

DAMMARANE-TYPE TRITERPENOIDS FROM THE STEMBARK OF
CHISOCHETON PENTANDRUS (MELIACEAE)

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ABSTRAK

Dua senyawa triterpenoid tipe damaran, cabraleadiol (**1**) dan cabraleahidrosilakton (**2**), telah diisolasi dari ekstrak *n*-heksana kulit batang *Chisocheton pentandrus* (Meliaceae). Struktur kimia senyawa **1** dan **2** diidentifikasi berdasarkan data-data spektroskopi terutama, NMR dan massa serta perbandingan data spektra dari laporan sebelumnya. Senyawa **1** dan **2** pertama kali dilaporkan pada tumbuhan *Chisocheton pentandrus*.

Kata kunci: Cabraleadiol, Cabraleahidrosilakton, *Chisocheton pentandrus*, Meliaceae.

ABSTRACT

Two dammarane-type triterpenoids, cabraleadiol (**1**) and cabraleahydroxylactone (**2**), have been isolated from *n*-hexane extract of the stem bark of *Chisocheton pentandrus* (Meliaceae). The structure of compounds **1** and **2** were determined by spectroscopic data mainly NMR and mass as well as by comparing with previously reported spectral data. Compounds **1** and **2** were reported for the first time from *Chisocheton pentandrus*.

Keywords: Cabraleadiol, Cabraleahydroxylactone, *Chisocheton pentandrus*, Meliaceae.

INTRODUCTION

Triterpenoids are the most important group of terpenoids because they exhibit a great diversity of biological activities and they are the major constituents of tropical higher plants. Recently, research of new biologically active compounds from plants used in traditional medicine has led to the isolation of numerous triterpenoids with important biological activities (Fu *et al.*, 2015; Nguyen *et al.*, 2015). Higher plants are major source of triterpenoids with several biological activities and numerous reports have shown that family of Meliaceae, Rhamnaceae, Cucurbitaceae, Ganodermataceae and Apocynaceae produce a wide variety of tetracyclic triterpenoids, whereas, family of Ranunculaceae, Burseraceae, Cappariaceae, Celastraceae and Lamiaceae families are recognized to contain

active pentacyclic triterpenoids (Harneti *et al.*, 2012; Farabi *et al.* 2017; Tian *et al.*, 2005; Zhang *et al.*, 2005).

Chisocheton genera is belong to the Meliaceae family, which consist of more than 50 plant species distributed mainly in tropical countries (Yang *et al.*, 2009; Heyne, 1982). Previous phytochemical studies on *Chisocheton* species have yielded a number of interesting compounds, including limonoids (Supratman *et al.*, 2019; Supriatno *et al.*, 2018; Katja *et al.*, 2016) and triterpenoids (Inada *et al.*, 1993; Katja *et al.*, 2017).

In our continuous search for novel constituents from *Chisocheton pentandrus*, we isolated new limonoids, pentandricin A from the stem bark of *C. pentandrus* (Suprianto *et al.*, 2018). In the further investigation for novel compounds from non polar fraction of *C. pentandrus*, we found two dammarane-type

triterpenoids from the *n*-hexane extract. In this paper, we report the isolation and structural determination of two dammarane-type triterpenoids, cabraleadiol (**1**) and cabraleahydroxylactone (**2**).

MATERIAL AND METHODS

Plant material

The stem bark of *C. pentandrus* were collected in Bogor Botanical Garden, Bogor, West Java Province, Indonesia in June 2016. The plant was identified by Mr. Ismail, the staff of the Herbarium and a voucher specimen (No. Bo-104) was deposited at the Herbarium.

Extraction and isolation

The dried ground stem bark (1.8 kg) of *C. pentandrus* was extracted with methanol (3 x 4 L) at room temperature for 6 days. After removal of the solvent under vacuum, the viscous concentrate of methanol extract (340 g) was first suspended in water and then partitioned with *n*-hexane, ethyl acetate and *n*-butanol, successively. Evaporation resulted in the crude extracts of *n*-hexane (10.9 g), ethyl acetate (25.2 g) and *n*-butanol (228.6 g), respectively. The *n*-hexane extract (10.9 g) was fractionated by column chromatography on silica gel using a gradient *n*-hexane-ethyl acetate-methanol to give eight fractions (I–VIII). Fraction II (5.4 g) was subjected to column chromatography over silica gel using a gradient mixture of *n*-hexane-dichloromethane-ethyl acetate (5% stepwise) as eluting solvents to afford thirteen subfractions (II₁–II₁₃). Subfraction II₉ (912.2 mg) was column chromatographed on silica gel, eluted with *n*-hexane:dichloromethane:ethyl acetate (5:4:1), to give six subfractions (II_{9.1}–II_{9.6}). subfraction II_{9.3} (62.2 mg) was further subjected to column chromatography over silica gel using a gradient mixture of *n*-hexane-dichloromethane-ethyl acetate (7:2:1) to give **1** (12.2 mg) and **2** (8.3 mg).

