

New Isoprenylated Aurones from the Flowers of *Rosa damascena*

by Yin-Ke Li^{a)}), Ji-Qing Sun^{a)}), Xue-Mei Gao^{c)}), and Chun Lei^{*a)}

^{a)} Department of Pharmacognosy, School of Pharmacy, Fudan University, 826 Zhang Heng Road, Shanghai 201203, P. R. China

^{b)} College of Resource and Environment, Yuxi Normal University, Yuxi 653100, P. R. China

^{c)} Key Laboratory of Chemistry in Ethnic Medicinal Resources, State Ethnic Affairs Commission & Ministry of Education, Yunnan University of Nationalities, Kunming 650031, P. R. China
(phone: +86-21-51980173; e-mail: chunlei@fudan.edu.cn)

A phytochemical investigation of the flowers of *Rosa damascena* resulted in the isolation of three new isoprenylated aurones, damaurones C–E (**1–3**, resp.), together with six known compounds, **4–9**. The structures of the new compounds were elucidated by spectroscopic methods, including extensive 1D- and 2D-NMR experiments.

Introduction. – The genus *Rosa* has long been appreciated in history for the utilization of gardening, food, and medication [1]. The fruits, petals, and buds of some species are used as herbal tea, drinks, juice, jam, and medicine to treat stomachache, diarrhea and gynecological diseases [2]. *Rosa damascena* MILL., as one of the most important *Rosa* species, is widely cultivated in the world. Their flowers are renowned for fine fragrance and commercially processed for rose oil to manufacture high-quality perfume. Many reports are available on the studies of the volatile constituents from petals oil of this species, and phenolics were found to be the major components from this plant [3]. To identify bioactive natural compounds and multipurpose utilization of the species *R. damascena*, we carried out phytochemical investigation on the flowers of this species. Separation of the AcOEt-soluble portion from an 95% EtOH extract of the flowers afforded three new isoprenylated aurones, damaurones C–E (**1–3**, resp.), and six known compounds, rugaurone B (**4**) [4], maritimein (**5**) [5], 6,7,3',4'-tetrahydroaurone (**6**) [6], (*E*)-3'-*O*- β -D-glucopyranosyl-4,5,6,4'-tetrahydroxy-7,2'-dimethoxyaurone (**7**) [7], siamaurone A (**8**) [8], and damaurone B (**9**) [3a] (*Fig. 1*). In this article, we present the structure elucidation of compounds **1–3**. This is the first report of isoprenylated aurones isolated from *R. damascena*.

Results and Discussion. – *Structure Elucidation.* The AcOEt-soluble portion from a 95% EtOH extract of *R. damascena* was purified by repeated column chromatography over silica gel, *Sephadex LH-20*, and *RP-18*, as well as preparative HPLC, to yield three new isoprenylated aurones, **1–3**, and six known ones, **4–9** (*Fig. 1*). The structures of these compounds were determined by spectroscopic methods.

Damaurone C (**1**) was obtained as a yellow gum. The ESI mass spectrum showed a molecular ion at m/z 359, and the molecular formula was determined as $C_{20}H_{16}O_5$ from the HR-ESI-MS (m/z 359.0892 ($[M + Na]^+$, $C_{20}H_{16}NaO_5^+$; calc. 359.0895), requiring 13

