Speciosins A−K, Oxygenated Cyclohexanoids from the Basidiomycete *Hexagonia speciosa*

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Eleven new oxygenated cyclohexanoids, speciosins A−K (**1**−**11**), and aporpinone A (**12**) were isolated from broth cultures of the basidiomycete *Hexagonia speciosa*. Their structures were elucidated on the basis of extensive spectroscopic analysis, while the structure of **8** was additionally confirmed by analysis of single-crystal X-ray diffraction data.

Oxygenated cyclohexanoids, mainly epoxides, which occur in bacteria, fungi, higher plants, and mollusks, possess a wide range of bioactivities (e.g., antifungal, antibacterial, antitumor, antibiotic, and phytotoxic).[1−8] The basidiomycete *Hexagonia speciosa* is a fungus of the family Polyporaceae that is distributed in the tropical and subtropical zones of China, such as the Hainan and Yunnan Provinces.[9,10] The secondary metabolites of *H. speciosa* have received little attention. As a part of our search for naturally occurring secondary metabolites from higher fungi in China,[11−14] we have carried out chemical investigations of *H. speciosa* cultures, which led to the isolation of a series of new oxygenated cyclohexanoids, speciosins A−K (**1**−**11**) and aporpinone A (**12**).[15]

Results and Discussion

The organism was cultured in a modified PDA medium shaken at 150 rpm. After culturing for 25 days at 25 °C, the whole culture broth (21 L) was filtered and the filtrate was extracted three times with EtOAc. The crude EtOAc extract (5.3 g) was subjected to repeated column chromatography to give pure **1** (4.7 mg), **2** (31.0 mg), **3** (5.7 mg), **4** (1.0 mg), **5** (103.0 mg), **6** (2.1 mg), **7** (2.0 mg), **8** (2.8 mg), **9** (2.7 mg), **10** (6.1 mg), **11** (3.8 mg), and **12** (10.0 mg).

Speciosin A (**1**) was isolated as a colorless oil whose molecular formula was established as $C_{10}H_8O_3$ by means of a positive HRFABMS $[M + H]^+$ ion at *m*/*z* 177.0557 (calcd for $C_{10}H_9O_3$, 177.0552). The IR spectrum showed absorptions of hydroxy, carbonyl, and acetylenic groups at 3431, 1729, and 2229 cm^{-1}, respectively. The presence of a cyclohex-2,3-enone group was suggested from the ^{13}C NMR spectrum (δ_C 189.4, s; 125.6, d; 144.3, d; Table 1) and the ^{1}H NMR spectrum (δ_H 6.07, dd, $J = 10.6$, 1.1 Hz, H-2; 6.67, ddd, $J = 10.6$, 4.7, 2.5 Hz, H-3). The proton resonating at δ_H 6.67 showed coupling ($J = 4.7$ Hz) to an oxymethine proton at δ_H 4.67 (m, H-4), which in turn was coupled to another oxymethine at δ_H 4.04 (dd, $J = 2.5$, 1.0 Hz, H-5). Signals for three olefinic protons, observed at δ_H 5.87 (dd, $J = 17.6$, 10.0 Hz, H-9) and a terminal methylene at δ 5.81 (dd, $J = 17.6$, 3.1 Hz, H-10a) and 5.63 (dd, $J = 10.0$, 3.1 Hz, H-10b) indicated a terminal double-bond function. The connectivity between this group and the cyclohex-2,3-enone moiety involved acetylenic carbons (δ_C 86.0, s; 81.4, s). Six degrees of unsaturation have thus been assigned, and the remaining one may be accommodated by assuming the presence of an epoxide ring at C-5 and C-6 of the cyclohexenone. Except for a methyl resonance, the NMR data were very similar to those reported for 5-hydroxy-1-(3-methyl-3-buten-1-ynyl)-7-oxabicyclo[4.1.0]hept-3-en-2-one (**1a**),[1] which was confirmed by X-ray diffraction. This suggested that **1** was a nor-methyl cyclohexenone derivative. The structural assignment was further confirmed by the following HMBC correlations: from the terminal methylene protons to an alkynyl carbon at δ 86.0 (C-8); from an olefinic proton at δ 5.87 (H-9) to the carbons at δ 81.4 (C-7) and 86.0 (C-8); and from the oxymethine proton at δ 4.04 (H-5) to the carbonyl carbon at δ 189.4 (C-1), the olefinic carbon at δ 144.3(C-3), and the acetylenic carbon at δ 81.4 (C-7). The magnitude and sign of the specific rotation and the NMR data, especially the coupling constants in the ^{1}H NMR spectrum of **1**, were in good accord with **1a**, suggesting that the two metabolites had the same relative configuration. Consequently, the structure of **1** was established as that shown in Figure 1 and named speciosin A.

