

# Chaetoindicins A–C, Three Isoquinoline Alkaloids from the Fungus *Chaetomium indicum*

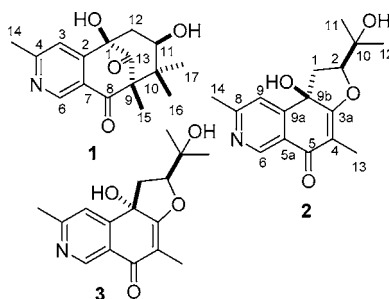
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Received June 21, 2006

## ABSTRACT



Chaetoindicins A–C (1–3), three isoquinolines with novel skeletons, were isolated from the solid-state fermented culture of *Chaetomium indicum*. Their structures were elucidated on the basis of spectral data. X-ray crystallographic analysis confirmed the structure of 2.

Isoquinoline alkaloids, one of the largest groups of natural products, are mainly discovered in plants and possess interesting bioactivities such as cytotoxicity,<sup>1–5</sup> antimalarial,<sup>6</sup> apoptosis,<sup>7</sup> antileukemic,<sup>8</sup> and antimicrobial activities.<sup>9</sup> Only

a limited number of isoquinoline alkaloids have been reported from other sources such as sponges,<sup>10–15</sup> lichens,<sup>16,17</sup> and fungi.<sup>18</sup> In the course to investigate the alkaloids from the fungus *Chaetomium indicum*, three novel isoquinolines with an unprecedented skeleton were isolated from its solid-state fermented rice culture. Here, the isolation and structural elucidation of chaetoindicins A–C are described.

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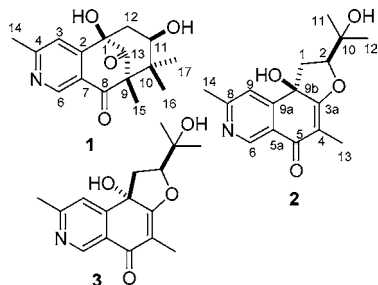
Chaetoindicin A (**1**) was obtained as colorless cubic crystals.<sup>19</sup> Its molecular formula C<sub>16</sub>H<sub>19</sub>NO<sub>4</sub> was established from the quasimolecular ion peak at *m/z* 290.1373 [M + H]<sup>+</sup> in the HRESIMS. Four methyl singlets were recognized in the <sup>1</sup>H NMR spectrum (Table 1). The <sup>13</sup>C NMR spectrum

**Table 1.** NMR Data of **1** in CD<sub>3</sub>OD<sup>a,b</sup>

no.	<b>1</b>	
	$\delta_{\text{H}}$ (mult., <i>J</i> Hz)	$\delta_{\text{C}}$
1		76.4
2		154.3
3	7.74 (s)	118.7
4		165.1
6	8.95 (s)	146.8
7		124.7
8		196.8
9		68.5
10		48.0
11	3.34 (dd, 12.8, 5.0)	70.1
12	2.15 (dd, 12.8, 5.0) 2.24 (t, 12.8)	44.8
13		206.7
14	2.68 (3H, s)	23.2
15	1.33 (3H, s)	10.0
16	0.90 (3H, s)	13.7
17	1.07 (3H, s)	20.3

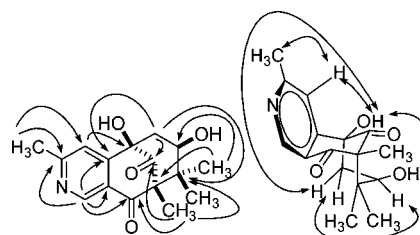
<sup>a</sup> <sup>1</sup>H: 600 MHz; <sup>13</sup>C: 150 MHz. <sup>b</sup> Assignments were based on HSQC and HMBC experiments.

