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# Cytotoxic Neoclerodane Diterpenoids from *Scutellaria barbata*

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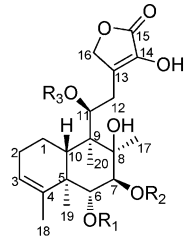
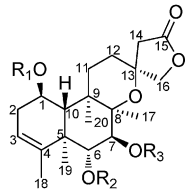
Seven new neoclerodane diterpenoids, scutebatas A−G (**1**−**7**), have been isolated from *Scutellaria barbata*. Compounds **1**−**3** possess a rare α-hydroxy group in their α,β-unsaturated lactone rings. Their structures were elucidated by spectroscopic analysis, and the relative configuration of scutebata A was deduced using ROESY data and the computational DFT method. Compounds **1**, **2**, **4**, **5**, and **6** were evaluated for *in vitro* cytotoxicity against six human cancer cell lines: HL-60, SMMC-7721, A-549, SK-BR-3, CACO-2, and PANC-1. Scutebata A (**1**) showed weak cytotoxicity against SK-BR-3 with an $IC_{50}$ value of 15.2 μM.

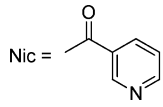
*Scutellaria* is a cosmopolitan genus of the Lamiaceae (or Labiatae) family.[1] The use of species belonging to this genus in Chinese traditional medicine has a long history. For example, dried whole plants of *Scutellaria barbata* D. Don, named "Ban zhi lian" in Chinese, are commonly used in folk medicines to treat tumors, hepatitis, cirrhosis, and other diseases.[2−4] In recent years, several cytotoxic neoclerodane diterpenoids have been isolated from *S. barbata*.[5−9] With the aim of discovering compounds with potential antitumor properties, we initiated a phytochemical study of the aerial parts of *S. barbata*, which led to the isolation of seven new neoclerodane diterpenoids, scutebatas A−G (**1**−**7**). Importantly, scutebatas A−C (**1**−**3**) were shown to possess a rare α-hydroxy group in their α,β-unsaturated lactone rings. In this paper, we describe the isolation and structural identification of scutebatas A−G and the *in vitro* cytotoxic activities of compounds **1**, **2**, **4**, **5**, and **6** against six human cancer cell lines: HL-60, SMMC-7721, A-549, SK-BR-3, CACO-2, and PANC-1.



| | $R_1$ | $R_2$ | $R_3$ |
|---|---|---|---|
| 1 | Bz | Bz | Ac |
| 2 | Nic | Bz | Ac |
| 3 | Nic | H | Ac |



| | $R_1$ | $R_2$ | $R_3$ |
|---|---|---|---|
| 4 | Bz | Ac | Ac |
| 5 | MePr | Ac | Ac |
| 6 | Nic | Ac | Ac |
| 7 | Bz | Nic | Bz |




Scutebata A (**1**), obtained as white powder, had the molecular formula $C_{36}H_{40}O_{10}$ according to its HRESIMS at *m/z* 655.2527 [M + Na]$^+$ (calcd 655.2519). Its IR data exhibited absorption bands for a hydroxy (3431 cm$^{-1}$), ester carbonyls (1727, 1762 cm$^{-1}$), an olefinic bond (1638 cm$^{-1}$), and phenyl (1604, 1026, 710 cm$^{-1}$) groups. In addition to the signals of one acetoxy and two benzoyloxy groups, the $^{13}$C NMR spectrum revealed 20 carbon resonances, which were classified into seven quaternary carbons (4 × sp$^2$, 3 × sp$^3$), five methines (1 × sp$^2$, 4 × sp$^3$), four sp$^3$ methylenes, and four methyl groups. The $^1$H NMR spectrum of compound **1** showed signals of four methyl singlets [$\delta_H$ 0.89 (s); 1.13 (s); 1.38 (s); 1.50 (s)], a broad singlet at $\delta_H$ 5.27 (1H), an AB system [$\delta_H$ 5.63 (d, $J$ = 10.2 Hz), 5.53 (d, $J$ = 10.2 Hz)], and an ABX system [$\delta_H$ 5.37 (d, $J$ = 10.8 Hz), 2.74 (dd, $J$ = 13.8, 10.8 Hz), 3.22 (d, $J$ = 13.8 Hz)]. Comparison of $^1$H and $^{13}$C NMR spectra of compound **1** (Table 1) with scuterulein A[10] suggested that compound **1** is a neoclerodane diterpenoid with an oxygenated substituent at C-11, which was confirmed by analysis of the 2D NMR data. HMBC correlations of H-6/C-1′, H-7/C-1″, and H-11/C-1‴ also established that benzoyloxy groups were present at C-6 and C-7 and that the acetoxy group was present at C-11. Furthermore, the HMBC correlations (Figure 1) between $H_2$-16 and C-13 (129.9 s), C-14 (138.3 s), and C-15 (169.7 s) indicated the presence of an α,β-unsaturated lactone ring with a hydroxy group at C-14.