Table 1. ^{13}C NMR Data of Speciosins A−J (**1**−**10**)

pos.	**1**[a]	**2**[b]	**3**[c]	**4**[c]	**5**[d]	**6**[d]	**7**[e]	**8**[b]	**9**[b]	**10**[c]
1	189.4	188.8	67.4	67.1	67.9	68.1	152.4	74.0	67.3	69.0
2	125.6	126.2	128.2	128.1	129.1	129.3	117.2	28.9	31.3	27.3
3	144.3	143.8	125.9	125.9	126.7	126.9	118.3	31.2	29.3	27.3
4	62.1	62.7	62.7	62.5	63.2	63.4	151.1	65.8	70.6	63.3
5	66.0	65.4	63.7	63.2	63.6	63.8	119.4	36.8	35.6	33.4
6	52.7	52.2	53.7	53.1	54.3	54.4	112.0	39.6	40.9	35.9
7	81.4	84.8	85.3	80.3	83.5	83.4	87.2	31.1	31.1	30.2
8	86.0	75.6	84.7	81.1	83.8	83.7	92.7	122.5	122.3	122.6
9	115.5	39.5	125.5	39.5	63.9	62.0	118.7	133.2	133.1	132.8
10	129.6	48.6	123.9	48.6	66.8	77.2	126.6	25.9	25.8	25.8
11			23.0			59.4		17.8	17.8	17.8

[a] Spectra of **1** were collected in $CDCl_3$−methanol-d_4 = 4:1 at 100 MHz. [b] Spectra were collected in $CDCl_3$ at 125 MHz. [c] Spectra were collected in $CDCl_3$ at 100 MHz. [d] Spectra were collected in methanol-d_4 at 100 MHz. [e] The spectrum was collected in methanol-d_4 at 125 MHz.

Speciosin B (**2**) was obtained as a colorless oil with the molecular formula $C_{10}H_8O_4$, as determined from the pseudomolecular ion observed at *m*/*z* 215.0325 $[C_{10}H_8O_4 + Na]^+$ using HRESIMS techniques. The IR spectrum indicated the presence of hydroxy (a broad band 3449 cm^{-1}), carbonyl (1732 cm^{-1}), and alkyne (2256 cm^{-1}) groups. The ^{1}H NMR signals (Tables 1 and 2) were similar to those of speciosin A (**1**), suggesting that **2** was also a cyclohexenone derivative. The presence of the core cyclohex-2,3-enone structure was characterized from the ^{13}C (δ_C 188.8, s; 126.2, d; 143.8, d; Table 1) and ^{1}H NMR spectra (δ_H 6.01, dd, $J = 10.6$, 1.1 Hz, H-2; 6.67, ddd, $J = 10.6$, 4.8, 2.6 Hz, H-3). The prominent differences between the two compounds were as follows: the resonances from the terminal double bond were absent and replaced by signals belonging to an epoxide ring at δ_C 39.5, δ_H 3.45 (1H, dd, $J = 4.0$, 2.6 Hz, H-9) and δ_C 48.6, δ_H 2.99 (1H, dd, $J = 5.5$, 2.6 Hz, H-10a), 2.96 (1H, dd, $J = 5.5$, 4.0 Hz, H-10b) in the NMR spectra of **2**. The presence of an additional oxygen atom in **2** compared with **1** was also confirmed from its HRESIMS data. The HMBC spectrum also showed the significant correlations from H-9

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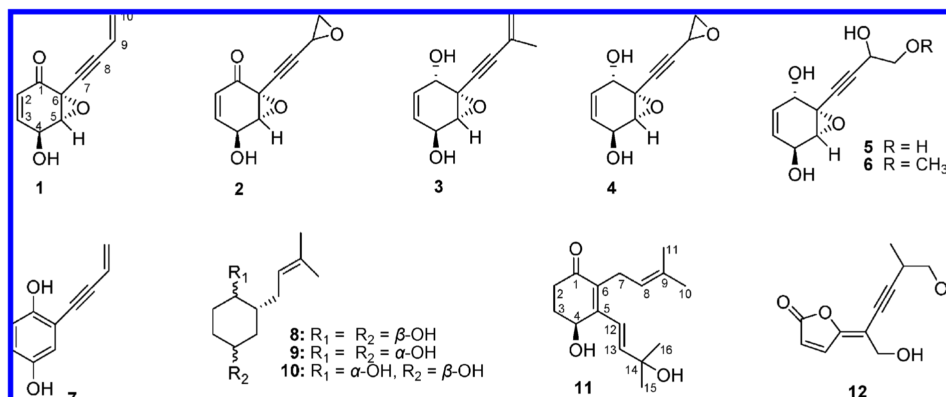

Figure 1. Structures of **1**−**12**.

