

NMR Spectroscopy, X-ray Crystallographic, and Molecular Modeling Studies on a New Pyranone from *Haloxylon salicornicum*

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A new pyranone, 5-hydroxy-3-methoxy-4*H*-pyran-4-one (**1**), was isolated from the aerial parts of the desert shrub *Haloxylon salicornicum*. The structure was elucidated by X-ray structural analysis, NMR spectroscopy, and mass spectrometry. The monoacetate was also prepared, and molecular modeling studies and full NMR data were recorded.

As part of a continuing study into the chemistry and chemotaxonomy of the desert flora of Kuwait,¹ we have studied the chemistry of *Haloxylon salicornicum* (Moq.) Bunge ex Boiss. (Chenopodiaceae) and isolated a new simple pyranone (**1**) from a chloroform extract of the aerial parts of the plant. Previous phytochemical studies on this species has led to the isolation of simple quinolines² and tyramine derivatives.³ The family Chenopodiaceae is well represented in Kuwait,⁴ and some of the most hardy perennial desert plants belong to this taxon. The family is known for its propensity for tolerance of salty and xeric conditions.

By Biotage flash chromatography and recrystallization, compound **1** was isolated as fine needles. The EIMS of compound **1** indicated a molecular ion at *m/z* 142. By recrystallization from methanol–ethyl acetate (1:1), a crystal of sufficient quality of **1** was obtained for X-ray structural analysis, which on completion led to the solution of the structure of compound **1** (Table 1, Figure 1). Natural product **1** is assigned as the new pyranone, 5-hydroxy-3-methoxy-4*H*-pyran-4-one monohydrate.

NMR studies on this natural product confirmed the X-ray analysis of **1**. Signals in the ¹H and ¹³C NMR spectra indicated the presence of a methoxyl (δ_{H} 3.77, δ_{C} 57.8), two superimposed olefinic protons (δ_{H} 8.01 2H, δ_{C} 139.5, 142.0), and a broad hydroxyl moiety (δ_{H} 7.62). Compound **1** was acetylated to yield its acetate (**2**), which was subjected to 1D and 2D NMR studies. Both olefinic protons were now separate, but each of their corresponding carbon signals appeared as two signals. Analysis of the HMBC spectrum of **2** revealed correlations between an olefinic proton (H-6) and C-5 (²*J*), the C-4 carbonyl and C-2 (both ³*J*). The remaining olefinic proton, H-2, exhibited correlations with C-3 (²*J*), C-4, and C-6 (³*J*), which completed the pyran-4-one nucleus. The remaining cross-peaks could be attributed to the methoxyl protons to C-5 (³*J*) and the acetyl methyl group to its carbonyl (²*J*). To account for the doubling of the carbon resonances at positions C-2 and C-6 in compound **2**, molecular modeling was carried out to determine

Table 1. Atomic Positional and Isotropic Displacement Parameters

atom	<i>x</i>	<i>y</i>	<i>z</i>	<i>U</i> _{eq} (Å ²)
O(1)	0.1981(2)	0.3086(1)	0.0090(1)	0.0753(9)
C(2)	0.2678(2)	0.3768(2)	0.1033(2)	0.063(1)
C(3)	0.3205(2)	0.5379(2)	0.1100(1)	0.0466(8)
O(3)	0.3934(2)	0.6112(1)	0.20263(8)	0.0554(7)
C(31)	0.4276(3)	0.5034(2)	0.2961(2)	0.062(1)
C(4)	0.3036(2)	0.6489(2)	0.0145(1)	0.0445(8)
O(4)	0.3515(2)	0.7995(1)	0.01601(9)	0.0605(7)
C(5)	0.2284(2)	0.5708(2)	-0.0837(1)	0.0475(8)
O(5)	0.2117(2)	0.6711(2)	-0.17438(9)	0.0616(7)
C(6)	0.1807(3)	0.4072(2)	-0.0827(2)	0.062(1)
O(0)	0.5526(2)	0.9921(2)	0.1643(1)	0.0683(8)
H(2)	0.278(3)	0.303(3)	0.162(2)	0.086(6)
H(31a)	0.500(3)	0.566(3)	0.353(2)	0.084(6)
H(31c)	0.499(3)	0.405(3)	0.274(2)	0.079(6)
H(31b)	0.319(2)	0.467(2)	0.326(2)	0.072(5)
H(5)	0.154(3)	0.612(3)	-0.233(2)	0.083(6)
H(6)	0.133(3)	0.345(3)	-0.144(2)	0.080(6)
H(0a)	0.491(4)	0.926(3)	0.118(2)	0.103(8)
H(0b)	0.607(3)	1.058(3)	0.121(2)	0.093(7)