The ROESY correlations of H-7/$H_3$-17, $H_3$-19, and $H_3$-20 indicated that they were cofacial and α-orientated, whereas the ROESY correlation of H-10/H-6 indicated that they were on the opposite face and β-oriented. The relative configuration of C-11 was deduced from the ROESY experiment and molecular modeling (Gaussian 03 D.01)[11] using *ab initio* calculations. The ROESY correlations of H-11/$H_3$-17, $H_3$-20, and $H_b$-16 suggested that in the preferred conformation of the pendant chain, H-11 was close to these groups. DFT calculations applied to compound **1** indicated that, in the minimum energy conformation, the calculated interatomic distances were H-11···Me-20 = 2.66 Å, H-11···Me-17 = 1.96 Å, and H-11···$H_b$-16 = 2.93 Å, which accounts for the cross-peaks observed in the ROESY spectrum and indicated that compound **1** had an 11*S** relative configuration. Together, these results showed that compound **1** was 11(*S**)-11-acetoxy-6α,7β-dibenzoyloxy-8β,14-dihydroxy-3,13(14)-neocleroden-15,16-olide.

Scutebata B (**2**), obtained as a white, amorphous powder, had a molecular formula of $C_{35}H_{39}NO_{10}$ according to its HRESIMS at *m/z* 656.2470 [M + Na]$^+$ (calcd 656.2471). Comparison of the $^1$H and $^{13}$C NMR spectra of compounds **2** and **1** showed that they were closely related, with the exception that one benzoyloxy group in compound **1** was replaced by one nicotinoyloxyl group in compound **2**. The $^1$H−$^1$H COSY, HMQC, HMBC, and ROESY data further confirmed that the structure of compound **2** was 11(*S**)-11-acetoxy-6α-nicotinoyloxy-7β-benzoyloxy-8β,14-dihydroxy-3,13(14)-neocleroden-15,16-olide.

The molecular formula of scutebata C (**3**) was found to be $C_{28}H_{35}NO_9$ according to its HRESIMS at *m/z* 552.2212 [M + Na]$^+$

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**Table 1.** $^1$H NMR Data of Scutebatas A−G (**1**−**7**) ($\delta$ in ppm, $J$ in Hz)