Table 2. ^{1}H NMR Data of Speciosins A−F (**1**−**6**)

pos.	**1**[a]	**2**[b]	**3**[c]	**4**[c]	**5**[d]	**6**[d]
1			4.58 (brs)	4.58 (m)	4.55 (brs)	4.52 (overlapped)
2	6.07 (dd, 10.6, 1.0)	6.01 (dd, 10.6, 1.1)	5.70 (dd, 10.6, 2.5)	5.69 (brd, 10.5)	5.58 (brd, 10.6)	5.57 (dd, 10.5, 2.2)
3	6.67 (ddd, 10.6, 4.7, 2.5)	6.67 (ddd, 10.6, 4.8, 2.6)	5.77 (m)	5.77 (m)	5.68 (m)	5.67 (m)
4	4.67 (m)	4.63 (d, 4.8)	4.43 (brd, 4.7)	4.43 (m)	4.28 (brd, 3.5)	4.25 (brd, 3.9)
5	4.04 (dd, 2.5, 1.0)	4.03 (dd, 2.6, 1.1)	3.69 (brs)	3.69 (m)	3.51 (brs)	3.47 (d, 1.8)
9	5.87 (dd, 17.6, 10.0)	3.45 (dd, 4.0, 2.6)		3.43 (dd, 7.3, 3.7)	4.39 (m)	4.52 (overlapped)
10	5.81 (dd, 17.6, 3.1)	2.99 (dd, 5.5, 2.6)	5.39 (s)	2.96 (m)	3.60 (m)	3.48 (m)
	5.63 (dd, 10.0, 3.1)	2.96 (dd, 5.5, 4.0)	5.31 (s)			
11			1.89 (s)			3.39 (s)

[a] Spectra of **1** were collected in $CDCl_3$−methanol-d_4 = 4:1, at 400 MHz. [b] Spectra were collected in $CDCl_3$ at 500 MHz. [c] Spectra were collected in $CDCl_3$ at 400 MHz. [d] Spectra were collected in methanol-d_4 at 400 MHz.

to C-7 and C-8, as well as from H-10 to C-8. Thus, the structure of **2** was elucidated as that shown in Figure 1.

Speciosin C (**3**) was obtained as a colorless oil. Its molecular formula was established as $C_{11}H_{12}O_3$ on the basis of HRESIMS and NMR spectra and contained six degrees of unsaturation. Its IR spectrum suggested the presence of hydroxy (3464, 3374 cm^{-1}) and alkynyl (2224 cm^{-1}) groups. Its ^{1}H and ^{13}C NMR spectra displayed resonances corresponding to a methyl, one bis-substituted double bond, one terminal double bond, one triple bond, three oxymethines, and a fully substituted carbon. Analysis of its NMR spectra indicated that it had the same skeleton as speciosin A (**1**). The main differences between compounds **1** and **3** were as follows: the ketone carbonyl in **1** was replaced by an oxymethine at δ_C 67.4, δ_H 4.58 (1H, brs) in **3**, and the latter had one more methyl group at δ_C 23.0, δ_H 1.89 (3H, s) than **1**. The ^{1}H−^{1}H COSY experiment delineated a spin system from H-1 to H-4. The HMBC cross-peaks of H-5 with C-1, C-3, C-6, and C-7 indicated a planar structure for **3**, as shown in Figure 1. The relative configuration of **3** was deduced from ^{1}H NMR coupling constants with the aid of Dreiding models. The $J_{3,4}$ coupling constant of 4.7 Hz indicated that the 4-OH was in a pseudoaxial position in the "closed boat" conformation of a 7-oxabicyclo[4.1.0]hept-3-ene ring system.[7,16−18] A small coupling constant was observed between H-1 and H-2, indicating the pseudoequatorial orientation of 1-OH.[4] Thus, the structure of speciosin C was established.

Speciosin D (**4**), speciosin E (**5**), and speciosin F (**6**) displayed NMR and IR features similar to those of speciosin C (**3**). Speciosin D (**4**) was obtained as a colorless oil and possessed a molecular formula of $C_{10}H_{10}O_4$, as determined by a HRESIMS signal at m/z 217.0476 ($C_{10}H_{10}O_4Na$, calcd 217.0476). Its ^{13}C NMR and DEPT (Table 1) spectra exhibited 10 carbon resonances, comprising a triple bond, a bis-substituted double bond, a methylene, four methines, and a fully substituted carbon. The ^{1}H NMR (Table 2) spectrum of **4** confirmed the presence of the functional groups indicated above. In addition, the spectrum displayed two deuterium-exchangeable signals at δ 2.27 (1H, d, J = 10.0 Hz, 1-OH) and 1.88 (1H, d, J = 7.8 Hz, 4-OH), suggesting the presence of two hydroxy groups. The data were similar to that of speciosin C (**3**), except that the isopropylene group in **3** had been replaced by an epoxide ring in **4**. Thus, the structure of **4** was elucidated to be that shown in Figure 1.

Speciosin E (**5**) was isolated as a colorless oil with a molecular formula of $C_{10}H_{12}O_5$. Its ^{1}H and ^{13}C NMR spectra were similar to those of **4**. The main differences were as follows: **5** had signals suggesting an oxymethine (δ_C 63.9; δ_H 4.39) and an oxymethylene (δ_C 66.8; δ_H 3.60) instead of the epoxy ring found in speciosin D (**4**). Therefore, the structure of **5** was established as that shown in Figure 1.