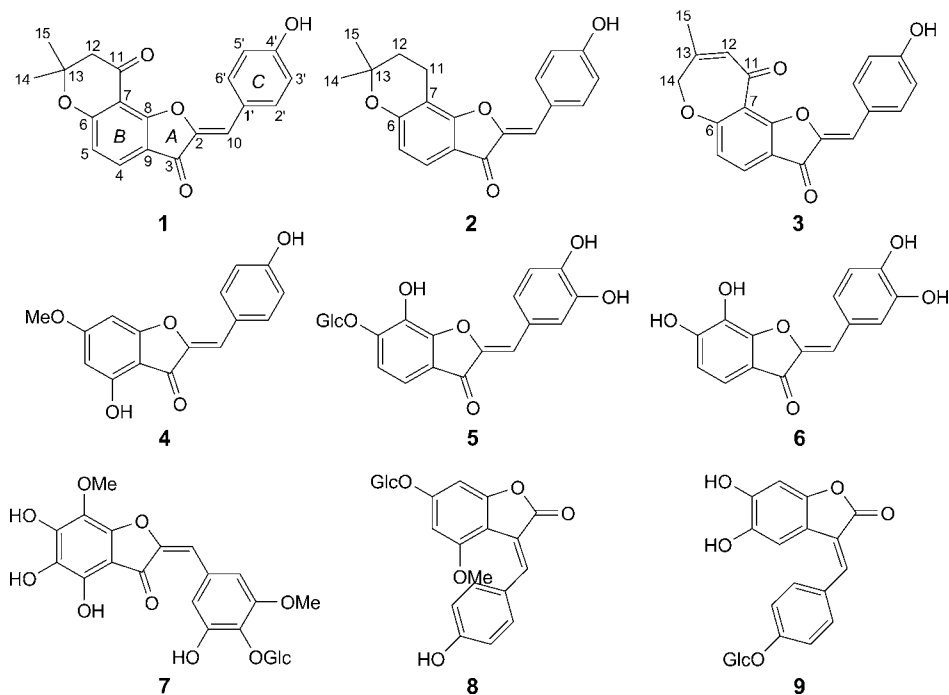


Fig. 1. Structures of compounds 1–9

degrees of unsaturation. The UV spectrum of **1** exhibited absorption maxima at 210, 262, and 350 nm, confirming the presence of the aromatic functions. The IR absorptions of **1** implied the presence of OH (3423 cm^{-1}) and C=O ($1692, 1668\text{ cm}^{-1}$) groups, and aromatic-ring ($1547, 1487\text{ cm}^{-1}$) moieties. The $^1\text{H-NMR}$ spectrum (*Table*) showed signals of a phenolic OH group at $\delta(\text{H})$ 11.20 (*s*, 1 H), a group of aromatic *AA'BB'* spin system at $\delta(\text{H})$ 7.81 (*d*, $J=8.8$, 2 H) and 6.83 (*d*, $J=8.8$, 2 H), an aromatic *AB* spin system at $\delta(\text{H})$ 7.72 (*d*, $J=8.6$, 1 H) and 6.72 (*d*, $J=8.6$, 1 H), an olefinic H-atom at $\delta(\text{H})$ 6.63 (*s*, 1 H), a broad *singlet* at $\delta(\text{H})$ 2.62 (2 H), and two Me *singlets* at $\delta(\text{H})$ 1.57 (*s*, 6 H). The $^{13}\text{C-NMR}$ spectrum exhibited 20 C-atom signals attributable to two CO groups, 14 sp^2 C-atoms, one O-bearing quaternary sp^3 C-atom, one CH_2 sp^3 C-atom, and two Me groups (*Table*). These data suggest that **1** is an isoprenylated flavone or aurone with one phenolic OH group. Two groups of HMBCs from the olefinic H-atom H–C(10) ($\delta(\text{H})$ 6.63) to the CO C-atom C(3) ($\delta(\text{C})$ 181.1), C(2) ($\delta(\text{C})$ 146.9), C(1') ($\delta(\text{C})$ 126.8), and C(2') and C(6') ($\delta(\text{C})$ 131.5), and from H–C(4) ($\delta(\text{H})$ 7.72) to C(3) clearly confirmed that **1** is an aurone (*Fig. 2*).