| no. | **1**[b] | **2**[b] | **3**[b] | **4**[c] | **5**[c] | **6**[c] | **7**[c] |
|---|---|---|---|---|---|---|---|
| 1α | 1.76, m | 1.76, m | 1.69, m | 5.76, dt (9.6, 6.2) | 5.40, dt (9.6, 6.2) | 5.76, dt (9.6, 6.2) | 5.84, dt (9.6, 6.1) |
| 1β | 2.05,[a] | 2.05,[a] | 2.00,[a] | | | | |
| 2α | 2.09,[a] | 2.10,[a] | 2.07,[a] | 2.71, m | 2.59, m | 2.68,[a] | 2.77, m |
| 2β | 2.17,[a] | 2.18,[a] | 2.13,[a] | 2.17, m | 1.97,[a] | 2.15, m | 2.23, m |
| 3 | 5.27, br s | 5.29, br s | 5.23, br s | 5.31, br s | 5.28, br s | 5.29, br s | 5.36, br s |
| 6 | 5.63, d (10.2) | 5.65, d (10.2) | 5.20, d (10.2) | 5.41, d (10.2) | 5.36, d (10.2) | 5.38, d (10.2) | 5.88, d (10.2) |
| 7 | 5.53, d (10.2) | 5.54, d (10.2) | 3.69, d (10.2) | 5.25, d (10.2) | 5.22, d (10.2) | 5.22, d (10.2) | 5.66, d (10.2) |
| 10 | 2.35, d (12.0) | 2.35, d (12.0) | 2.21, d (12.0) | 2.72, d (9.6) | 2.53, d (9.6) | 2.70, d (9.6) | 2.87, d (9.6) |
| 11α | 5.37, br d (10.8) | 5.37, br d (10.8) | 5.36, br d (10.8) | 1.56, m | 1.59,[a] | 1.58, m | 1.64,[a] |
| 11β | | | | 2.07,[a] | 2.02,[a] | 2.01,[a] | 2.16,[a] |
| 12α | 2.74, dd (13.8, 10.8) | 2.73, dd (13.8, 10.8) | 2.69, dd (13.8, 10.8) | 1.68,[a] | 1.70,[a] | 1.68,[a] | 1.74, m |
| 12β | 3.22, br d (13.8) | 3.22, br d (13.8) | 3.15, br d (13.8) | 2.09,[a] | 1.98,[a] | 2.03,[a] | 2.17,[a] |
| 14α | | | | 2.57, d (17.2) | 2.53, d (17.4) | 2.54, d (17.2) | 2.68, d (17.2) |
| 14β | | | | 3.14, d (17.2) | 3.10, d (17.4) | 3.09, d (17.2) | 3.15, d (17.2) |
| 16α | 4.59, d (16.4) | 4.59, d (16.4) | 4.57, d (16.4) | 4.12, d (8.6) | 4.12, d (8.6) | 4.10, d (8.6) | 4.12, d (8.9) |
| 16β | 4.67, d (16.4) | 4.67, d (16.4) | 4.64, d (16.4) | 4.19, d (8.6) | 4.22, d (8.6) | 4.16, d (8.6) | 4.20, d (8.9) |
| 17 | 1.13, 3H, s | 1.13, 3H, s | 1.20, 3H, s | 1.12, 3H, s | 1.12, 3H, s | 1.11, 3H, s | 1.20, 3H, s |
| 18 | 1.50, 3H, s | 1.51, 3H, s | 1.52, 3H, s | 1.67, 3H, s | 1.63, 3H, s | 1.65, 3H, s | 1.68, 3H, s |
| 19 | 1.38, 3H, s | 1.38, 3H, s | 1.28, 3H, s | 1.36, 3H, s | 1.29, 3H, s | 1.34, 3H, s | 1.57, 3H, s |
| 20 | 0.89, 3H, s | 0.89, 3H, s | 0.78, 3H, s | 1.09, 3H, s | 1.05, 3H, s | 1.07, 3H, s | 1.20, 3H, s |
| 2′ | | | | | 2.24, m | | |
| 3′ | 7.66, m | 8.76, d (1.6) | 9.15, br s | 7.95, br d (7.2) | 1.12, 3H, d (7.0) | 9.12, d (1.7) | 7.99, m |
| 4′ | 7.34, m | | | 7.46, br t (7.2) | 1.14, 3H, d (7.0) | | 7.31, m |
| 5′ | 7.50, m | 8.66, dd (4.8, 1.6) | 8.85, d (5.0) | 7.59, br t (7.2) | | 8.78, br d (4.9) | 7.47, m |
| 6′ | 7.34, m | 7.39,[a] | 7.63, dd (7.9, 5.0) | 7.46, br t (7.2) | | 7.40, dd (4.9, 7.9) | 7.31, m |
| 7′ | 7.66, m | 7.97, dt (8.0, 1.6) | 8.37, br d (7.9) | 7.95, br d (7.2) | | 8.18, br d (7.9) | 7.99, m |
| 2″ | | | | 2.01, 3H, s | 1.99, 3H, s | 1.99, 3H, s | |
| 3″ | 7.82, m | 7.81, m | | | | | 8.92, d (1.9) |
| 4″ | 7.34, m | 7.36,[a] | | | | | |
| 5″ | 7.50, m | 7.54, m | | | | | 8.61, d (4.8) |
| 6″ | | | | | | | 7.19, m |
| 7″ | | | | | | | 7.96, m |
| 2‴ | 1.99, 3H, s | 2.00, 3H, s | 1.96, 3H, s | 2.09, 3H, s | 2.08, 3H, s | 2.06, 3H, s | |
| 3‴, 7‴ | | | | | | | 7.82, m |
| 4‴, 6‴ | | | | | | | 7.49, m |
| 5‴ | | | | | | | 7.61, br t |

[a] Overlapped, without denoting multiplicity. [b] In DMSO-$d_6$. [c] In $CDCl_3$.