showed 16 signals (Table 1). Two <sup>1</sup>H NMR singlets at  $\delta$  8.95 (H-6) and 7.74 (H-3), five <sup>13</sup>C NMR signals at  $\delta$  154.3 (C-2), 118.7 (C-3), 165.1 (C-4), 146.8 (C-6), and 124.7 (C-7), and the HMBC correlations of H-14/C-4 and C-3, H-3 and H-6/C-2, C-4, and C-7 indicated the presence of a 2-methyl-4,5-substituted pyridine moiety (Figure 2). One conjugated ketonic carbon suggesting by the <sup>13</sup>C NMR signal at  $\delta$  196.8, and the IR absorption at  $\nu_{\text{max}}$  1678 cm<sup>-1</sup>, was assigned as C-8 from the HMBC correlation of H-6/C-8, C-7, and C-2. The connection of C-9–C-10–C-11–C-12–C-1 was deduced from the spin system of H-11/H-12 and the HMBC correlations of H-15/C-9 and C-10, H-11/C-9 and C-10, and H-11 and H-12/C-1. C-9 was connected with C-8 in view of the HMBC correlations of H-15/C-8 and C-9,



**Figure 1.** Structures of **1–3**.

which in turn supported the assignment of C-15. The connection of C-1 with C-2 was confirmed by the HMBC correlations of H-3 and H-12/C-2 and C-1. The <sup>13</sup>C NMR signal at  $\delta$  206.7 and the IR absorption at  $\nu_{\text{max}}$  1733 cm<sup>-1</sup> suggested a ketonic carbon (C-13), which was connected with C-1 and C-9 from the HMBC correlations of H-15/C-9 and C-13 and H-12/C-1 and C-13. CH<sub>3</sub>-16 and CH<sub>3</sub>-17 were assigned upon the HMBC correlations of H-16 and H-17/C-9, C-10, and C-11. Thus, the structure of **1** was finally determined (Figure 1). The relative stereochemistry was determined by the NOESY cross signals of OH-1/H-12a and OH-11 (in DMSO-*d*<sub>6</sub>), H-11/H-12b, and H-17 (Figure 2).



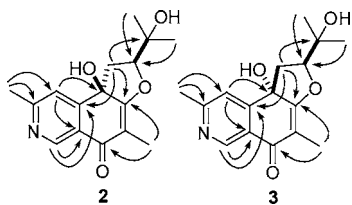
**Figure 2.** Key HMBC and NOESY correlations of **1**.

Chaetoindicin B (**2**) was isolated as colorless cubic crystals.<sup>20</sup> Its molecular formula was determined to be C<sub>16</sub>H<sub>19</sub>NO<sub>4</sub> from the HRESIMS quasimolecular ion peak at *m/z* 290.1381 [M + H]<sup>+</sup>. The IR spectrum suggested the presence of hydroxyl groups (3413 cm<sup>-1</sup>). From the <sup>1</sup>H and <sup>13</sup>C NMR data (Table 2) and HMBC correlations (Figure 3), one 2-methyl-4,5-substituted pyridine moiety and three other methyls were concluded. An  $\alpha,\beta$ -unsaturated ketone

**Table 2.** NMR Data of **2** and **3** in C<sub>5</sub>D<sub>5</sub>N<sup>a,b</sup>

no.	<b>2</b>		<b>3</b>	
	$\delta_{\text{H}}$ (mult., <i>J</i> Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (mult., <i>J</i> Hz)	$\delta_{\text{C}}$
1	2.63 (dd, 13.8, 9.9) 3.18 (dd, 13.8, 1.6) 4.86 (dd, 9.7, 1.4)	35.4	2.63 (dd, 12.3, 10.1) 3.08 (dd, 12.3, 4.5) 5.34 (dd, 10.1, 4.5)	36.8
2		91.6		91.9
3a		174.7		174.9
4		107.2		107.1
5		185.2		185.1
5a		122.0		122.2
6	9.53 (s)	148.0	9.42 (s)	148.1
8		162.1		161.2
9	7.55 (s)	120.3	7.60 (s)	120.2
9a		149.1		149.5
9b		71.5		74.2
10		70.5		69.8
11	1.49 (s)	27.3	1.44 (s)	26.7
12	1.71 (s)	26.8	1.63 (s)	26.1
13	2.08 (s)	8.1	1.98 (s)	8.0
14	2.61 (s)	24.4	2.60 (s)	24.5

<sup>a</sup> <sup>1</sup>H: 600 MHz; <sup>13</sup>C: 150 MHz. <sup>b</sup> Assignments were based on HSQC and HMBC experiments.