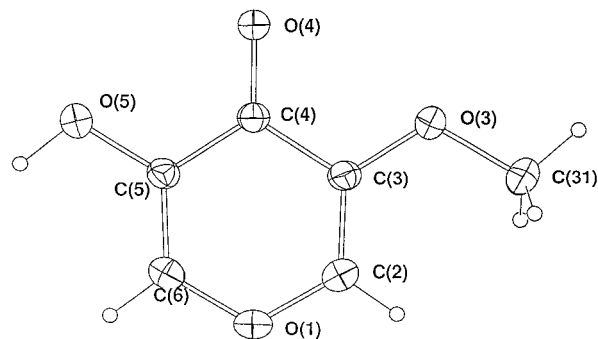


Figure 1. Molecular projection, normal to the plane of the molecule, showing 20% thermal ellipsoids for the non-hydrogen atoms, hydrogen atoms having arbitrary radii of 0.1 Å.

whether the acetylate adopted two low-energy conformations. Using semiempirical quantum mechanical energy calculations (MOPAC), conformational analysis about the C(4)–C(3)–O(3)–acetyl torsion angle revealed two different low-energy conformers that cannot interconvert (Figure 2). This would explain the doubling of NMR signals for the carbons at positions C-2 and C-6.

The demethyl parent compound, rubiginol (**3**), has been reported to be an insect sex pheromone from the male

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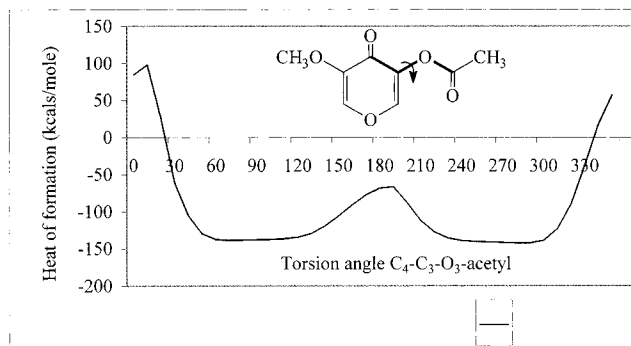
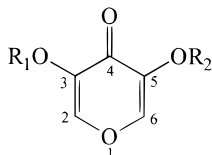


Figure 2. The change in the energy of the compound as the torsion angle shown was varied. The angle was changed in 10° increments, and the energy of the compound was measured with the semiempirical quantum mechanics program MOPAC using the AM1 Hamiltonian.

cotton harlequin bug *Tectoris diophthalmus*.⁵ It is possible that *H. salicornicum* stores rubiginol as its monomethyl ether (**1**), which may then be demethylated to the sex pheromone, which could possibly act as a pollination aid.



- 1 $R_1 = \text{Me}, R_2 = \text{H}$
 2 $R_1 = \text{Ac}, R_2 = \text{Me}$
 3 $R_1 = R_2 = \text{H}$

Experimental Section

General Experimental Procedures. The NMR spectra of **1** and **2** (acetone- d_6 , 400 MHz for ^1H , 100 MHz for ^{13}C) were run using a Bruker AMX-400 spectrometer with acetone as internal standard. EIMS were recorded on a Fisons AutospecQ instrument.

A sphere of X-ray data (8129 reflections) was measured using a Bruker AXS CCD detector instrument at ca. 300 K [specimen-detector 5 cm, $2\theta_{\text{max}} = 58^\circ$; 5 s frames, ω scan (0.3° increments); monochromatic Mo $K\alpha$ radiation, $\lambda = 0.71073 \text{ \AA}$], and processed using proprietary software (SAINT/SADABS/XPREP) to yield 1382 independent reflections ($R_{\text{int}} = 0.031$), all being used in the full-matrix least-squares refinement, refining anisotropic thermal parameter forms for C, O as well as (x, y, z, U_{iso})_H. Some deterioration (dehydration?) of the crystal occurred during data collection and was compensated for by appropriate scaling. Conventional residuals R , R_w (statistical weights) on $|F|$ were 0.044, 0.059. Pertinent results are given in Figure 1 and Table 1. Crystallographic data for **1** has been deposited with the Cambridge Crystallographic Data Centre. Copies of data can be obtained, free of charge, on application to the Director, CCDC, 12 Union Road, Cambridge CB2 1EZ, U.K. (fax: +44-(0)1223-336033 or e-mail: deposit@ccdc.cam.ac.uk). Neutral atom complex scattering factors were employed, computation using the Xtal 3.5 program system.⁶

A search of the Cambridge Structural Database⁷ revealed no structurally similar 3,5-disubstituted 4H-pyranone derivatives that have been crystallized. The structure was built using the molecular modeling package CHEMX⁸ and was optimized by the ab initio quantum mechanics program GAMESS⁹ with the 3-21G basis set. The conformational flexibility about the C(4)–C(3)–O(3)–acetyl torsion angle was examined by adjusting the angle in 10° increments with CHEMX and optimizing the structure using the semiempirical quantum mechanics package MOPAC (version 6) with the AM1 Hamiltonian.¹⁰

The torsion angle was fixed in each case. The keywords PRECISE, GEO-OK, GNORM=0.1 were specified with the BFGS optimizer.