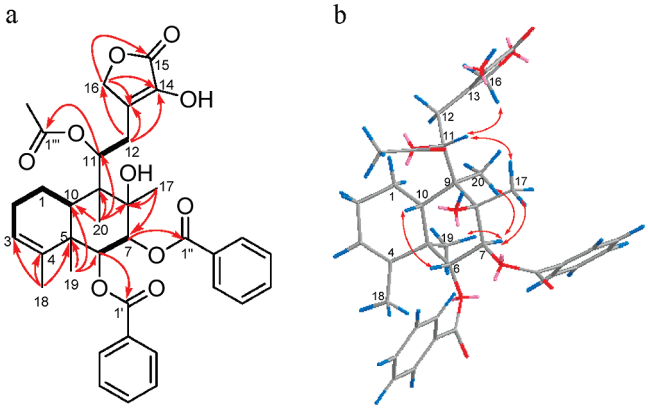


**Figure 1.** (a) $^1$H−$^1$H COSY (—) and selected HMBC (→) correlations of **1**. (b) ROESY (↔) correlations of **1**.

(calcd 552.2209 for $C_{28}H_{35}NO_9Na$). Its $^1$H and $^{13}$C NMR spectra showed a similar signal pattern to those of compound **2**, with the exception that a benzoyloxy group was absent in compound **2**. The 2D NMR data established that compound **3** was 11(*S**)-11-acetoxy-6α-nicotinoyloxy-7β-hydroxy-8β,14-dihydroxy-3,13(14)-neocleroden-15,16-olide.

Scutebata D (**4**) was assigned the molecular formula $C_{31}H_{38}O_9$ from its HRESIMS, which displayed a quasi-molecular ion at *m*/*z* 577.2431 [M + Na]$^+$ (calcd 577.2413). Its IR data showed absorptions for a γ-spirolactone (1787 cm$^{-1}$), ester carbonyls (1749, 1720, 1710 cm$^{-1}$), an olefinic bond (1638, 1629 cm$^{-1}$), and a phenyl group (1026, 716 cm$^{-1}$). The $^1$H and $^{13}$C NMR data (Tables 1 and 2) exhibited one benzoyloxy and two acetoxy groups. In addition, the 1D NMR data showed signals of a neoclerodane diterpenoid with a 3-en-γ-13-spiro-15,16-lactone moiety [$\delta_H$ 1.09 (s, $H_3$-20), 1.12 (s, $H_3$-17), 1.36 (s, $H_3$-19), 1.67 (s, $H_3$-18), 5.31 (br s, H-3), 2.57 (d, $J$ = 17.3 Hz, $H_\alpha$-14), 3.14 (d, $J$ = 17.3 Hz, $H_\beta$-14), 4.12 (d, $J$ = 8.6 Hz, $H_\alpha$-16), 4.19 (d, $J$ = 8.6 Hz, $H_\beta$-16), $\delta_C$ 21.1 (q, C-20), 19.6 (q, C-17), 16.6 (q, C-19), 20.0 (q, C-18), 120.2 (d, C-3), 143.1 (s, C-4), 44.3 (t, C-14), 76.5 (t, C-16)].[4,6] Detailed analysis of the 2D NMR data, including the HMQC, $^1$H−$^1$H COSY, and HMBC data (Figure 2), confirmed the above findings. The locations of benzoyloxy and acetoxy groups were determined by the HMBC correlations from H-1 ($\delta$ 5.76) to C-1′, H-6 ($\delta$ 5.41) to C-1″, and H-7 ($\delta$ 5.25) to C-1‴, respectively.

The relative configuration of compound **4** was deduced from its ROESY spectrum. The ROESY correlations of H-7/$H_3$-17, $H_3$-19, $H_3$-20 and H-1/$H_3$-19, $H_3$-20 indicated that they were cofacial and α-orientated, whereas the ROESY correlation of H-10/H-6 indicated that they were on the opposite face and β-oriented. The configuration of C-13 was determined to be *S** by the ROESY correlation between $H_2$-16 and $H_3$-17.[12] Thus, the structure of compound **4** was found to be 13(*S**)-1β-benzoyloxy-6α,7β-diacetoxy-8β,13-epoxy-3-neocleroden-15,16-olide.