The *AA'BB'* spin system in the $^1\text{H-NMR}$ and the HMBC spectra of the phenolic OH ($\delta(\text{H})$ 11.20) with C(4') ($\delta(\text{C})$ 158.3) indicated that the OH group was attached to C(4') of the C-ring. According to the molecular formula and the 13 degrees of unsaturation, in addition to the eleven degrees of unsaturation for the aurone skeleton of A–C rings, there should be another O-containing ring with a CO group ($\delta(\text{C})$ 191.5) present in compound **1**. The O-containing ring should be a six-membered 2,2-

Table. ^1H - and ^{13}C -NMR Data (500 and 125 MHz, resp., in (D_5) pyridine) of Compounds **1**–**3**. δ in ppm, J in Hz.

Position ^{a)}	1		2		3	
	$\delta(\text{H})$	$\delta(\text{C})$	$\delta(\text{H})$	$\delta(\text{C})$	$\delta(\text{H})$	$\delta(\text{C})$
2	–	146.9	–	146.1	–	145.9
3	–	181.1	–	181.3	–	182.1
4	7.72 (<i>d</i> , $J=8.6$)	128.0	7.39 (<i>d</i> , $J=8.6$)	121.0	7.75 (<i>d</i> , $J=8.6$)	130.1
5	6.72 (<i>d</i> , $J=8.6$)	113.5	6.66 (<i>d</i> , $J=8.6$)	113.0	6.80 (<i>d</i> , $J=8.6$)	113.5
6	–	165.3	–	164.0	–	167.8
7	–	116.8	–	115.4	–	116.2
8	–	162.8	–	160.6	–	164.0
9	–	115.0	–	113.9	–	115.8
10	6.63 (<i>s</i>)	112.4	6.62 (<i>s</i>)	112.2	6.64 (<i>s</i>)	112.1
11	–	191.5	2.58 (<i>dd</i> , $J=6.8, 6.8$)	21.2	–	188.1
12	2.62 (<i>br. s</i>)	48.8	1.72 (<i>dd</i> , $J=6.8, 6.8$)	33.1	6.39 (<i>s</i>)	132.9
13	–	80.7	–	73.9	–	154.3
14	1.57 (<i>s</i>)	26.9	1.52 (<i>s</i>)	26.3	4.41 (<i>s</i>)	72.0
15	1.57 (<i>s</i>)	26.9	1.52 (<i>s</i>)	26.3	1.89 (<i>s</i>)	20.8
1'	–	126.8	–	126.3	–	126.5
2',6'	7.81 (<i>d</i> , $J=8.8$)	131.5	7.82 (<i>d</i> , $J=8.8$)	131.1	7.83 (<i>d</i> , $J=8.8$)	131.8
3',5'	6.83 (<i>d</i> , $J=8.8$)	116.2	6.89 (<i>d</i> , $J=8.8$)	116.3	6.87 (<i>d</i> , $J=8.8$)	116.8
4'	–	158.3	–	158.2	–	158.9
HO–C(4')	11.20 (<i>s</i>)	–	11.20 (<i>s</i>)	–	11.18 (<i>s</i>)	–

^{a)} C-Atom numbering as indicated in Fig. 1.

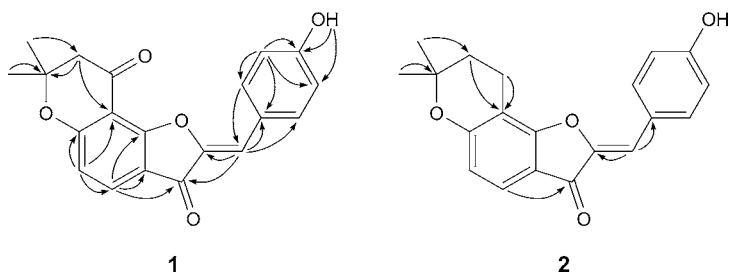


Fig. 2. Selected HMBCs ($\text{H} \rightarrow \text{C}$) of compounds **1** and **2**

dimethylpyran ring, since two Me *singlets* were observed in the ^1H - and ^{13}C -NMR spectra. The CO group was determined to be at C(11) in the pyran ring, deduced from the key HMBCs from Me(14,15) ($\delta(\text{H})$ 1.57) to C(12) ($\delta(\text{C})$ 48.8) (Fig. 2) and the ring was established to be fused along C(6) ($\delta(\text{C})$ 165.3) and C(7) ($\delta(\text{C})$ 116.8) of *B*-ring by the occurrence of aromatic *AB* spin system of H–C(4) with H–C(5) ($\delta(\text{H})$ 6.72) in the *B*-ring and the key HMBC from CH_2 (12) ($\delta(\text{H})$ 2.62) to C(7) (Fig. 2).

