A New Friedelane Type Triterpene from *Euonymus hederaceus*

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**Abstract:** *Euonymus hederaceus* is distributed widely in the south of China; its stems and leaves have been used as folk medicines to treat many diseases such as renal deficiency and chronic diarrhea, traumatic injury, and abnormal menstruation. Chemical investigation of the leaves and stems of *Euonymus hederaceus* resulted in the isolation for the first time and characterization of a new friedelane type triterpene with a molecular mass of 472 and molecular formula of C_{30}H_{48}O_{4} by high resolution mass spectrometry. The 1H-NMR 13C-NMR and DEPT1350 spectra matched the characteristic data of the proposed triterpene skeleton. The compound was finally identified as 28-hydroxyfriedelan-3-one-29-oic acid on the basis of spectroscopic evidence, including two dimensional nuclear magnetic resonance as well as its IR spectrum.

**Keywords:** *Euonymus hederaceus*; new friedelane triterpene; 28-hydroxyfriedelan-3-one-29-oic acid; structure elucidation

**Introduction**

Celastraceae plants have been the subject of continued and growing interest due to the range of biological activities shown by many members of this family. Pharmaceutical studies and clinical practice have demonstrated that their sesquiterpenes and triterpenes possess notable antibacterial, anti-tumor, insect antifeedant and cytotoxic activities. More than 100 compounds have been isolated
and purified from three species, including *Celastrus hypoleucus* (Oliv.) Warb [1], *Celastrus hypoleucus* [2], and *Microtropis triflora* Merr [3] belonging to the *Celastraceae* family. The structures were determined by IR, UV, MS, and NMR, respectively, and 13 of them were new compounds. Friedelin ring triterpenes are a very important class, and so far, these compounds were reported to display a lot of biological activity. For example, 28,30-dihydroxyfriedelan-3-one showed good anti-tumor activity against P388 [4].

*Euonymus hederaceus* (Celastraceae) is a scramble shrub cultivated as an ornamental or hedge plant and widely distributed in Anhui, Jiangsu, Zhejiang, Jiangxi, Fujian, Hunan, Guangdong, and Guangxi provinces in China and Taiwan. Its decoctions are reputed in traditional medicine for their antibiotic and anti-tumor properties [5]. *Euonymus hederaceus* was investigated for the first time in our group, and five known friedelane triterpenes: 3-friedelone, 28-hydroxyfriedelan-3-one (canophyllal), 28-hydroxy-3-friedelanone (canophyllol), 30-hydroxy-3-friedelanone, 29-hydroxy-3-friedelanone and three olean-type triterpenoids including 3β-methoxyolean-11-oxo-18-ene, olean-12-ene-3,11-dione and 28-hydroxyolean-12-ene-3,11-dione were separated and their structures elucidated [6]. We report here the isolation and structure elucidation of a new triterpene by a combination of NMR techniques, including 1H-NMR, 13C-NMR, DEPT1350, 1H-1HCOSY, HMQC and HMBC.

