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Two new sesquiterpene lactones and other chemical constituents of *Artemisia roxbughiana*

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ABSTRACT

Two new sesquiterpene lactones, named roxbughianins A and B, were isolated together with thirteen other compounds from the leaves of *Artemisia roxbughiana* Bess. (Asteraceae). Their structures were determined by spectroscopic methods.

1. Subject and source

The genus *Artemisia* comprises over 500 species worldwide with fifteen species described in the Flora of Vietnam and is one of the largest of 1535 genera in the family Asteraceae (Vallès et al., 2003; Tariku et al., 2010). The genus *Artemisia* is usually presented by small herbs and shrubs with aromatic and bitter taste. Many species of the genus are of economic values because of their importance in the pharmaceutical and food industries. The genus has been divided into five sections, *Absinthium* (Tournefort) de Cand., *Artemisia* Tournefort (=section *Abrotanum* Besser), *Dracunculus* Besser, *Seriphidium* Besser (Hayat et al., 2009a), and *Tridentatae* (Rydb.) McArthur, which is endemic to North America (McArthur et al., 1981). The biology of *Artemisia* is diversified because of the high number of taxa and species are known to be difficult to identify (Kelsey, 1984). Chemical studies of *Artemisia* have been extensively published in the last 50 years (Tan et al., 1998) and compounds isolated can be utilized as an important aid in the systematic classification of *Artemisia*. For instance, in the Asteraceae sesquiterpene lactones have been used as chemical characteristics to understand the systematic relationships of the genera *Centaurea* (Bruno et al., 1998), *Scalesia* (Spring et al., 1999), and *Artemisia* (Kelsey and Shafizadeh, 1979). Our systematic study of *Artemisia roxbughiana* was carried out to evaluate whether the profile of sesquiterpene lactones in the species could be used as a chemical tool for differentiating species from other in the genus.

The leaves of *A. roxburghiana* Bess. (Vietnamese name: Ngải rừng) were collected in Ha Giang province, Viet Nam at an altitude of 600 m above sea level in November 2008. The plant was identified by Dr. Nguyen Quoc Binh, a botanist of the

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Institute of Biological Resources and Ecology, Vietnam Academy of Science and Technology, Hanoi, Viet Nam. Voucher specimen of the plant (voucher number: VMN-B0000302) was deposited at the same Institute.

2. Previous work

The chemistry of the genus *Artemisia* is diversified and characterized by the occurrence of essential oils (Güvenalp et al., 1998; Lopes-Lutz et al., 2008), sesquiterpene lactones (Kelsey and Shafizadeh, 1979; Lee et al., 2003), sesquiterpene alkaloids (Su et al., 2010), diterpenoids (Li et al., 1990), triterpenoids (Zheng, 1994; Sharma and Ali, 1996; Hu and Feng, 2000), polyacetylenic compounds (Marco et al., 1994; Lee et al., 2003), alkamides (Saadali et al., 2001), and phenolic compounds (Tan et al., 1998; Sheu and Tan, 1999; Lee et al., 2003). The common skeletal types of sesquiterpene lactones found in *Astemisia* are germacranolide, guaianolide, and eudesmanolide. The only report on the constituents of the essential oil obtained by hydrodistillation of the aerial parts of *A. roxbughiana* Bess. revealed the presence of mono- and sesquiterpenoids as its main constituents (Bicchi et al., 1998). Oxygenated sesquiterpenoids are very minor compounds in this oil of which several compounds are of eudesmane and eremophilane structures.