The configuration of the olefinic bond in **1** was determined as (*Z*) by the chemical-shift value of C(10) ($\delta(\text{C})$ 112.4), since the configuration of the olefinic C=C bond in aurones can be established on the chemical shift of olefinic CH resonance at $\delta(\text{C})$ 119.9–121.5 in (*E*)-aurones and 105.9–112.8 in (*Z*)-aurone [9]. Thus, the structure of **1**

was elucidated as (2*Z*)-2-(4-hydroxybenzylidene)-7,7-dimethyl-7*H*-furo[2,3-*f*]chromene-3,9(2*H*,8*H*)-dione and named damaurone C (Fig. 1).

Damaurone D (**2**) was obtained as a yellow gum. Its molecular formula was deduced as C₂₀H₁₈O₄ from HR-ESI-MS (*m/z* 345.1110 ([*M* + Na]⁺ C₂₀H₁₈NaO₄⁺; calc. 345.1103), indicating twelve degrees of unsaturation. The ¹H- and ¹³C-NMR spectra (Table) displayed almost the same signals for *A*- and *C*-ring as those of compound **1**, indicating **2** was an aurone with a OH group at C(4') of *C*-ring. Except signals of *A*- and *C*-ring, the ¹H-NMR displayed an aromatic *AB* spin system at δ(H) 7.39 (*d*, *J* = 8.6, 1 H) and 6.66 (*d*, *J* = 8.6, 1 H), a spin system of CH₂(11) and CH₂(12) at δ(H) 2.58 (*dd*, *J* = 6.8, 6.8, 2 H) respectively, and 1.72 (*dd*, *J* = 6.8, 6.8, 2 H), and two Me *singlets* at δ(H) 1.52 (*s*, 6 H) (Table). The ¹³C-NMR spectrum exhibited signals of two CH₂ sp³-C-atoms in compound **2**, compared to those of one CH₂ sp³-C-atom and one CO moiety in compound **1** (Table). These data suggested that a 2,2-dimethylpyran ring without a CO group was fused with *B*-ring in **2**. The HMBs from Me(14,15) (δ(H) 1.52) to C(12) (δ(C) 33.1), and from CH₂(11) (δ(H) 2.58) to C(7) (δ(H) 115.4) (Fig. 2) further confirmed the 2,2-dimethylpyran ring substructure, which was also fused along C(6)–C(7), as in **1** by the HMBC of H–C(4) (δ(H) 7.39) with C(3) (δ(C) 181.3) (Fig. 2), and by the *AB* spin system of H–C(4) and H–C(5) (δ(H) 6.66) in the ¹H-NMR spectrum (Table). Thus, the structure of **2** was elucidated as (2*Z*)-8,9-dihydro-2-(4-hydroxybenzylidene)-7,7-dimethyl-7*H*-furo[2,3-*f*]chromen-3(2*H*)-one and named damaurone D (Fig. 1).