A comparison of $^1$H and $^{13}$C NMR data showed that scutebatas E (**5**, $C_{28}H_{38}O_9$) and F (**6**, $C_{30}H_{37}NO_9$) possessed the same 1β,6α,7β-trihydroxy-8β,13-epoxy-3-neocleroden-15,16-olide structural framework as scutebata D (**4**). The only difference between these compounds was the presence of an ester substituent at C-1 in compounds **5** and **6**, which was corroborated by their HMBC correlations. The 1β-benzoyloxy group in compound **4** was replaced by an isobutyroyloxy group in compound **5** [$\delta_H$ 1.12 (3H, d, $J$ = 7.0 Hz), 1.14 (3H, d, $J$ = 7.0 Hz), 2.24 (1H, m); $\delta_C$ 176.3 (s), 34.3 (d), 19.2 (q), 18.5 (q)] and a nicotinoyloxy group in compound **6** [$\delta_H$ 9.12 (d, $J$ = 1.7 Hz), 8.78 (br d, $J$ = 4.9 Hz), 7.40 (dd, $J$ = 4.9, 7.9 Hz), 8.18 (br d, $J$ = 7.9 Hz); $\delta_C$ 164.3 (s), 125.8 (s), 150.7

**Table 2.** $^{13}$C NMR Data of Scutebatas A−G (**1**−**7**)

| no. | **1**[a] | **2**[a] | **3**[a] | **4**[b] | **5**[b] | **6**[b] | **7**[b] |
|---|---|---|---|---|---|---|---|
| 1 | 18.9 $CH_2$ | 18.9 $CH_2$ | 18.8 $CH_2$ | 70.8 CH | 70.3 CH | 71.6 CH | 70.9 CH |
| 2 | 25.4 $CH_2$ | 25.4 $CH_2$ | 25.4 $CH_2$ | 33.0 $CH_2$ | 32.8 $CH_2$ | 33.0 $CH_2$ | 33.1 $CH_2$ |
| 3 | 123.2 CH | 123.4 CH | 122.7 CH | 120.2 CH | 120.1 CH | 119.9 CH | 120.5 CH |
| 4 | 140.9 C | 140.8 C | 141.4 C | 143.1 C | 143.4 C | 143.2 C | 143.1 C |
| 5 | 42.6 C | 42.5 C | 42.1 C | 44.2 C | 44.1 C | 44.2 C | 44.7 C |
| 6 | 74.9 CH | 75.7 CH | 78.3 CH | 73.2 CH | 73.1 CH | 73.0 CH | 74.6 CH |
| 7 | 75.3 CH | 75.3 CH | 72.2 CH | 74.1 CH | 74.0 CH | 73.9 CH | 74.5 CH |
| 8 | 76.9 C | 76.8 C | 77.8 C | 80.8 C | 80.7 C | 80.7 C | 81.2 C |
| 9 | 47.0 C | 47.0 C | 46.4 C | 38.7 C | 38.6 C | 38.6 C | 38.9 C |
| 10 | 40.1 CH | 40.1 CH | 39.9 CH | 43.1 CH | 43.1 CH | 43.0 CH | 43.5 CH |
| 11 | 75.1 CH | 75.0 CH | 75.3 CH | 28.5 $CH_2$ | 28.3 $CH_2$ | 28.5 $CH_2$ | 28.6 $CH_2$ |
| 12 | 28.2 $CH_2$ | 28.2 $CH_2$ | 28.0 $CH_2$ | 29.3 $CH_2$ | 29.3 $CH_2$ | 29.2 $CH_2$ | 29.3 $CH_2$ |
| 13 | 129.9 C | 129.9 C | 130.2 C | 76.5 C | 76.3 C | 76.4 C | 77.0 C |
| 14 | 138.3 C | 138.3 C | 138.1 C | 44.3 $CH_2$ | 44.2 $CH_2$ | 44.2 $CH_2$ | 44.5 $CH_2$ |