Speciosin F (**6**) had the molecular formula $C_{11}H_{14}O_5$, as determined by HRESIMS and NMR spectra. A comparison of the NMR data of compound **6** with those of compound **5** indicated that the only difference was one additional *O*-methyl group (δ_C 59.4; δ_H 3.39, s, H-11) in **6**. The HMBC cross-peak of H-11 with C-10 (δ_C 77.2) indicated that the *O*-methyl was located at C-10, which causes C-10 to be shifted downfield.

The magnitude and signs of the specific rotation and the NMR data, especially the coupling constants in the ^{1}H NMR spectrum of speciosins D−F (**4**−**6**), were in accord with those of **3**, suggesting that the these metabolites had the same relative configuration.

The molecular formula of speciosin G (**7**) was found to be $C_{10}H_8O_2$ by negative ESIMS, which indicated seven degrees of unsaturation. Its ^{1}H and ^{13}C NMR (Tables 1 and 3) spectra revealed the presence of one terminal double bond at δ 126.6 (t) and 118.7 (d), one triple bond at δ 92.7 (s) and 87.2 (s), three olefinic methines at δ 119.4 (d), 118.3 (d), and 117.2 (d), and three fully substituted carbons at δ 152.4 (s), 151.1 (s), and 112.0 (s). Analysis of the NMR spectra of compound **7** revealed a typical pattern for a 1,2,4-trisubstituted aromatic ring: H-2 (δ 6.66, d, J = 8.8 Hz), H-3 (δ 6.63, dd, J = 8.8, 2.4 Hz), and H-5 (δ 6.68, d, J = 2.4 Hz). The HMBC correlations of H-5 with C-1 (δ 152.4), C-3 (δ 118.3), and C-7 (δ 87.2); of H-2 with C-4 (δ 151.1) and C-6 (δ 112.0); and of H-3 with C-1 (δ 152.4) and C-5 (δ 119.4) led to the assignment of the structure of **7** as that shown in Figure 1. Speciosin G is an aromatic acetylenic metabolite.

Table 3. ^{1}H NMR Data of Speciosins G−J (**7**−**10**)

pos.	**7**[a]	**8**[b]	**9**[b]	**10**[c]
1		3.35 (ddd, 9.2, 9.2, 3.7)	3.83 (brs)	3.91 (brs)
2	6.66 (d, 8.8)	1.76 (overlapped)	1.88 (ddd, 14.1, 6.8, 3.4)	1.87 (overlapped)
		1.68 (m)	1.48 (ddd, 14.1, 3.4, 3.0)	1.62 (m)
3	6.63 (dd, 8.8, 2.4)	1.78 (m)	1.76 (m)	1.87 (overlapped)
		1.55 (m)	1.56 (m)	1.51 (overlapped)
4		4.02 (brt, 2.7)	3.59 (dddd, 10.7, 10.7, 4.7, 4.7)	4.08 (brs)
5	6.68 (d, 2.4)	1.83 (ddd, 16.3, 6.3, 2.7)	1.74 (overlapped)	1.68 (m)
		1.25 (ddd, 16.3, 11.4, 2.7)	1.37 (ddd, 10.7, 10.7, 10.7)	1.51 (overlapped)
6		1.76 (overlapped)	1.44 (m)	1.87 (overlapped)
7		2.31 (ddd, 14.6, 12.6, 7.4)	2.09 (ddd, 14.1, 14.1, 7.3)	2.07 (ddd, 14.1, 14.1, 6.8)
		1.93 (ddd, 14.6, 14.6, 7.2)	2.01 (ddd, 14.1, 14.1, 7.3)	1.94 (ddd, 14.1, 14.1, 7.2)
8		5.19 (dd, 7.4, 7.2)	5.13 (t, 7.3)	5.14 (dd, 7.2, 6.8)
9	6.06 (dd, 17.1, 11.2)			
10	5.68 (dd, 17.1, 1.9)	1.70 (s)	1.70 (s)	1.70 (s)
	5.49 (dd, 11.2, 1.9)			
11		1.63 (s)	1.63 (s)	1.62 (s)

[a] Spectra were collected in methanol-d_4 at 500 MHz. [b] Spectra were collected in $CDCl_3$ at 500 MHz. [c] Spectra were collected in $CDCl_3$ at 400 MHz.

