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FLAVONOIDS FROM *LOTUS CRETICUS*

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Key Word Index—*Lotus creticus*; Leguminosae; isoflavonoids; flavanone; lupinalbin A–C; euchrestaflavanone A, wighteone methyl ether.

Abstract—One flavanone, euchrestaflavanone A and four isoflavonoids (lupinalbin A–C, and a previously unreported 4′-methyl ether of wighteone) were isolated from *Lotus creticus*. The structures were elucidated by spectroscopy.

INTRODUCTION

Chemical investigations of the genus *Lotus* have not been extensive, although flavones and isoflavanes have been reported from three species [1–3]. We report here the isolation of a flavanone as well as four isoflavonoids from *Lotus creticus* L.

RESULTS AND DISCUSSION

An aqueous ethanol extract of the roots of *Lotus creticus* yielded the previously reported flavanone euchrestaflavanone A (**3**) and three recently described isoflavonoids, namely, lupinalbin A–C (**5**, **2**, **4**), as well as the previously unreported 4′-methyl ether of wighteone (**1**). This is only the second report of **2**, **4** and **5**.

In the UV spectrum of **1**, Band II (268 nm) and Band I (330 nm) exhibited a relative intensity of *ca* 2 to 1. These data as well as an ^{1}H NMR signal at δ8.35, which is characteristic for H-2 in isoflavones, together indicated an isoflavone skeleton for **1** [4]. The ^{1}H NMR spectrum of **1** also indicated that the B-ring of **1** was 4′-substituted (δ7.49, 2H, *d*, J = 8 Hz, H-2′, 6′; 7.02, 2H, *d*, J = 8 Hz, H-3′, 5′), and that the A-ring was either 5,6,7 or 5,7,8 trisubstituted (δ6.46, 1H, *s*, H-6 or H-8). Other signals indicated the presence of one methoxyl group (δ3.79, 3H, *s*) and a prenyl sidechain (δ3.21, 2H, *br d*, J = 7 Hz, 5.18, 1H, *br t*, J = 7 Hz, 1.62, 3H, *br s*, and 1.73, 3H, *br s*). A 2D COSY NMR experiment recorded at 500 MHz established the above signal assignments. The EIMS of **1** exhibited a molecular ion at *m/z* 352 ($C_{21}H_{20}O_5$) in agreement with one methoxyl, one prenyl and two hydroxyl groups for **1**. Fragments at *m/z* 297 $[M-C_4H_7]^+$, 165 $[A_1-C_4H_7]^+$ and 69 $[prenyl]^+$ further supported the presence of a prenyl group (for MS nomenclature see [5]). The NOE experiment of **1** recorded at 500 MHz in DMSO-d_6 confirmed a 4′-methoxyl group, that is, irradiation of the methoxyl signal at δ3.79 enhanced the signals for H-3′ and 5′ at δ7.02 but not the singlet signal at δ6.46. The 11 nm bathochromic shift of Band II ($AlCl_3$–HCl–MeOH) relative to the spectrum in MeOH suggested a C-5 hydroxyl group in **1** [4]. The MS fragment at *m/z* 165 $[A_1-55]^+$ supported the prenyl group being attached to the A-ring, and the singlet signal at δ6.46 (a chemical shift typical for H-8) also suggested that the prenyl group is at the C-6 position. This assignment was supported by the tetramethylsilane-derivatization results, namely, the C-5 hydroxyl group could not be derivatized even standing overnight with the reagents following standard procedures [4], thus indicating steric hindrance of the C-5 hydroxyl by the prenyl group at the C-6 position. Therefore, the structure of **1** could be assigned as depicted.

Euchrestaflavanone A (**3**) could be easily identified by its ^{1}H NMR data which indicated a flavanone skeleton with three hydroxyl and two prenyl groups. One of the prenyl groups could be assigned to the C-3′ position on the basis of chemical shifts and coupling patterns of the B-ring signals. The other prenyl group could be assigned to the C-6 position since **3** also resisted trimethylsilylation of the C-5 hydroxyl group. The mp, UV and MS data all agreed with published information [6].

The UV, ^{1}H NMR and MS data for lupinalbin A-C (**5**, **2**,**4**) also agreed with those reported in the literature [7]. Previously unreported ^{13}C NMR data are presented for compounds **2** and **5** (Table 1).