Damaurone E (**3**) was obtained as a yellow gum. The HR-ESI-MS exhibited a parent-ion peak at *m/z* 357.0732 ([*M* + Na]⁺, C₂₀H₁₄NaO₅⁺; calc. 357.0739) corresponding to the molecular formula C₂₀H₁₄O₅, requiring 14 degrees of unsaturation. Comparing the ¹H- and ¹³C-NMR spectra data with those of compound **1**, they exhibited almost the same signals of *A*- and *C*-ring (Table). Considering the aurone skeleton, together with the molecular formula and 14 degrees of unsaturation, there should be another ring with two O-atoms and a moiety possessing two degrees of unsaturation. The aromatic *AB* spin system at δ(H) 7.75 (*d*, *J* = 8.6, 1 H) and 6.80 (*d*, *J* = 8.6, 1 H) in the ¹H-NMR spectrum (Table) and the HMBC from H–C(4) (δ(H) 7.75) to C(3) (δ(C) 182.1) (Fig. 3) restricted the difference between compound **1** and **3** to the newly-built ring fused along C(6)–C(7) (δ(C) 167.8 and 116.2, resp.). The ¹H-NMR spectrum (Table) of **3** displayed signals of an olefinic H-atom at δ(H) 6.39 (*s*, 1 H), an O-bearing CH₂ group at δ(H) 4.41 (*s*, 2 H), and a Me group at δ(H) 1.89 (*s*, 3 H). The ¹³C-NMR (Table) evidenced the presence of an additional olefinic bond and just one Me group. These data suggested that one of two Me groups in **3** was O-bearing to build a seven-membered oxepane ring with one C=C bond and a CO moiety. The key HMBs from Me(15) (δ(H) 1.89) to C(12) (δ(C) 132.9), C(13) (δ(C) 154.3), and C(14) (δ(C) 72.0), and from H–C(12) (δ(H) 6.39) to C(7) (δ(C) 116.2), C(11) (δ(C) 188.1), C(13), and C(14) confirmed the oxepane using with a α,β-unsaturated ketone moiety as depicted (Fig. 3). Thus, the structure of compound **3** was established as (2*Z*)-2-(4-hydroxybenzylidene)-8-methylfuro[2,3-*g*][1]benzoxepine-3,10(2*H*,7*H*)-dione, named damaurone E (Fig. 1).

By comparison of the physical and spectroscopic data with those previously published, the remaining six known compounds were identified as rugaurone B (**4**) [4], maritimein (**5**) [5], 6,7,3',4'-tetrahydroxyaurone (**6**) [6], (*E*)-3'-*O*-β-D-glucopyranosyl-

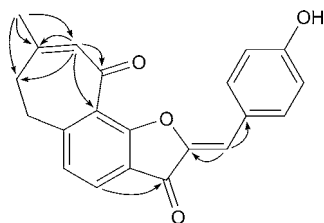


Fig. 3. Selected HMBCs (H → C) of compound **3**

4,5,6,4'-tetrahydroxy-7,2'-dimethoxyaurone (**7**) [7], siamaurone A (**8**) [8], and damaurone B (**9**) [3a].

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Experimental Part

General. Column chromatography (CC): silica gel *H* (200–300 mesh; *Yantai Institute of Chemical Technology*, P. R. China), *Chromatorex RP-18* gel (20–45 μm ; *Fuji Silysia Chemical, Ltd.*, Japan), and *Sephadex LH-20* (*GE Healthcare Amersham Biosciences*, Sweden). TLC: Precoated silica gel *GF₂₅₄* plates (10–40 μm ; *Yantai Institute of Chemical Technology*, P. R. China). HPLC: *Agilent 1200* (*Agilent Technologies*, USA), *Sepax Amethyst C₁₈* column (10 \times 50 mm, 5 μm ; *Sepax Technologies, Inc.*, USA). UV Spectra: *Shimadzu UV-2401PC* spectrophotometer; λ_{max} (log ϵ) in nm. IR Spectra: *Nicolet Avatar-360* spectrometer; $\tilde{\nu}_{\text{max}}$ in cm^{-1} . NMR Spectra: *Bruker DRX-500* instruments; δ in ppm rel. to residual solvent peaks of (*D*₅)pyridine ($\delta(\text{H})$ 8.74, 7.58, 7.22; $\delta(\text{C})$ 150.3, 135.9, 123.9); *J* in Hz. ESI-MS and HR-ESI-MS: *VG Auto Spec-3000* mass spectrometer; in *m/z* (rel. %).