**Results and Discussion**

**Compound 1:** acicular crystals, m.p. 294-296 °C, $[\alpha]_D^{20} = -8.016$ (MeOH). Its molecular formula was deduced to be $C_{30}H_{48}O_4$ (ESI-MS $m/z$: 473.3629 [M+H]$^+$, calcd. 473.3625). The IR spectrum showed absorptions at $\nu_{\text{max}}$ 3,426, 1,718 and 1,707 cm$^{-1}$, compatible with the presence of hydroxyl (OH) and two carbonyl functionalities, respectively. The 1H-NMR (C$_5$D$_5$N) spectra exhibited 46 protons, while the 13C-NMR and DEPT1350 spectra showed a total of 30 carbon signals: six primary carbons, twelve secondary carbons, four tertiary carbons and eight quaternary carbons, which were consistent with the characteristic of triterpene skeleton. Six methyl group signals at $\delta$ 0.66, 0.79, 1.04, 1.44, 1.45 (s, 3H each) and 0.95 (d, 3H) were observed in the 1H-NMR spectrum, in combination with six carbon signals at $\delta$ 14.6, 18.5, 15.7, 18.7, 33.0, and 7.2 (Table 1) in the 13C-NMR spectrum, which exhibited one methyl less than that of 28-hydroxy-friedelan-3-one. The HMBC correlations (Table 1) for the signals H-2 ($\delta$ 2.20), H-23 ($\delta$ 0.95), and H-4 ($\delta$ 2.12) with carbonyl carbon at $\delta$ 211.8 and for the signals H-28 ($\delta$ 3.89, 3.94) with C16, C22, and C18 were consistent with a 28-hydroxy-3-one type friedelane triterpene. Long-range correlations (HMBC) observed for the signals H-19 ($\delta$ 1.60), H-30 ($\delta$ 1.45), and H-21 ($\delta$ 1.77) with carbonyl carbon at $\delta$ 181.0 suggested the presence of another carbonyl group at C29, in agreement with the absorption at $\nu_{\text{max}}$ 1,707 cm$^{-1}$ revealed by the IR spectrum. Comparing with the carbon spectrum with those of 28-hydroxy-friedelan-3-one (Figure 1) and 2-hydroxy-3-oxo-friedelan-29-oic acid [7-8] as well as 2D data (Table 1), differences were also observed: specifically a downfield shift at C20 ($\delta$ 40.8, for compound 1; $\delta$ 28.1, for 28-hydroxy-friedelan-3-one) and C29 ($\delta$ 181.0, for compound 1; $\delta$ 32.9, for 28-hydroxy-friedelan-3-one). Therefore, compound 1 was identified as 28-hydroxyfriedelan-3-one-29-oic acid (Figure 2).
As reported, the D and E-rings adopted boat-boat conformation for 3-friedelin, because the repulsion between C29 and C27 is very strong [9]. However, when C29 was oxidized to a carboxylic acid or aldehyde, the D and E-ring would favor chair-chair conformation due to release of the repulsion [4,10]. So it was speculated that rings D/E in compound 1 adopted a chair-chair conformation (Figure 3).