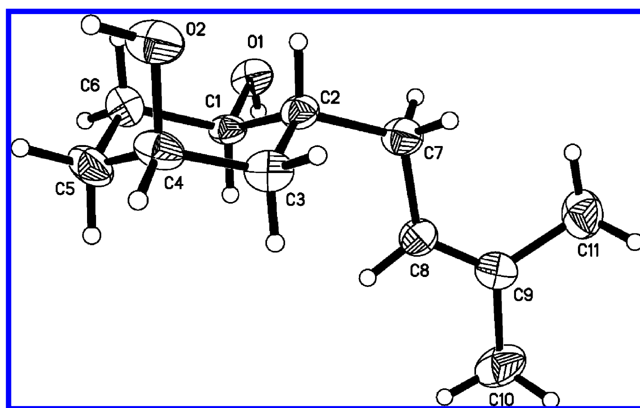

Figure 2. ORTEP drawing of **8**.

Speciosins H−J (**8**−**10**) all had identical molecular formulas of $C_{11}H_{20}O_2$ on the basis of their HRESIMS. Their ^{1}H and ^{13}C NMR spectra (Tables 1 and 3) showed two methyl singlets, four aliphatic methylenes, a trisubstituted double bond, and three methines, two of which were oxygenated. By interpretation of HSQC and HMBC spectra, their structures were determined to be based on a 6-isoprenylcyclohexane-1,4-diol framework. The relative configurations of the three metabolites were deduced from ^{1}H NMR coupling constants. The ^{1}H NMR spectrum of **8** revealed axial−axial coupling constants ($J_{a,a}$ = 9.2, 9.2, 11.4 Hz) between H-1/H-2*ax*, H-1/H-6, and H-5*ax*/H-6, and axial−equatorial or equatorial−equatorial coupling constants ($J_{a,e/e,e}$ = 3.7, 2.7, 2.7 Hz) between H-1/H-2*eq*, H-4/H-3, and H-4/H-5. These values were consistent with a chair conformation for the cyclohexane ring in which the 1-OH was in an equatorial position, while the 4-OH and H-6 were in axial positions. The structure of **8** (Figure 2) was also determined by single-crystal X-ray diffraction studies, which confirmed the proposed structure. The $J_{a,a}$ values observed in the ^{1}H NMR spectrum of compound **9** between H-4/H-3*ax* (10.7 Hz), H-4/H-5*ax* (10.7 Hz), and H-5*ax*/H-6 (10.7 Hz), as well as the $J_{a,e/e,e}$ values for H-1/H-2 and H-1/H-6, are in accord with a chair conformation for a cyclohexane ring displaying axial 1-OH and H-6 moieties and an equatorial 4-OH group. In compound **10**, small $J_{a,e/e,e}$ values for H-1/H-2, H-4/H-3, and H-4/H-5 were observed, indicating a chair conformation for the cyclohexane ring and axial orientations of the 1-OH, 4-OH, and H-6 moieties. Therefore, the structures of speciosins H−J (**8**−**10**) were elucidated as those shown in Figure 1.

Speciosin K (**11**) was obtained as a colorless oil. Its molecular formula was determined to be $C_{16}H_{24}O_3$ on the basis of a positive HRESIMS signal at *m/z* 265.1798 (calcd for $C_{16}H_{25}O_3$, 265.1803) and of ^{13}C NMR and DEPT spectra. Its IR spectrum showed absorption bands at 3429, 3283, and 1736 cm^{-1}, which are ascribable to hydroxy and carbonyl groups. The ^{13}C NMR spectrum exhibited 16 carbon signals, including a carbonyl carbon at δ 198.7 (s), a bis-substituted double bond at δ 145.0 (d) and 123.0 (d), a trisubstituted double bond at δ 132.0 (s) and 121.5 (d), a tetrasubstituted double bond at δ 147.8 (s) and 136.4 (s), three methylenes at δ 32.3 (t), 29.9 (t), and 23.8 (t), an oxygenated methine at δ 63.8 (d), an oxygenated fully substituted carbon at δ 71.3 (s), and four methyl signals at δ 29.8 (q), 29.6 (q), 25.7 (q), and 18.0 (q). Correspondingly, the ^{1}H NMR spectrum revealed three olefinic protons at δ 6.69 (d, J = 17.2 Hz), 6.45 (d, J = 17.2 Hz), and 4.89 (t, J = 6.7 Hz), an oxymethine at δ 4.73 (t, J = 3.3 Hz), six upfield methylene protons from 2.16 to 3.12 ppm, and four methyl singlets at δ 1.73 (3H), 1.65 (3H), and 1.41 (6H). The HMBC data showed the following correlations: from H-2 (δ 2.78, 2.42) to C-4 δ (63.8, d) and C-6 (δ 136.4, s); from H-3 (δ 2.13−2.17, m) to C-1 (δ 198.7, s) and C-5 (δ 147.8, s); from H-7 (δ 3.12, d) to C-1, C-5 (δ 147.8, s), C-6 (δ 136.4, s), and C-9 (δ 132.2, s); from H-12 (δ 6.69, d) to C-4, C-6, and C-14 (δ 71.3, s); and from H-13 (δ 6.45, d) to C-5, C-15 (δ 29.8, q), and C-16 (δ 29.6, q). The absolute configuration of **11** was deduced by comparing its specific rotation with those of known analogues. In the case of 4-hydroxy-3-methyl-2-cyclohexen-1-one, the *R* enantiomer has a specific rotation of +34.1 (*c* 1.0, $CHCl_3$), while the *S* enantiomer has a specific rotation of −35.2 (*c* 1.0, $CHCl_3$).[19] Another analogue, (*S*)-2-allyl-4-hydroxycyclohex-2-enone, has a reported specific rotation of −39.3 (*c* 1.0, $CHCl_3$).[20] Since compound **11** has only one stereogenic center and has a negative value for its specific rotation, −14.5 (*c* 0.13, $CHCl_3$), compound **11** was assigned to have the 4*S* configuration. Consequently, the structure of **11** was established as that shown in Figure 1.

