H, J = 2.12 Hz, H-4), 3.75 (s, 3H, MeO), 4.91 (d, 1H, J = 7.4 Hz, H-1') 3.91-3.34 (m, H-sugar). Eims m/z: rel. int. %): M^+ 302(4), 140(100), 111(41) (15).

Continued elution of column B with $CHCl_3$-MeOH (7:3) (1.6 L) and with $CHCl_3$-MeOH (6.4) (1:8 L) gave β-sistosterol (43.1 mg, 0.00035 % yield) while elution with $CHCl_3$-MeOH (1:1) (1:2 L) and $CHCl_3$-MeOH (3:7) (0.8 L) yielded β-sitosterol-β-D-glucopyranose (28.7 mg, 0.00023% yield) (16).

D-Mannitol (VI): Continued elution of the column (column B) with $CHCl_3$-MeOH (2:8) (1.2 L); MeOH (1 L) and with MeOH-H_2O (1:9) (1.8 L) yielded white crystals of D-mannitol **(VI)** (16.8 g, 0.138% yield), mp 167°C(17).

Chromatography of Fraction D: Fraction D (163.4 g) was chromatographed over Sephadex gel column (350g) using MeOH-H_2O (8:2) as eluent; 50 fractions were collected (50ml, each) and combined according to PC analysis. Fraction 1-38 gave D-mannitol **(VI)** (72.8g, 0.601% yield), and fractions 39-50 gave dulcitol **(VII)** (2.6g, 0.021% yield), mp 188°C (17).

The isolated materials were identified by mp, m.mp, IR, MS, PMR, $[\alpha]_D$, and direct co-chromatographic comparison with authentic samples and literature data.

RESULTS and DISCUSSION

Investigation of the roots of ***Eryngium creticum***, which grows wildly in Jordan and used in folk medicine as a diuretic and for the treatment of scorpion bites, led to the isolation and characterization of two coumarins, a quercitol, a monoterpene glycoside, a phloroglucinol glycoside, β-sitosterol and its glycoside and two sugars, mannitol and dulcitol.

Deltoin **(I)** and marmesin **(II)** were identified by comparison with literature data (11,13). These two compounds have been isolated from ***E. ilicifolium*** (11). The PMR spectrum of **IV** showed three methyl groups; one appeared as a singlet at δ 1.95 and attributed to an unsaturated carbon atom at C-2. The other two methyl groups at δ 1.28 and 1.31 were typical gem-dimethyl and assigned to C-4. The spectrum also revealed the presence of two olefinic protons, AB-system (J= 9.7 Hz) ortho-coupled at δ 6.14 and 6.94 which were assigned