| 15 | 169.7 C | 169.8 C | 169.6 C | 173.7 C | 173.7 C | 173.4 C | 173.7 C |
| 16 | 68.7 $CH_2$ | 68.7 $CH_2$ | 68.6 $CH_2$ | 76.5 $CH_2$ | 76.4 $CH_2$ | 76.3 $CH_2$ | 76.6 $CH_2$ |
| 17 | 20.5 $CH_3$ | 20.5 $CH_3$ | 21.0 $CH_3$ | 19.6 $CH_3$ | 19.6 $CH_3$ | 19.5 $CH_3$ | 19.8 $CH_3$ |
| 18 | 19.8 $CH_3$ | 19.8 $CH_3$ | 19.9 $CH_3$ | 20.0 $CH_3$ | 20.0 $CH_3$ | 20.0 $CH_3$ | 20.2 $CH_3$ |
| 19 | 17.0 $CH_3$ | 16.9 $CH_3$ | 16.8 $CH_3$ | 16.6 $CH_3$ | 16.4 $CH_3$ | 16.6 $CH_3$ | 16.8 $CH_3$ |
| 20 | 16.1 $CH_3$ | 16.1 $CH_3$ | 15.9 $CH_3$ | 21.1 $CH_3$ | 21.1 $CH_3$ | 21.0 $CH_3$ | 21.2 $CH_3$ |
| 1′ | 165.2 C | 164.2 C | 164.4 C | 165.6 C | 176.3 C | 164.3 C | 165.7 C |
| 2′ | 128.9 C | 125.4 C | 126.8 C | 130.0 C | 34.3 CH | 125.8 C | 128.9 C |
| 3′ | 128.8 CH | 149.6 CH | 149.6 CH | 129.4 CH | 18.5 $CH_3$ | 150.7 CH | 129.5 CH |
| 4′ | 128.1 CH | | | 128.7 CH | 19.2 $CH_3$ | | 128.3 CH |
| 5′ | 133.1 CH | 153.6 CH | 152.9 CH | 133.4 CH | | 153.8 CH | 133.3 CH |
| 6′ | 128.1 CH | 123.7 CH | 124.0 CH | 128.7 CH | | 123.5 CH | 128.3 CH |
| 7′ | 128.8 CH | 136.6 CH | 137.2 CH | 129.4 CH | | 136.7 CH | 129.5 CH |
| 1″ | 165.7 C | 165.7 C | | 169.9 C | 169.8 C | 169.7 C | 163.5 C |
| 2″ | 129.8 C | 128.7 C | | 21.5 $CH_3$ | 21.4 $CH_3$ | 21.4 $CH_3$ | 125.9 C |
| 3″ | 129.5 CH | 129.4 CH | | | | | 150.7 CH |
| 4″ | 128.4 CH | 128.2 CH | | | | | |
| 5″ | 133.2 CH | 133.4 CH | | | | | 153.3 CH |
| 6″ | 128.4 CH | 128.2 CH | | | | | 123.1 CH |
| 7″ | 129.5 CH | 129.4 CH | | | | | 136.7 CH |
| 1‴ | 170.6 C | 170.6 C | 170.4 C | 170.9 C | 170.9 C | 170.8 C | 166.3 C |
| 2‴ | 20.6 $CH_3$ | 20.7 $CH_3$ | 20.6 $CH_3$ | 20.8 $CH_3$ | 20.8 $CH_3$ | 20.7 $CH_3$ | 130.0 C |
| 3‴, 7‴ | | | | | | | 129.8 CH |
| 4‴, 6‴ | | | | | | | 128.7 CH |
| 5‴ | | | | | | | 133.5 CH |

[a] In DMSO-$d_6$. [b] In $CDCl_3$.

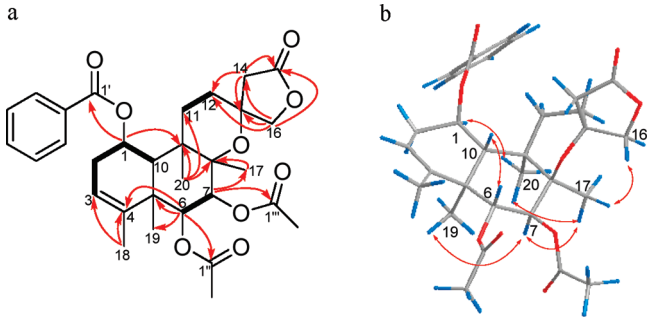


**Figure 2.** (a) $^1$H−$^1$H COSY (—) and selected HMBC (→) correlations of **4**. (b) ROESY (↔) correlations of **4**.

(d), 153.8 (d), 123.5 (d), 136.7 (d)]. The ROESY data also showed that the relative configurations of compounds **4**, **5**, and **6** were identical.