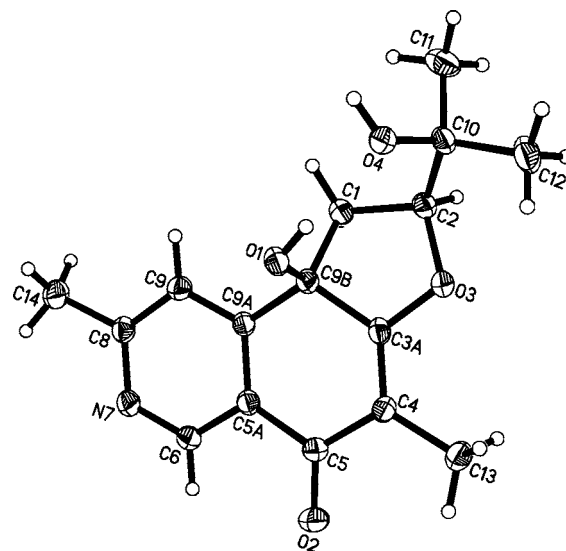


**Figure 3.** Important HMBC correlations of **2** and **3**.

was suggested by the IR peaks at  $\nu_{\max}$  1643  $\text{cm}^{-1}$ , the  $^{13}\text{C}$  NMR signals at  $\delta$  185.2 (C-5), 107.2 (C-4), and 174.7 (C-3a), and the HMBC correlations of H-6/C-5a and C-5, H-13/C-3a, C-4, and C-5. The  $^1\text{H}$  NMR signals at  $\delta$  4.86 (dd,  $J = 9.7, 1.4$  Hz, H-2), 3.18 (dd,  $J = 13.8, 1.6$  Hz, H-1a), and 2.63 (1H, dd,  $J = 13.8, 9.9$  Hz, H-1b) and the HMBC correlations of H-11 and H-12/C-2 and C-10 led to the elucidation of a substructure of  $-\text{CH}_2-\text{CH}-\text{C}(\text{CH}_3)_2$  [C-1–C-2–C-10]. The connection of C-1 to C-9b was determined from the HMBC correlations of H-1/C-2, C-3a, C-9a and C-10, and H-2/C-3a and C-9b. The relative stereochemistry and structure of **2** were confirmed by the X-ray crystallographic analysis (Figure 4).<sup>22</sup>

Chaetoindicin C (**3**) was obtained as a white amorphous powder.<sup>21</sup> Its molecular formula  $\text{C}_{16}\text{H}_{19}\text{NO}_4$  was provided by the HRESIMS quasimolecular ion peak at  $m/z$  290.1376  $[\text{M} + \text{H}]^+$ , which resembled closely that of **2**. Its NMR data were very similar to those of **2**, but the  $^1\text{H}$  NMR signal at  $\delta$  4.86 (H-2) and the  $^{13}\text{C}$  NMR signal at  $\delta$  71.5 (C-9b) in **2** were, respectively, shifted to  $\delta$  5.34 and  $\delta$  74.2 in **3** (Table 2). The HSQC and HMBC correlations led to the structural elucidation of **3**. Therefore, it was a diastereoisomer of **2** (Figure 3).

Most isoquinoline alkaloids discovered from plants exhibit complicated structures, which were mainly synthesized from



**Figure 4.** ORTEP diagram of **2**.

tyrosine, whereas most isoquinolines from lichens, fungi, and sponges show simple skeletons and are possibly biosynthesized via mixed pathways.<sup>10–18</sup> Compounds **1–3** represent a unique and unprecedented carbon skeleton. They are possibly biosynthesized via mixed routes.

**Supporting Information Available:** HRESIMS, 1D and 2D NMR spectra of **1–3**, and X-ray crystallographic data of **2** (CIF). This material is available free of charge via the Internet at <http://pubs.acs.org>.