Plant Material. The flowers of *Rosa damascena* MILL. were collected in Chenggong County, Yunnan Province, in September 2011. The plant was identified by Prof. *Y.-J. Chen* (Yunnan Nationalities University). A voucher specimen (YNNI 12-09-65) has been deposited with our laboratory.

Extraction and Isolation. The air-dried and powdered flowers of *R. damascena* (5 kg) were soaked in 95% EtOH (20:1) at r.t. for 48 h before extraction. After extracting three times, the combined residue without solvents was partitioned between AcOEt and H₂O. The AcOEt crude extract (130 g) was subjected to CC (SiO₂ (200–300 mesh); CH₂Cl₂/acetone 50:1, 20:1, 9:1, 8:2, 7:3, 6:4, 5:5), to give seven *Frs. A–G*. The further separation of *Fr. D* (7:3, 21.2 g) by CC (SiO₂; CHCl₃/MeOH 9:1, 8:2, 7:3, 6:4, 1:1) yielded mixtures *D1–D6*. *Fr. D1* (8:2, 1.8 g) was subjected to CC (*Sephadex LH-20* and *Chromatorex RP-18* gel) and then prep. HPLC (30–60% MeOH, 3 ml/min) to give **1** (18 mg), **2** (13 mg), and **3** (16.5 mg). The further separation of *Fr. F* (6:4, 40 g) by CC (SiO₂ and *Chromatorex RP-18* gel; 20% MeOH) to give **4** (10.4 mg), **5** (15 mg), **6** (13.2 mg), **7** (18.5 mg), **8** (12.5 mg), and **9** (7.6 mg).

Damaurone C (= (2*Z*)-2-(4-Hydroxybenzylidene)-7,7-dimethyl-7H-furo[2,3-*f*]chromene-3,9(2*H*, 8*H*)-dione; **1**). Yellow gum. UV (MeOH): 350 (3.28), 262 (3.92), 210 (4.18). IR (KBr): 3423, 2916, 2875, 1692, 1668, 1614, 1547, 1487, 1426, 1269, 1149, 1068, 876, 780. ¹H- and ¹³C-NMR: see the *Table*. ESI-MS (pos.): 359 ([*M* + Na]⁺). HR-ESI-MS (pos.): 359.0892 ([*M* + Na]⁺, C₂₀H₁₆NaO₅⁺; calc. 359.0895).

Damaurone D (= (2*Z*)-8,9-Dihydro-2-(4-hydroxybenzylidene)-7,7-dimethyl-7H-furo[2,3-*f*]chromen-3(2*H*)-one; **2**). Yellow gum. UV (MeOH): 344 (3.12), 258 (3.85), 210 (4.24). IR (KBr): 3431, 2964, 2927, 2869, 1664, 1612, 1545, 1482, 1436, 1368, 1273, 1151, 1076, 881, 764. ¹H- and ¹³C-NMR: see the *Table*. ESI-MS (pos.): 345 ([*M* + Na]⁺). HR-ESI-MS (pos.): 345.1110 ([*M* + Na]⁺, C₂₀H₁₈NaO₄⁺; calc. 345.1103).

Damaurone E (= (2*Z*)-2-(4-Hydroxybenzylidene)-8-methylfuro[2,3-*g*][1]benzoxepine-3,10(2*H*,7*H*)-dione; **3**). Yellow gum. UV (MeOH): 368 (3.09), 276 (3.92), 210 (4.20). IR (KBr): 3431, 2924, 2870, 1672,

1663, 1610, 1553, 1482, 1457, 1382, 1274, 1146, 1065, 868, 754. ^1H - and ^{13}C -NMR: see the *Table*. ESI-MS (pos.): 357 ($[M + \text{Na}]^+$). HR-ESI-MS (pos.): 357.0732 ($[M + \text{Na}]^+$, $\text{C}_{20}\text{H}_{14}\text{NaO}_5^+$; calc. 357.0739).

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