Scutebata G (**7**) was assigned to the molecular formula $C_{40}H_{41}NO_9$ by positive HRESIMS. Its NMR data were similar to those of compound **4**, with the exception that a nicotinoyloxy group and a benzoyloxy group in compound **7** replaced two acetoxy groups in compound **4**. HMBC correlations of H-1 ($\delta_H$ 5.84)/C-1′, H-6 ($\delta_H$ 5.88)/C-1″, C-19, and H-7 ($\delta_H$ 5.66)/C-1‴ indicated that a nicotinoyloxy group is present at C-6 and that benzoyloxy groups are present at C-1 and C-7. The structure of scutebata G was thus determined to be 13(*S**)-1$\beta$,7$\beta$-dibenzoyloxy-6$\alpha$-nicotinoyloxy-8$\beta$,13-epoxy-3-neocleroden-15,16-olide.

Compounds **1**, **2**, **4**, **5**, and **6** were tested for *in vitro* cytotoxicity against six human cancer cell lines (HL-60, SMMC-7721, A-549, SK-BR-3, CACO-2, and PANC-1) using the MTT method. Scutebata A (**1**) showed weak cytotoxic activity against SK-BR-3 with an $IC_{50}$ value of 15.2 $\mu$M.

## Experimental Section

**General Experimental Procedures.** Optical rotations were measured on a JASCO DIP-370 digital polarimeter. UV spectra were obtained using a Shimadzu UV-2401A spectropolarimeter. A Tenor 27 spectrophotometer was used for scanning IR spectroscopy. NMR spectra were recorded on Bruker AM-400 and DRX-500 spectrometers with TMS as internal standard. ESIMS were recorded using a Finnigan MAT 90 instrument, and FABMS spectra were recorded using a VG Auto Spec-3000 spectrometer. Column chromatography was performed on silica gel (200−300 mesh, Qingdao Marine Chemical Inc., Qingdao, People's Republic of China), Sephadex LH-20 (40−70 $\mu$m, Amersham Pharmacia Biotech AB, Uppsala, Sweden), and Lichroprep RP-18 gel (40−63 $\mu$m, Merck Darmstadt, Germany). HPLC separations were performed using an Agilent 1100 series pump equipped with a UV detector and a Zorbax SB-C18 (10 $\mu$m, Agilent Co. Ltd. Wilmington, DE) column (i.d. 9.4 × 250 mm).

**Plant Material.** The dried aerial parts of *S. barbata* (20 kg) were collected in Guangxi Province of China and were sourced from Kunming Juhua medicinal material market in September 2007. A voucher specimen (HH2007092801) has been deposited at the State Key Laboratory of Phytochemistry and Plant Resources in West China, Kunming Institute of Botany, Chinese Academy of Sciences.

**Extraction and Isolation.** Dried and powered aerial parts of *S. barbata* (20 kg) were extracted with MeOH (3 × 80 L) at 50 °C. After evaporation of the solvent, the residue was dissolved in $H_2O$ and extracted successively with petroleum ether, EtOAc, and *n*-BuOH. The EtOAc extract (220 g) was chromatographed over silica gel (200−300 mesh) and eluted with petroleum ether/EtOAc (9:1, 7:3, 1:1, 0:1) to

produce four fractions (A−D). Fraction B (54 g) was purified by column chromatography over MCI gel using 90% MeOH/$H_2O$ and MeOH as eluents. The fraction eluted by 90% MeOH was further purified by Sephadex LH-20 (eluted with MeOH) and silica gel (eluted with petroleum ether/$Me_2CO$, 9:1) to produce compounds **1** (63 mg), **4** (28 mg), and **5** (31 mg). Fraction C (48 g) was subjected to an RP-18 column (eluted with MeOH/$H_2O$ at 30, 45, 60, 75, 90%) to produce five fractions (C1−C5). Fraction C3 was purified by Sephadex LH-20 in MeOH to produce fractions C3-1, C3-2, and C3-3. Fraction C3-1 was subjected to semipreparative HPLC using MeOH/$H_2O$ (70%) as the mobile phase (2 mL/min, 30 °C) to yield compound **3** (2.9 mg), whereas fraction C3-2 was subjected to CC on silica gel eluted with petroleum/$Me_2CO$ (8:2−7:3) to produce compounds **2** (31 mg) and **6** (74 mg). Fraction C3-3 was subjected to CC on silica gel eluted with petroleum/EtOAc (7:3) to yield compound **7** (4.2 mg).