It is interesting to note that a metabolite with a core cyclohex-2,3-enone structure was reported to show activity against phytopathogens and plant growth promotion.[1]

Experimental Section

General Experimental Procedures. Melting points were measured on a PHMK 79/2289 micromelting point apparatus and are uncorrected. Optical rotations were measured on a Horiba SEPA-300 polarimeter. UV spectra were recorded on a Shimadzu UV-2401PC spectrophotometer. IR spectra were obtained on a Bruker Tensor 27 FT-IR spectrometer using KBr pellets. NMR spectra were acquired on Bruker DRX-500 and AV-400 instruments at room temperature. MS were recorded on a VG Autospec-3000 spectrometer and an API QSTAR Pulsar i spectrometer. X-ray crystallographic data were collected on a Bruker Smart APEX II CCD diffractometer with graphite-monochromated Mo Kα radiation. Silica gel (200−300 mesh, Qingdao Marine Chemical Inc., China) and Sephadex LH-20 (Amersham Biosciences, Sweden) were used for column chromatography. Preparative HPLC was performed on an Agilent 1100 liquid chromatography system equipped with a Zorbax SB-C_{18} column (9.4 mm × 150 mm).

Fungal Material and Cultivation Conditions. The fungus *H. speciosa* was isolated from a tissue culture of its fruiting bodies collected in the Gaoligong Mountains, Yunnan Province, People's Republic of China, in July 2007, and identified by Prof. Yang Zhu-Liang, Kunming Institute of Botany. A voucher specimen (HFG 07061) was deposited in the Herbarium of the Kunming Institute of Botany. The culture medium consisted of potato (peel) (200 g), glucose (20 g), KH_2PO_4 (3 g), $MgSO_4$ (1.5 g), citric acid (0.1 g), and thiamine hydrochloride (10 mg) per L of deionized H_2O. The pH was adjusted to 6.5 before autoclaving, and the fermentation was carried out in a shaker at 25 °C and 150 rpm for 25 days.

Extraction and Isolation. The entire culture broth of *H. speciosa* (21 L) was initially filtered, and the filtrate extracted three times with EtOAc. The organic layer was concentrated under reduced pressure to give a crude extract (5.3 g), and this residue was subjected to column chromatography over silica gel (200−300 mesh) using a $CHCl_3$−MeOH gradient to afford fractions A−F. Fraction B (879 mg) was further purified by Sephadex LH-20 ($CHCl_3$−MeOH, 1:1) column chromatography and preparative HPLC to yield compound **1** (4.7 mg). Fraction C (450 mg) was purified by Sephadex LH-20 ($CHCl_3$−MeOH, 1:1) column chromatography to provide four subfractions (C1−C4). Each subfraction was further purified by repeated reversed-phase chromatography on a C_{18} column (MeOH−H_2O) and preparative HPLC (MeCN−H_2O). Subsequently, compound **11** (3.8 mg) was obtained from subfraction C1, while compounds **10** (6.1 mg), **8** (2.8 mg), and **9** (2.7 mg) were obtained from subfraction C2. Compounds **3** (5.7 mg) and **2** (31.0 mg) were isolated from subfractions C3 and C4, respectively. Fraction D (248 mg) was subjected to repeated Sephadex LH-20 ($CHCl_3$−MeOH, 1:1) and preparative HPLC (MeCN−H_2O) purification to give **4** (1.0 mg). Fraction E (1.2 g) was chromatographed over Sephadex LH-20 (MeOH), subjected to repeated reversed-phase chromatography on a C_{18} column (MeOH−H_2O), and finally purified by preparative HPLC (MeCN−H_2O) to yield compounds **12** (10.0 mg), **6** (6.5 mg), **7** (2.0 mg), and **5** (103.0 mg).

Speciosin A (1): colorless oil; $[\alpha]^{16}_D$ +297.9 (*c* 0.24, CH_3OH); IR (KBr) ν_{max} 3431, 2929, 2229, 1729, 1689, 1187, 1036 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; FABMS (positive) *m/z* 177 $[M + H]^+$; HRFABMS (positive) *m/z* 177.0557 $[M + H]^+$, calcd for $C_{10}H_9O_3$ 177.0552.