Plant Material. Plant material was collected from the Jal-Az-Zoor in March 1998. A voucher specimen has been deposited at the Kuwait University Herbarium (KTUH), Khaldiyah, Kuwait.

Extraction and Isolation. The dried aerial parts of *H. salicornicum* (500 g) were extracted at room temperature in chloroform ($3 \times 2.5 \text{ L}$) and methanol ($2 \times 2.5 \text{ L}$). The chloroform extract was reduced under vacuum to give a residue of 20 g. This was then subjected to Biotage flash chromatography (Flash 75 S Si gel cartridge) eluting with hexane with increasing 10% amounts of ethyl acetate and finally 10% methanol in ethyl acetate. The residue from the fraction eluted with 70% ethyl acetate in hexane was then recrystallized from ethyl acetate to yield compound **1** (200 mg).

5-Hydroxy-3-methoxy-4H-pyran-4-one (1): colorless needles (acetone); mp $107\text{--}108^\circ\text{C}$; UV (methanol) λ_{max} (log ϵ) 299 (1.3) nm; IR ν_{max} (film) 3200, 3070, 1580, 1470, 1312, 1216, 1079, 952 cm^{-1} ; ^1H NMR (acetone- d_6 , 400 MHz) δ 8.01 (2H, s, H-2, H-6), 7.62 (1H, s, OH), 3.77 (3H, s, MeO); ^{13}C NMR (acetone- d_6 , 100 MHz) δ 169.8 (s, C-4), 148.2 (s, C-5), 147.1 (s, C-3), 142.0 (d, C-2), 139.5 (d, C-6), 57.8 (q, OMe); EIMS m/z 142 $[\text{M}]^+$ (100), 124 (35), 112 (55), 96 (30), 83 (35), 71 (20), 58 (60); FABMS m/z 165 $[\text{M}+\text{Na}]^+$ (40), 143 (100), 139 (25); HRFABMS m/z 143.0341 (calcd for $\text{C}_6\text{H}_7\text{O}_4$, 143.03443).

Crystal data: $\text{C}_6\text{H}_8\text{O}_5$, $M_r = 160.1$; monoclinic, space group $P2_1/c$ (C_{2h}^2 , no. 14), $a = 7.615(2) \text{ \AA}$, $b = 7.892(2) \text{ \AA}$, $c = 12.013(3) \text{ \AA}$, $\beta = 91.682(5)^\circ$, $V = 721.6 \text{ \AA}^3$. D_c ($Z = 4$) = 1.474 g cm^{-3} ; $F(000) = 336$. μ_{Mo} (no correction) = 1.3 cm^{-1} ; specimen: $0.48 \times 0.28 \times 0.15 \text{ mm}$. $n_D = 133$; $|\Delta\rho_{\text{max}}| = 0.23 \text{ e \AA}^{-3}$.

Acetylation of Compound 1: Compound **1** (10 mg) was dissolved in pyridine (2 mL), and then acetic anhydride (1 mL) was added. The mixture was left overnight and then evaporated under nitrogen. The resulting solid was then recrystallized from acetone to afford compound **2** (8 mg).

3-Acetoxy-5-methoxy-4H-pyran-4-one (2): cubes (acetone); mp $87\text{--}88^\circ\text{C}$; UV (acetone) λ_{max} (log ϵ) 324 (0.18) nm; IR ν_{max} (KBr) 2917, 2849, 1759, 1625, 1369, 1283, 1180, 1067 cm^{-1} ; ^1H NMR (acetone- d_6 , 400 MHz) δ 8.21 (1H, s, H-2), 8.00 (1H, s, H-6), 3.76 (3H, s, MeO), 2.24 (3H, s, Me); ^{13}C NMR (acetone- d_6 , 100 MHz) δ 168.3 (s, C-4), 168.3 (s, acetyl carbonyl), 150.5 (s, C-5), 149.3 and 149.2 (d, C-2), 141.5 (s, C-3), 141.0 and 140.9 (d, C-6), 57.5 (q, OMe), 20.2 (q, Me); EIMS m/z 368 $[2\text{M}]^+$ (10), 340 (5), 312 (10), 181 (20), 137 (30); FABMS m/z 207 $[\text{M}+\text{Na}]^+$ (100), 185 $[\text{M}+\text{H}]^+$ (35), 176 (30), 143 (60); HRFABMS m/z 207.0270 (calcd for $\text{C}_8\text{H}_8\text{O}_5\text{Na}$, 207.0269).

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