SUPPLEMENTARY MATERIAL
Two new polyhydroxysterols produced by Fusarium solani, an endophytic fungus from Chloranthus multistachys

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A highly antagonistic endophytic fungus, designated strain CL39, was originated from the leaves of Chloranthus multistachys collected in Wulong of Chongqing municipality of China in November 2015. The strain was identified as Fusarium solani based on morphological characteristics, 5.8S gene and internal transcribed spacer sequence analysis. Two new compounds, 2β, 9α-dihydroxy-5α-methoxyergosta-7, 22-diene (1), 2β, 6β-dihydroxy-5α-methoxyergosta-7, 22-diene (2) have been isolated from the culture broth of the strain. Structures of the new compounds were elucidated by detailed analysis of their spectroscopic data aided by the comparison with reported data of related derivatives, and found to belong to the polyhydroxylated steroids with a hydroxyl at C-2 instead of C-3, a rare structure among the steroids. The extract of this strain and all isolated compounds were evaluated for their antagonistic activities.

Keywords: Chloranthus multistachys; Endophytic fungus; Polyhydroxysterols; Fusarium; Antagonistic activities.

Table S1. NMR spectroscopic data for compounds 1 and 2 (600 Hz, in acetone-d6, δ in ppm, J in Hz).

| No. | 1 | 2 |
|-----|---|---|
|     | δH (m, 1H), 1.43 (m, 1H) | δC (t) |
| 1   | 1.73 | 33.0 |
| 2   | 3.98 (m, 1H) | 66.7 (d) |
| 3   | 2.10 (m, 1H), 1.66 (m, 1H) | 40.4 (t) |
| 4   | 1.75 (m, 1H), 1.40 (m, 1H) | 31.3 (t) |
| 5   | 75.2 (s) | 79.1 (s) |
| 6   | 3.57 (bs, 1H) | 73.3 (t) |
| 7   | 5.21 (t, 1H) J = 8.2 | 119.0 (d) |
| 8   | 141.1 (s) | 119.6 (d) |
| 9   | 2.05 (m, 1H) | 42.8 (d) |
| 10  | 37.0 (s) | 74.5 (s) |
| 11  | 1.56 (m, 1H), 1.36 (m, 1H) | 21.7 (t) |
| 12  | 2.05 (m, 1H), 1.39 (m, 1H) | 39.5 (t) |
| 13  | 43.3 (t) | 44.9 (s) |
| 14  | 1.32 (m, 1H) | 56.0 (d) |
| 15  | 1.56 (m, 1H), 1.47 (m, 1H) | 22.8 (t) |
| 16  | 1.47 (m, 1H), 1.50 (m, 1H) | 27.9 (t) |
| 17  | 1.92 (t, 1H) | 54.6 (d) |
| 18  | 0.62 (s, 3H) | 11.3 (q) |
| 19  | 1.07 (s, 3H) | 17.8 (q) |
| 20  | 2.04 (s, 3H) | 40.3 (d) |
| 21  | 1.04 (d, 3H) J = 6.6 | 20.6 (q) |
|     | 5.24 (dd, 1H) $J = 15.0, 7.2$ | 135.8 (d) | 5.25 (dd, 1H) $J = 15.2, 7.2$ | 135.4 (d) |
|-----|------------------------------|----------|------------------------------|----------|
| 22  | 5.25 (dd, 1H) $J = 15.2, 7.4$ | 131.8 (d) | 5.28 (dd, 1H) $J = 15.2, 7.2$ | 132.1 (d) |
| 23  | 1.84 (m, 1H)                  | 43.1 (d) | 1.84 (m, 1H)                  | 42.8 (d)  |
| 24  | 1.47 (m, 1H)                  | 32.9 (d) | 1.43 (m, 1H)                  | 32.9 (d)  |
| 25  | 0.85 (d, 3H) $J = 6.6$        | 19.1 (q) | 0.86 (d, 3H) $J = 6.8$        | 19.0 (q)  |
| 26  | 0.86 (d, 3H) $J = 6.6$        | 19.4 (q) | 0.87 (d, 3H) $J = 6.8$        | 19.4 (q)  |
| 27  | 0.92 (d, 3H) $J = 6.7$        | 17.2 (q) | 0.96 (d, 3H) $J = 6.8$        | 19.7 (q)  |
| 28  | 3.56 (s)                      | 56.8 (q) | 3.56 (s)                      | 56.8 (q)  |
| 29-OCH₃ | 3.56 (s)                   | 56.8 (q) | 3.56 (s)                      | 56.8 (q)  |

Figure S1. Key $^1$H–$^1$H COSY and HMBC correlations of 1 and 2.

COSY (→) HMBC (H→C)

Figure S2. Key NOESY correlations of 1 and 2.

NOESY (H H)
Figure S3. The ESI-MS spectra of two new compounds.

Figure S4. The NMR (1H, 13C, COSY HMBC, HMQC) spectra of new compound 1.
Figure S5. The NMR (1H, 13C, DEPT, COSY HMBC, HMQC, NOESY) spectra of new compound 2.