EXPERIMENTAL

All UV spectra and TMSi derivatizations were carried out by standard procedures [4]

Roots of *Lotus creticus* L were collected during the flowering season in February 1985 in Egypt in El-kaser Descrite, 12 km from Marsa-matrouh. The plant was identified by Prof. El-Hadidy, the Department of Botany, Cairo University, Egypt

Air-dried powdered roots (800 g) were defatted with petrol, the material was then extracted with 70% aq EtOH After evapn of the solvent, the residue (22 g) was chromatographed over 400 g of silica gel (Merck) The column was first eluted with

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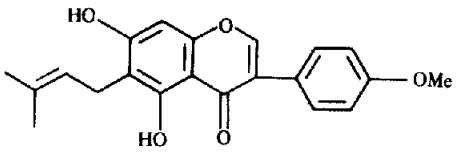

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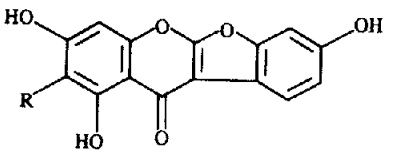
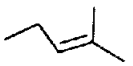


2 R = 

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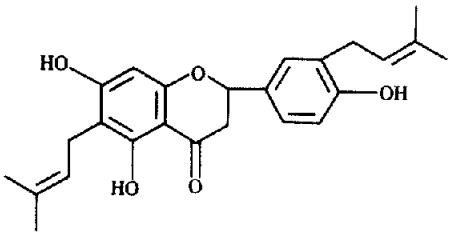

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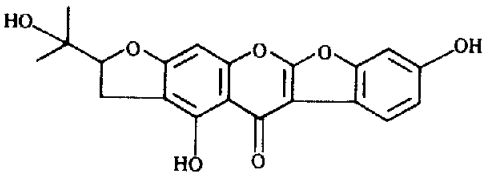

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Table 1 ^{13}C NMR chemical shifts* of compounds 2 and 5 (125 MHz, DMSO-d_6, TMS as int standard

C	2	5
A and C- ring		
2	149.8	149 8
3	113 4	113 4
4	178 2	178 2
5	161 1	162 0
6	111 8	99 8
7	164 2	164 2
8	94 0	94 3
9	156 2	156 2
10	102 5	102 5
B-ring		
1′	96 9	96 9
2′	152 3	154 6
3′	98 7	98 7
4′	159 0	162 0
5′	113 7	113 7
6′	121 0	121 0
Prenyl group		
1″	21.0	
2″	122 0	
3″	130 8	
4″	17 7	
5″	25 5	

*Signal assignments in the δ149–164 range are tentative

$CHCl_3$ (16 fractions, 150 ml each), then 2% MeOH in $CHCl_3$ (10 fractions, 150 ml each). The residue from fractions 8–9 (0.4 g) was rechromatographed on a small silica gel column (30 g) eluted with petrol–ether mixture (4 1); this procedure afforded compound 1 after crystallization from MeOH (40 mg) Rechromatography and repeated prep TLC separation of the concentrate of fractions 13–16 yielded, after crystallization in petrol, 175 mg of 3 as colourless needles. Crystallization of the material from fractions 17–24 (2.4 g) from MeOH afforded 2 (350 mg, colourless crystals). Repeated silica gel chromatography yielded 4 (2 mg, colourless crystals) and 5 (6 mg, colourless crystals)

Compound 1. Pale yellow crystals, mp 216–218° (MeOH). EIMS (probe) 70 eV *m/z* (rel int): 352 $[M]^+$(62), 337 $[M-Me]^+$(20), 309 $[M-C_3H_7]^+$ (89), 297 $[M-C_4H_7]^+$ (100), 165 $[A_1-55]^+$ (4), 132 $[B_1]^+$ (7), 69 [prenyl]$^+$ (7) UV λ_{max}^{MeOH} 268, 330 (sh); (NaOMe), 277, 345, ($AlCl_3$), 279, 310 (sh), 375; ($AlCl_3$/HCl), 279, 310 (sh), 375, (NaOAc), 273, 330, (NaOAc–H_3BO_3), 273, 330 (sh) ^{1}H NMR (500 MHz, DMSO-d_6, TMS): δ8 35 (1H, *s*, H-2), 6.46 (1H, *s*, H-8), 7 49 (2H, *d*, *J* = 8 Hz, H-2′, 6′), 7.02 (2H, *d*, *J* = 8 Hz, H-3′, 5′), 3 21 (2H, *br d*, *J* = 7 Hz, H-1″), 5.18 (1H, *br t*, *J* = 7 Hz, H-2″), 1 62 and 1 73 (each 3H, *br s*, H-4″, 5″).

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