**Scutebata A (1):** white, amorphous powder; $[\alpha]_D^{21}$ −62.1 (*c* 0.065, MeOH); IR (KBr) $\nu_{max}$ 3432, 1762, 1727, 1638, 1604, 1278, 1119, 710 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive FABMS *m/z* 633 $[M]^+$; HRESIMS *m/z* 655.2527 $[M + Na]^+$ (calcd 655.2519 for $C_{36}H_{40}O_{10}Na$).

**Scutebata B (2):** white, amorphous powder; $[\alpha]_D^{23}$ −82.6 (*c* 0.065, $CHCl_3$/MeOH, 1:1); IR (KBr) $\nu_{max}$ 3476, 1763, 1726, 1598, 1286, 1112, 711 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 634 $[M + 1]^+$; HRESIMS *m/z* 656.2470 $[M + Na]^+$ (calcd 656.2471 for $C_{35}H_{39}NO_{10}Na$).

**Scutebata C (3):** white, amorphous powder; $[\alpha]_D^{23}$ −34.8 (*c* 0.13, MeOH); IR (KBr) $\nu_{max}$ 3433, 1753, 1736, 1721, 1710, 1638, 1629, 1285, 1027, 592 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 530 $[M + 1]^+$; HRESIMS *m/z* 552.2212 $[M + Na]^+$ (calcd 552.2209 for $C_{28}H_{35}NO_9Na$).

**Scutebata D (4):** white, amorphous powder; $[\alpha]_D^{26}$ −40.4 (*c* 0.095, $CHCl_3$); IR (KBr) $\nu_{max}$ 3431, 1786, 1749, 1720, 1710, 1638, 1629, 1247, 1026, 716 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 577 $[M + Na]^+$; HRESIMS *m/z* 577.2431 $[M + Na]^+$ (calcd 577.2413 for $C_{31}H_{38}O_9Na$).

**Scutebata E (5):** white, amorphous powder; $[\alpha]_D^{23}$ −49.0 (*c* 0.26, $CHCl_3$); IR (KBr) $\nu_{max}$ 3436, 2976, 1791, 1749, 1374, 1248, 1027 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 543 $[M + Na]^+$; HRESIMS *m/z* 543.2589 $[M + Na]^+$ (calcd 543.2570 for $C_{28}H_{40}O_9Na$).

**Scutebata F (6):** white, amorphous powder; $[\alpha]_D^{23}$ −47.2 (*c* 0.26, $CHCl_3$); IR (KBr) $\nu_{max}$ 3440, 2979, 1788, 1748, 1719, 1591, 1248, 1025 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 578 $[M + Na]^+$; HRESIMS *m/z* 578.2389 $[M + Na]^+$ (calcd 578.2366 for $C_{30}H_{37}NO_9Na$).

**Scutebata G (7):** white, amorphous powder; $[\alpha]_D^{26}$ −117.2 (*c* 0.15, $CHCl_3$); IR (KBr) $\nu_{max}$ 3424, 1792, 1721, 1639, 1591, 1277, 1104, 1025, 713 $cm^{-1}$; for $^1H$ and $^{13}C$ NMR data see Tables 1 and 2; positive ESIMS *m/z* 680 $[M + 1]^+$; HRESIMS *m/z* 702.2668 $[M + Na]^+$ (calcd 702.2679 for $C_{40}H_{41}NO_9Na$).

**Cytotoxicity Assays.** HL-60, SMMC-7721, A-549, SK-BR-3, CACO-2, and PANC-1 cell lines were maintained in RPMI 1640 medium and seeded in 96-well tissue culture plates. After 12 h incubation at 37 °C, the test compound (40 $\mu$M) was added and the plate was further incubated for 48 h. Cell growth was then evaluated using an MTT assay procedure.[13] Compounds that inhibited 50% of the growth of the cancer cells were evaluated again at five concentrations; each concentration of the compound was tested in three parallel wells. The $IC_{50}$ value was calculated using the Reed−Muench method. Cisplatin was included as a positive control.

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**Supporting Information Available:** 1D and 2D NMR spectra of scutebatas A (**1**) and D (**4**) and 1D NMR spectra of compounds **2**, **3**, **5**, **6**, and **7** are available free of charge via the Internet at http://pubs.acs.org.

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