OL061525K

(19) Compound **1**: colorless crystal, mp 231–232 °C ( $\text{CH}_3\text{OH}$ );  $[\alpha]_{\text{D}}^{20} +8.5^\circ$  ( $c$  0.10,  $\text{CH}_3\text{OH}$ ); IR(KBr)  $\nu_{\max}$  3459, 2974, 2936, 1733, 1768, 1590, 733, 688  $\text{cm}^{-1}$ ; UV( $\text{CH}_3\text{OH}$ )  $\lambda_{\max}$  (log  $\epsilon$ ) 213 (4.29), 248 (4.20) nm;  $^1\text{H}$  and  $^{13}\text{C}$  NMR data, see Table 1; HRESIMS  $m/z$  290.1373  $[\text{M} + \text{H}]^+$ .

(20) Compound **2**: colorless crystal, mp 255–256 °C ( $\text{CH}_3\text{OH}$ );  $[\alpha]_{\text{D}}^{20} -7.5^\circ$  ( $c$  0.10,  $\text{CH}_3\text{OH}$ ); IR(KBr)  $\nu_{\max}$  3413, 2955, 2927, 1643, 1602, 1188, 703  $\text{cm}^{-1}$ ; UV( $\text{CH}_3\text{OH}$ )  $\lambda_{\max}$  (log  $\epsilon$ ) 210 (4.36), 244 (4.20), 285 (3.92), 309 (3.96) nm;  $^1\text{H}$  and  $^{13}\text{C}$  NMR data, see Table 2; HRESIMS  $m/z$  290.1381  $[\text{M} + \text{H}]^+$ .

(21) Compound **3**: white amorphous powder;  $[\alpha]_{\text{D}}^{20} +9.6^\circ$  ( $c$  0.10,  $\text{CH}_3\text{OH}$ ); IR(KBr)  $\nu_{\max}$  3427, 2927, 1641, 1600, 1195, 709  $\text{cm}^{-1}$ ; UV( $\text{CH}_3\text{OH}$ )  $\lambda_{\max}$  (log  $\epsilon$ ) 211 (4.35), 245 (4.21), 285 (3.91), 310 (3.95);  $^1\text{H}$  and  $^{13}\text{C}$  NMR data, see Table 2; HRESIMS  $m/z$  290.1376  $[\text{M} + \text{H}]^+$ .

(22) X-ray data of compound **2**:  $\text{C}_{16}\text{H}_{19}\text{N}_1\text{O}_4$ ;  $M_r = 289.32$ ; orthorhombic, space group  $P2_12_12_1$ ,  $a = 8.777$  (8) Å,  $b = 13.634$  (12) Å,  $c = 12.888$  (18) Å,  $\alpha = 90^\circ$ ,  $\beta = 90^\circ$ ,  $\gamma = 109.04^\circ$ ,  $V = 1457.78$  (43) Å<sup>3</sup>,  $Z = 4$ ,  $D_{\text{calcd}} = 1.318$  g/cm<sup>3</sup>,  $\lambda = 0.71073$  Å,  $\mu$  (Mo K $\alpha$ ) = 0.095 mm<sup>-1</sup>,  $F(000) = 616$ ,  $T = 287$  (2) K. Of the 3841 reflections that were collected, 3354 were unique ( $R_{\text{int}} = 0.0157$ ). The structure was solved by direct methods with SHELXL-97 and refined by full-matrix least-squares on  $F^2$ . Final refinement: data/restraints/parameters = 3354/0/195;  $R1 = 0.0417$  (all data),  $wR2 = 0.0936$  (all data). Absolute structure parameter = 0.0090 (12) and  $\text{GOF} = 0.945$ . The maximum and minimum peaks on the final difference Fourier map corresponded to 0.229 and  $-0.159$  e<sup>-</sup>/Å<sup>3</sup>, respectively. Crystallographic data (including structure factors) for compounds **2** (CCDC609952) reported in this paper have been deposited with the Cambridge Crystallographic Data Center. Copies of the data can be obtained, free of charge, on application to CCDC, 12 Union Road, CB2 1EZ, UK [fax: +44-0-1223-336033 or e-mail: deposit@ccdc.cam.ac.uk].