Speciosin B (2): colorless oil; $[\alpha]^{26}_D$ +346.3 (*c* 0.40, $CHCl_3$); UV($CHCl_3$) λ_{max} (log ε) 241 (3.61) nm; IR (KBr) ν_{max} 3449, 3000, 2919, 2256, 1732, 1689, 1411, 1372, 1186, 912 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; EIMS *m/z* (%) 192 $[M]^+$ (10), 163 (55), 97 (100); HRESIMS *m/z* 215.0325 $[M + Na]^+$, calcd for $C_{10}H_8O_4Na$ 215.0320.

Speciosin C (3): colorless oil; $[\alpha]^{24}_D$ +177.7 (*c* 0.28, $CHCl_3$); UV($CHCl_3$) λ_{max} (log ε) 241 (3.65) nm; IR (KBr) ν_{max} 3464, 3374, 2224, 1831, 1613, 1436, 1374, 1299, 1030 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; EIMS *m/z* (%) 192 $[M]^+$ (20), 163 (25), 97 (100); HRESIMS *m/z* 215.0681 $[M + Na]^+$, calcd for $C_{11}H_{12}O_3Na$ 215.0684.

Speciosin D (4): colorless oil; $[\alpha]^{21}_D$ +116.7 (*c* 0.12, $CHCl_3$); IR (KBr) ν_{max} 3407, 2997, 2918, 1636, 1376, 1027 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; ESIMS (positive) *m/z* 217 $[M + Na]^+$, 411 $[2 M + Na]^+$; HRESIMS *m/z* 217.0476 $[M + Na]^+$, calcd for $C_{10}H_{10}O_4Na$ 217.0476.

Speciosin E (5): colorless oil; $[\alpha]^{24}_D$ +152.0 (*c* 0.5, MeOH); IR (KBr) ν_{max} 3396, 2922, 2069, 1630, 1421, 1035 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; FABMS (negative) *m/z* 211 $[M - H]^-$; HRESIMS (negative) *m/z* 211.0605 $[M - H]^-$, calcd for $C_{10}H_{11}O_5$ 211.0606.

Speciosin F (6): colorless oil; $[\alpha]^{16}_D$ +185.7 (*c* 0.11, MeOH); IR (KBr) ν_{max} 3416, 2926, 1633, 1450, 1035 cm^{-1}; ^1H NMR data, see Table 2; ^{13}C NMR data, see Table 1; FABMS (positive) *m/z* 227 $[M + H]^+$; HRESIMS (positive) *m/z* 249.0735 $[M + Na]^+$, calcd for $C_{11}H_{14}O_5Na$, 249.0738.

Speciosin G (7): colorless oil; UV(CH_3OH) λ_{max} (log ε) 326 (3.21), 279 (3.49), 264 (3.48), 250 (3.41), 203 (3.74) nm; IR (KBr) ν_{max} 3443, 2925, 2854, 2368, 2346, 1638 cm^{-1}; ^1H NMR data, see Table 3; ^{13}C NMR data, see Table 1; ESIMS (negative) *m/z* 159 $[M - H]^-$; HRESIMS (negative) *m/z* 159.0452 $[M - H]^-$, calcd for $C_{10}H_7O_2$ 159.0446.

Speciosin H (8): colorless crystals ($CHCl_3$); mp 103−104 °C; $[\alpha]^{24}_D$ −40.5 (*c* 0.11, $CHCl_3$); IR (KBr) ν_{max} 3315, 2960, 2909, 2723, 1494, 1446, 1359, 1059 cm^{-1}; ^1H NMR data, see Table 3; ^{13}C NMR data, see Table 1; FABMS (positive) *m/z* 185 $[M + H]^+$; HRFABMS (positive) *m/z* 207.1363 $[M + Na]^+$, calcd for $C_{11}H_{20}O_2Na$ 207.1360.

Speciosin I (9): colorless crystals ($CHCl_3$); mp 128−129 °C; $[\alpha]^{24}_D$ +15.9 (*c* 0.11, $CHCl_3$); IR (KBr) ν_{max} 3311, 2940, 2930, 2723, 1639, 1438, 1020 cm^{-1}; ^1H NMR data, see Table 3; ^{13}C NMR data, see Table 1; FABMS (positive) *m/z* 185 $[M + H]^+$; HRESIMS (positive) *m/z* 207.1361 $[M + Na]^+$, calcd for $C_{11}H_{20}O_2Na$ 207.1360.

Speciosin J (10): colorless oil; $[\alpha]^{26}_D$ +12.3 (*c* 0.37, $CHCl_3$); IR (KBr) ν_{max} 3383, 3382, 2932, 2857, 2724, 1639, 1452, 1366, 1075, 995 cm^{-1}; ^1H NMR data, see Table 3; ^{13}C NMR data, see Table 1; EIMS *m/z* (%) 184 $[M]^+$ (15), 166 (30), 149 (61), 141 (100); HRESIMS *m/z* 207.1356 $[M + Na]^+$, calcd for $C_{11}H_{20}O_2Na$ 207.1360.

