SUPPLEMENTARY MATERIAL

A new helvolic acid derivative from an endophytic Fusarium sp. of Ficus carica

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Abstract: A new helvolic acid derivative named helvolic acid methyl ester (1), together with two known helvolic acid compounds, helvolic acid (2) and hydrohelvolic acid (3), were isolated from the fermentation of endophytic fungus Fusarium sp. in Ficus carica leaves. Their structures were elucidated and identified by spectroscopic methods. Compounds 1-3 showed potent antifungal and antibacterial activities.

Keywords: Helvolic acid methyl ester; endophytic fungus; Fusarium sp.; Ficus carica; antifungal activity

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**Supplementary material 1**: The NMR of compound 1

![Helovic acid ester](image)

| Position | δ_C  | δ_H (J in Hz) | Position | δ_C  | δ_H (J in Hz) |
|----------|------|---------------|----------|------|---------------|
| 1        | 157.1| 7.30 (1H, d, J=10.1) | 18       | 18.0 | 0.93 (3H, s)  |
| 2        | 127.8| 5.85 (1H, d, J=9.7) | 19       | 27.5 | 1.45 (3H, s)  |
| 3        | 201.4|               |          | 20   | 130.3         |
| 4        | 40.4 | 2.78 (1H, d, J=12.0) | 21       | 172.9|               |
| 5        | 47.3 | 2.29 (1H, d, J=12.0) | 22       | 28.8 | 2.46 (2H, m)  |
| 6        | 73.8 | 5.23 (1H, s) | 23       | 28.3 | 2.10 (1H, m), 2.14 (1H, m) |
| 7        | 208.8|               | 24       | 122.8| 5.11 (1H, s)  |
| 8        | 52.7 |              | 25       | 132.9|               |
| 9        | 41.8 | 2.63 (1H, s) | 26       | 17.7 | 1.61 (3H, s)  |
| 10       | 38.2 |            | 27       | 25.7 | 1.69 (3H, s)  |
| 11       | 23.9 | 1.57 (1H, m), 1.87 (1H, m) | 28       | 13.1 | 1.28 (3H, d, J=6.5) |
| 12       | 25.9 | 1.81 (1H, m), 2.41 (1H, m) | 29       | 18.3 | 1.18 (3H, s)  |
| 13       | 49.5 | 2.57 (1H, s) | 30       | 170.1|               |
| 14       | 46.6 |              | 31       | 20.5 | 1.95 (3H, s)  |
| 15       | 40.7 | 1.89 (1H, m), 2.21 (1H, m) | 32       | 168.8|               |
| 16       | 73.5 | 5.89 (1H, d, J=8.2) | 33       | 20.7 | 2.11 (3H, s)  |
| 17       | 147.7|               | 34       | 53.6 | 3.71 (3H, s)  |
H-NMR (400 MHz, CDCl₃) of compound 1

$^{13}$C-NMR (100 MHz, CDCl₃) of compound 1

Supplementary material 2: The NMR of compounds 2 and 3
$^{1}$H-NMR (400 MHz, CDCl$_3$) of compound 2

$^{13}$C-NMR (100 MHz, CDCl$_3$) of compound 2
DEPT-135 spectrum of compound 2

$^1$H-NMR (400 MHz, CDCl$_3$) of compound 3
$^{13}$C-NMR (100 MHz, CDCl$_3$) of compound 3

Supplementary material 3: HRESI-MS of compound 1
Supplementary material 4: Strain identification of FL10

The microscopic features of endophytic fungus FL10

The nucleotide sequence of FL10 (without primer sequences)

CCGTTCCTCTTAATTTTTGTGACCGTTCTACTGATCGAGGTCAGCATTACATTTTGTTAATTTTTGTTGTCGACGGTTCTACTGATCGAGGTCAACATTCAGAAGTTGGGGTTTTACGGCATGGCCGCGCCGTTCCAGTTGCGAGGTGTTAGCTACTACGCAATGGAGGCTGCAGCGAGACCGCCAATGTATTTCGGGGGCGGCACCGCCCAGAAGGGCAGAGGCCGATCCCCAACCAACCAAAACCGGGGCTTGAGGGTTGAAATGACGCTCGAACAGGCA

System phylogenetic tree of strain FL10 based 18s rDNA-ITS sequence

Hypocreales sp. EU620165.1
Salicornia bigelovii JX110823.1

Fusarium sporotrichioides HM805048.1
Fusarium avenaceum HM805046.1
Gibberella pulicaris FJ481029.1
Fusarium concolor DQ767593.1
Fusarium sp. FJ196601.1
Gibberella sp. FJ196601.1
Fusarium concolor DQ767593.1
Fusarium oxysporum EU594568.1
Fusarium concolor DQ767593.1
Gibberella avenacea FJ602971.1
Fusarium solani JF776160.1
Fusarium tricinctum FJ459974.1

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