A depside from *Frullania trichodes* Mitt.

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ABSTRACT

Chemical investigation of the dichloromethane extract of *Frullania trichodes* Mitt. led to the isolation of atranorin (1). The structure of 1 was elucidated by extensive 1D and 2D NMR spectroscopy and confirmed by comparison of its ¹H and ¹³C NMR data with those reported in the literature.

Keywords: *Frullania trichodes* Mitt., Frullaniaceae, liverwort, atranorin

INTRODUCTION

*Frullania trichodes* Mitt. (syn. *Frullania vethii* Sande Lac.) is a Bryophyte which belongs to a group of simple plants called liverworts [1]. There are no reported chemical constituents and biological activities of *F. trichotoma*. However, its synonym *F. vethii* was reported to afford guianolides and elemane type sesquiterpene lactones, as well as a large amount of 5-hydroxy-7,4′-dimethoxyflavone [2, 3]. Liverworts of the genus *Frullania* have also been studied chemically. Two new cadinane-type sesquiterpenes, frullanic acid and frullanic acid methyl ester, together with four known benzylns, brittonin B, 3,3′-dimethoxy-4,5-methylenedioxybibenyl, 3,4,5,3′,4′-pentamethoxybibenyl and 3-(4′-methoxybenzyl)-5,6-dimethoxyphthalide, were isolated from the Chinese liverwort *F. serrata* [4]. Another liverwort, *F. brasiliensis* afforded two eremophilanes, 5-epi-dilatanolide A [5] and 5-epi-dilatanolide B [5] and two eudesmanes, (+)-frullanolide [5] and nepalensolide A [6]. The Indian *F. inflata* was reported to contain the sesquiterpenes, (11S)-11,13-dihydrolipiferolide, α-cubebene, β-cubebene, α-copaene, β-caryophyllene, germacrene-A, α-selinene, β-selinene and β-guaiene [7]. Another study reported the isolation of tamariscene, valerena-4,7(11)-diene, pacifigorgia-1,10-diene, pacifigorgia-1(6),10-diene, pacifigorgia-1(9),10-diene, pacifigorgia-2,10-diene, and pacifigorgia-2(10),11-diene from *F. tamarisci*, *F. fragilifolia* and *Valeriana officinalis* [8]. In another study, *F. muscicola* yielded stigmasteryl arachidate, sitosterol, apigenin-7,4′-dimethy ether, scutellarin-6,4′-dimethyl ether, 6-hydroxyxuteolin-6,3′-dimethyl ether, scutellarin-6-methyl ether, and daucosterol[9]. Furthermore, ent-labdane type diterpenoid, muscinolone, two benzylns and four flavonoids were isolated from the liverwort *F. muscicola* Steph. [10]. Muscinolone showed cytotoxic effects to some human tumor cells [10]. A new pacifigorgiane sesquiterpenoid alcohol, tamariscol was isolated from another liverwort *F. tamarisci* [11]. The diterpenoids, 1,2-dehydro-3,7-dioxo-manoxy oxide, 1,2-dehydro-7β-hydroxy-3-oxo-manoxy oxide, 3,7-dioxo-manoxy oxide, 3-β-hydroxy-7-oxo-manoxy oxide and highly methoxylated benzylns were isolated from the liverwort *F. inouei* [12].
We report herein the isolation of atranorin (I) from *F. trichodes* Mitt. To the best of our knowledge this is the first report on the isolation of I from *F. trichodes*.

**MATERIALS AND METHODS**

**General Experimental Procedure**

NMR spectra were recorded on a Varian VNMR spectrometer in CDCl₃ at 600 MHz for ¹H NMR and 150 MHz for ¹³C NMR spectra. Column chromatography was performed with silica gel 60 (70-230 mesh). Thin layer chromatography was performed with plastic backed plates coated with silica gel F₂₅₄ and the plates were visualized by spraying with vanillin/H₂SO₄ solution followed by warming.

**General Isolation Procedure**

A glass column 12 inches in height and 0.5 inch internal diameter was used for the chromatography of the crude extract. Two milliliter fractions were collected. All fractions were monitored by thin layer chromatography. Fractions with spots of the same Rₚ values were combined and rechromatographed in appropriate solvent systems until TLC pure isolates were obtained. Final purifications were conducted using Pasteur pipettes as columns. One milliliter fractions were collected.

**Plant material**

The samples were found growing on palm tree trunks with direct sunlight exposure in the town of Banaue, Ifugao Province, Cordillera Administrative Region, Philippines. The sample location is N 16°55.60', E 121°03.199' at about 1350 meters above sea level. The area endures mountain climate with temperatures at the time of collection about 10-15°C, and frequent fog. Samples were collected on January 9, 2016. The liverwort was identified as *Frullania trichodes* Mitt. by Virgilio C. Linis of the Biology Department, De La Salle University, Manila, Philippines.

**Isolation**

The air-dried *F. trichodes* (0.80 g) was ground in an osterizer, soaked in CH₂Cl₂ for three days, and then filtered. The filtrate was concentrated under vacuum to afford a crude extract (0.59 g) which was chromatographed using increasing proportions of acetone in CH₂Cl₂ (10% increment) as eluents. The 70% to 90% acetone in CH₂Cl₂ fractions were combined and rechromatographed (2 ×) using CH₂CN:Et₂O:CH₂Cl₂ (2.5:2.5:5, v/v) to yield I (5 mg) after washing with petroleum ether, followed by Et₂O.

**Atranorin (I):** ¹H NMR(600 MHz, CDCl₃): δ 66.38 (s, H-5), 10.34 (s, H-8), 2.67 (s, CH₁-9), 6.50 (s, H-6'), 2.07 (s, CH₁-8'), 2.53 (s, CH₁-9), 3.97 (s, OCH₂) 12.48 (s, 2-OH), 12.53 (s, 4-OH), 11.93 (s, 3'-OH); ¹³C NMR(150 MHz, CDCl₃): 131.02 (C-1), 169.08 (C-2), 108.53 (C-3), 167.47 (C-4), 112.84 (C-5), 152.43 (C-6), 169.69 (C-7), 193.83 (C-8), 25.57 (C-9), 151.96 (C-1'), 116.77 (C-2'), 162.87 (C-3'), 110.24 (C-4'), 139.86 (C-5'), 115.79 (C-6'), 172.17 (C-7'), 9.41 (C-8'), 24.02 (C-9'), 52.33 (OCH₃).

**RESULTS AND DISCUSSION**

Silica gel chromatography of the dichloromethane extract of *Frullania trichodes* led to the isolation of atranorin (I). The structure of I was elucidated by extensive 1D and 2D NMR spectroscopy and confirmed by comparison of its ¹H and ¹³C NMR data with those reported in the literature [13].
Atranorinis found in a variety of lichen species. It exhibited anti-proliferative action against malignant cell lines [14], antinociceptive effects [15, 16] and antibiotic action against M. aurum [17]. It was found to inhibit leukotriene B4 synthesis in leukocytes, which might affect inflammatory processes [18] and modulates the wound healing process [19].

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REFERENCES