3. Present study

3.1. Extraction and isolation

The powder of dried *A. roxbughiana* leaves (6 kg) was extracted with MeOH at room temperature (three times, each time for three days). The combined MeOH extract was successively partitioned between water and *n*-hexane, CH_2Cl_2, and EtOAc to give the corresponding soluble fractions. Part of the *n*-hexane-soluble fraction (45 g) was subjected to silica gel open-column chromatography (CC) using *n*-hexane–acetone 19:1, 9:1, 6:1, and 3:1 to give eleven fractions. Fractions 2 (0.15 g) and 3 (61 mg) were purified by silica gel CC eluting with *n*-hexane–acetone 99:1, 49:1, and 30:1 to give compounds **1** (20 mg) and **2** (15 mg), respectively. Fraction 4 (1.1 g) was washed with *n*-hexane to give compound **3** (1 g). Compounds **4** (30 mg), **5** (0.1 g), and a mixture of compounds **6** and **7** (8 mg) were obtained from fraction 5 (0.8 g) and **9** (5 mg) from fraction 8 (2 g) by silica gel CC eluting with *n*-hexane–EtOAc 9:1, 6:1, and 3:1. Fraction 7 (0.4 g) was chromatographed on a silica gel column eluting with *n*-hexane–CH_2Cl_2 1:4, 1:9, and 1:15, which yielded, compound **8** (8 mg). Fraction 9 (1.9 g) was fractionated by silica gel CC eluting with *n*-hexane–acetone 6:1, 3:1, and 1:1 to afford a mixture of compounds **10** and **11** (65 mg). Part of the CH_2Cl_2-soluble fraction (42 g) was subjected to silica gel CC using *n*-hexane–acetone 49:1, 29:1, 19:1, 6:1, 3:1, and 1:1 to give eleven fractions. Silica gel CC of fraction 7 (3.8 g) eluting with *n*-hexane–acetone 19:1, 9:1, 6:1, 3:1, and 1:1 gave compounds **12** (3 mg), **13** (4 mg), and a mixture of compounds **8** and **14** (15 mg). The EtOAc-soluble fraction (3.0 g) was separated by silica gel CC eluting with *n*-hexane–EtOAc–HCOOH 20:19:1, 20:20:1, and 10:20:1 to give five fractions. Fraction 3 was purified by silica gel CC eluting with CH_2Cl_2–EtOAc 3:1 and 1:1 to give compound **15** (6 mg).

The structures of the isolated compounds (Fig. 1) were determined as friedelin (**1**) (Akihisa et al., 1992), friedelan-3β-ol (**2**) (Monkodkaew et al., 2009), tetracosanoic acid (**4**), β-sitosterol (**5**) (Goad and Akihisha, 1997), docosanoyl and tetracosanoyl p-coumarates (**6** and **7**) (Martínez et al., 1997), achillin (**8**) (Martínez et al., 1988), eicosanoic acid (**9**), 1-*O*-(tricosanoyl) and 1-*O*-(pentacosanoyl)glycerols (**10** and **11**) (Qi et al., 2004), palmitic acid (**12**), (23*Z*)-cycloart-3β,25-diol-23-ene (**13**) (Pei et al., 2007), 1β,10β-epoxyachilin (**14**) (Buděšínský and Šaman, 1995) by comparing their spectroscopic data (IR, MS, ^1H NMR and ^{13}C NMR including 2D NMR) with the reported literature values. Compounds **3** and **15** are new compounds.

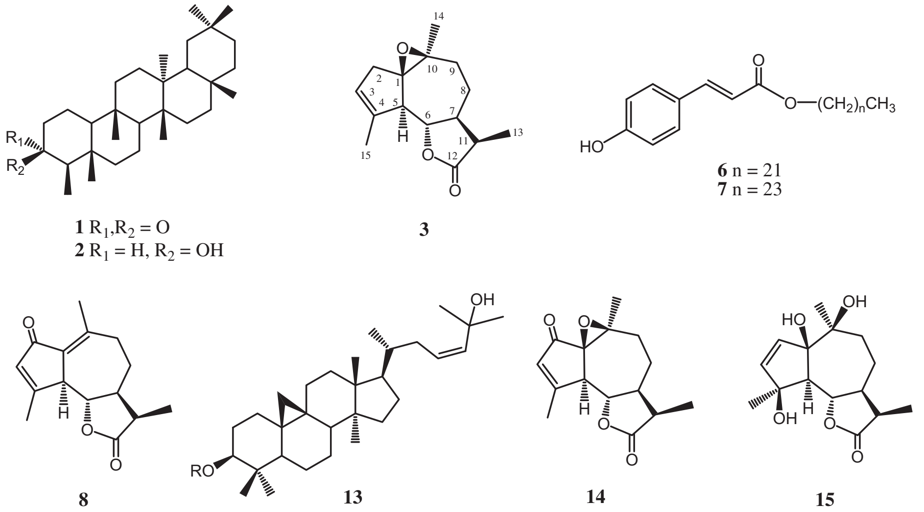

Fig. 1. Chemical structures of compounds **1–3**, **6–8**, and **13–15**.