Speciosin K (11): colorless oil; $[\alpha]^{24}_D$ −14.5 (*c* 0.13, $CHCl_3$); UV($CHCl_3$) λ_{max} (log ε) 282 (3.12), 235 (2.84), 226 (2.84), 205 (2.89) nm; IR (KBr) ν_{max} 3429, 3283, 2972, 2924, 2371, 1736, 1636, 1620, 1365, 1153 cm^{-1}; ^1H NMR (400 MHz, C DCl_3) δ 6.69 (1H, d, J = 17.2 Hz, H-12), 6.45 (1H, d, J = 17.2 Hz, H-13), 4.89 (1H, t, J = 6.7 Hz, H-8), 4.73 (1H, t, J = 3.3 Hz, H-4), 3.12 (2H, d, J = 6.7 Hz, H-7), 2.78 (1H, ddd, J = 17.2, 12.0, 6.0 Hz, H-2a), 2.42 (1H, dt, J = 17.2, 4.1 Hz, H-2b), 2.13−2.17 (2H, m, H-3), 1.73 (3H, s, H-10), 1.65 (3H, s, H-11), 1.41 (6H, s, H-15, 16); ^{13}C NMR (400 MHz, $CDCl_3$) δ 198.7 (s, C-1), 147.8 (s, C-5), 145.0 (d, C-13), 136.4 (s, C-6), 132.2 (s, C-9), 123.0 (d, C-12), 121.5 (d, C-8), 71.3 (s, C-14), 63.8 (d, C-4), 32.3 (t, C-2), 29.9 (t, C-3), 29.8 (q, C-15 or C-16), 29.6 (q, C-15 or C-16), 25.7 (q, C-10), 23.8 (t, C-7), 18.0 (q, C-11); EIMS *m/z* (%) 264 $[M]^+$ (1), 246 (5), 205 (100); HRESIMS *m/z* 265.1798 $[M + H]^+$, calcd for $C_{16}H_{25}O_3$ 265.1803.

Aporpinone A (12): colorless oil; $[\alpha]^{29}_D$ −43.9 (*c* 0.08, $CHCl_3$); ^1H NMR (500 MHz, CD_3OD) δ 8.08 (1H, d, J = 5.7, H-4), 6.29 (1H, d, J = 5.7 Hz, H-3), 4.31 (2H, s, H-1″), 3.63 (1H, dd, J = 10.6, 6.3 Hz, H-5′a), 3.52 (1H, dd, J = 10.6, 6.8 Hz, H-5′b), 2.85 (1H, m, H-4′), 1.24 (3H, d, J = 7.0 Hz, H-6′). ^{13}C NMR (500 MHz, $CDOD_3$) δ 171.0 (s, C-2), 155.3 (s, C-5), 141.9 (d, C-4), 120.5 (d, C-3), 111.0 (s, C-1′), 106.7 (s, C-3′), 78.2 (s, C-2′), 67.0 (t, C-5′), 61.2 (t, C-1″), 31.6 (d, C-4′), 17.4 (d, C-6′); EIMS *m/z* (%) 208 $[M]^+$ (28), 190 (10), 178 (61), 160 (100), 147 (27), 131 (58), 77 (38); HRESIMS *m/z* 231.0635 $[M + Na]^+$, calcd for $C_{11}H_{12}O_4Na$ 231.0633.

Crystallographic Data of Speciosin H (8). $C_{11}H_{20}O_2$, MW = 184.27, monoclinic, space group $P2(1)$, a = 6.1288(18) Å, b = 7.166(2) Å, c = 12.816(4) Å, β = 102.805(4)°, V = 548.8(3) Å^3, Z = 2, D_{calcd} = 1.115 g/cm^3, λ = 0.71073 Å, μ(Mo Kα) = 0.074 mm^{-1}, $F(000)$ = 204, and T = 293(2) K. A colorless crystal of dimensions 0.12 × 0.08 × 0.06 mm was selected for X-ray analysis. A total of 3367 reflections, collected in the range 3.26° ≤ θ ≤ 28.53°, yielded 2352 unique reflections. The structure was solved using direct methods and was refined by full-matrix least-squares on F^2 values for 2352 $I > 2\sigma(I)$. Hydrogen atoms were fixed at calculated positions. The final indices were R_1 = 0.0787, wR_2 = 0.1839 with goodness-of-fit = 0.977. Crystallographic data for structure **8** have been deposited at the Cambridge Crystallographic Data Centre as supplementary publication no. CCDC 722765. Copies of the data can be obtained free of charge on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK (fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk).

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Supporting Information Available: 1D and 2D NMR spectra for compounds **1**−**11**. This material is available free of charge via the Internet at http://pubs.acs.org.

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