**Figure 1.** The structure of 28-hydroxy-friedelan-3-one.

**Figure 2.** The structure of compound 1.

**Figure 3.** The chair-chair conformation of compound 1.
### Table 1. 1D NMR and 2D NMR data for compound 1.

| Position | δC (Compound 1 in pyridine-D$_5$) | 28-hydroxy-friedelan-3-one | 2-hydroxy-3-oxo-friedelan-29-oic acid | HMQC (δH) | HMBC |
|----------|----------------------------------|-----------------------------|---------------------------------------|-----------|------|
| C1       | 22.4 22.3 32.5                  | 1.50(m), 1.73(m)            | /                                     |           |      |
| C2       | 41.5 41.5 75.0                  | 2.20 (m), 2.38              | /                                     |           |      |
| C3       | 211.8 212.6 212.4               | /                           | H-2,4,24                              |           |      |
| C4       | 57.8 58.1 55.6                  | 2.12 (q, J=6.8)             | H-23,24                               |           |      |
| C5       | 41.9 42.1 43.0                  | /                           | H-4,10,23,24                          |           |      |
| C6       | 41.1 41.2 41.2                  | 1.10 (m), 1.54              | H-24                                  |           |      |
| C7       | 18.4 18.2 18.2                  | 1.23 (m)                    | H-8                                   |           |      |
| C8       | 50.4 52.4 50.8                  | 1.39 (m)                    | H-25,26                               |           |      |
| C9       | 37.6 37.4 37.4                  | /                           | H-8,10,25                             |           |      |
| C10      | 59.2 59.4 56.8                  | 1.40 (m)                    | H-24,25                               |           |      |
| C11      | 35.5 35.4 35.3                  | 1.30 (m)                    | H-25                                  |           |      |
| C12      | 29.7 30.1 29.5                  | 1.80 (m)                    | H-27                                  |           |      |
| C13      | 39.5 39.3 39.3                  | /                           | H-11,12,18                            |           |      |
| C14      | 39.4 38.1 39.3                  | /                           | H-7,12,18,26                          |           |      |
| C15      | 29.1 31.2 29.4                  | 1.41 (m)                    | H-26                                  |           |      |
| C16      | 31.4 29.1 36.1                  | 1.52 (m), 2.44              | H-28                                  |           |      |
| C17      | 35.6 35.1 30.1                  | /                           | H-18                                  |           |      |
| C18      | 40.3 39.4 44.2                  | 1.89 (m)                    | H-28                                  |           |      |
| C19      | 30.3 34.5 30.3                  | 1.60 (m), 2.62              | H-30                                  |           |      |
| C20      | 40.8 28.1 40.4                  | /                           | H-18,22,30                            |           |      |
| C21      | 31.1 31.4 29.7                  | 1.77 (m), 2.82              | /                                     |           |      |
| C22      | 32.1 33.3 36.7                  | 2.44 (m), 1.79              | H-28                                  |           |      |
| C23      | 7.2 6.8 6.6                     | 0.95 (d, J 6.8)             | H-4,5                                 |           |      |
| C24      | 14.6 14.7 14.7                  | 0.66 (s)                    | H-4                                   |           |      |
| C25      | 18.5 18.1 18.5                  | 0.79 (s)                    | H-8,10                                |           |      |
| C26      | 15.7 19.2 16.4                  | 1.04 (s)                    | /                                     |           |      |
| C27      | 18.7 19.1 18.1                  | 1.44 (s)                    | H-18                                  |           |      |
| C28      | 69.4 70.0 31.9                  | 3.89,3.93 (dd, J10.4)       | H-18                                  |           |      |
| C29      | 181.0 32.9 184.4                | /                           | H-19,21,30                            |           |      |
| C30      | 33.0 34.3 31.4                  | 1.45 (s)                    | H-21                                  |           |      |
Experimental

General

The IR spectrum was recorded in KBr pellets on a Nicolet NEXUS-470 FT-IR spectrometer. NMR spectra were recorded on a Bruker Avance DMX 500 NMR Instrument (Bruker Analytik, GmbH, Germany). The chemical shift values are given in ppm using pyridine-D$_5$ as solvent and TMS as the internal standard. Mass spectra were performed on an Apex III (7.0 Tesla) Fourier transformation ion cyclotron resonance mass spectrometer (FT-ICRMS) equipped with electrospray ionization source (ESI) (Bruker, Billerica, MA, USA).

Plant material

The whole plant of *Euonymus hederaceus* was collected in Jiulong Mountain, Suichang County, Zhejiang Province, P. R. China in October 2003, and identified by Dr. Haitong Wan (Zhejiang Chinese Medical University, Hangzhou, P. R. China.). A voucher specimen was deposited in the College of Agriculture and Biotechnology, Zhejiang University, Hangzhou, P. R. China.

Extraction and isolation

The shade-dried, powdered root barks, stem barks and leaves of *Euonymus hederaceus* (5.0 kg) were extracted three times with 95% EtOH (20 L) at room temperature for seven days. After removal of the solvent *in vacuo*, the extract was dissolved in H$_2$O (0.5 L) and extracted with EtOAc (2 L). The concentrated EtOAc extract (75 g) was subjected to column chromatography (CC) on silica gel, eluting with petroleum ether and increasing proportions of EtOAc. The eluate with 1:9 (petroleum ether/EtOAc) gave the new pure compound 1 (10 mg).

Conclusions

A new friedelane type triterpene was isolated from *Euonymus hederaceus*, its structure was determined to be 28-hydroxy-friedelan-3-one-29-oic acid by spectroscopic methods, including FT-ICRMS and NMR experiments, in combination to the comparison with known compounds. In addition, its stereo structure was proposed on the basis of the reference data.

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Sample Availability: Samples of compound 1 are available from the authors.

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