General Experimental Procedure

Melting points were determined on an electrothermal melting point apparatus. The IR spectra were recorded on a Perkin-Elmer spectrum-100 FT-IR in KBr. Mass spectra were obtained with a Synapt G2 mass spectrometer instrument. NMR data were recorded on a JEOL-ECZ-600 spectrometer at

600 MHz for ¹H and 150 MHz for ¹³C. Chemical shifts are given on a δ (ppm) scale with tetramethylsilane (TMS) as an internal standard. Column chromatography was conducted on silica gel 60 (Merck). TLC plates were precoated with silica gel GF₂₅₄ (Merck, 0.25 mm) and detection was achieved by spraying with 10% H₂SO₄ in ethanol, followed by heating.

RESULTS AND DISCUSSION

Cabraleadiol (1)

Physical properties: colourless needle crystal, m.p. 170-172°C. IR λ_{max} (KBr) cm⁻¹: 3457, 2948, 2861, 1461, 1380. HR-TOFI-MS *m/z*: 461.3879 [M+H]⁺. ¹H-NMR (600 MHz, CDCl₃) and ¹³C-NMR (125 MHz, CDCl₃) see Table 1.

Cabraleahydroxylactone (2)

Physical properties: white amorphous powder, m.p. 240-242 °C, IR λ_{max} (KBr) cm⁻¹: 3550, 2900, 2810, 1760, 1457, 1386, 1249, 1196. HR-TOFI-MS *m/z*: 416 [M+H]⁺; ¹H-NMR (600 MHz, CDCl₃) and ¹³C-NMR (125 MHz, CDCl₃) see Table 1.

Compound **1** was isolated as a colourless needle crystal. The molecular formula of **1** was determined to be C₃₀H₅₂O₃ from its molecular ion peak [M+H]⁺ at *m/z* 460.3916 (calcd. for C₃₀H₅₂O₃, 460.3920) in the LC-TOFMS. The IR spectrum showed absorption peaks due to hydroxyl (3457 cm⁻¹), C-C aliphatic (2948 and 2861 cm⁻¹) and ether functional groups (1380 cm⁻¹). The ¹H NMR spectrum (Table 1) displayed eight tertiary methyl singlets [δ_H 0.87, 0.82, 0.92, 1.09, 1.17, 1.13, 0.95 and 0.84 (each 3H, s)], two oxymethine protons [δ_H 3.38 (1H, t, *J*=3.0 Hz, H-3 and 3.62 (1H, dd, *J*=4.8, 10.2 Hz, H-24)], and some aliphatic protons in the up field region. The ¹³CNMR and DEPT-135 spectra (Table 1), exhibited the presence of eight methyl signals [δ_C 15.6 (Me-30), 22.2 (Me-29), 28.4 (Me-28), 24.1 (Me-27), 27.9 (Me-26), 27.3 (Me-21), 16.6 (Me-18) and 16.2 (Me-19)], ten methylenes [δ_C 33.7 (C-1), 25.4 (C-2), 18.3 (C-6), 34.8 (C-7), 21.7 (C-11), 27.1 (C-12), 31.5 (C-15), 25.9 (C-16), 35.3 (C-22), 26.4 (C-23)], six methines including two oxygenated methines [δ_C 76.4 (C-3), 49.6 (C-5), 50.7 (C-9), 42.8 (C-13), 49.8 (C-17), and

86.3 (C-24)] and eight quaternary carbon signals [δ_C 37.3 (C-4), 40.7 (C-8), 37.7 (C-10) and 50.2 (C-14)]. The NMR data suggested that **1** had a triterpenoid tetracyclic skeleton similar to the dammarane-type triterpenoid (Harneti et al., 2014). The structure of the tetracyclic system in **1** was determined by analysis of COSY and HMBC spectra (Figure 2). Key HMBC spectra were the 2J correlations from the eight methyl groups (Me-18, Me-19, Me-21, Me-14, Me-26, Me-27, Me-29 and Me-30) to their attached carbons enabled the assignment of the eight singlet methyls. A secondary alcohol was assigned at C-3 by the HMBC correlations from H-1 (δ_H 1.42), H-2 (δ_H 1.55), and H-5 (δ_H 1.24) to C-3 (δ_C 76.4), whereas a tertiary alcohol was located at C-25 by the HMBC correlation from Me-26 (δ_H 1.17), Me-27 (δ_H 1.09), H-24 (δ_H 3.62) to C-25 (δ_C 70.3).

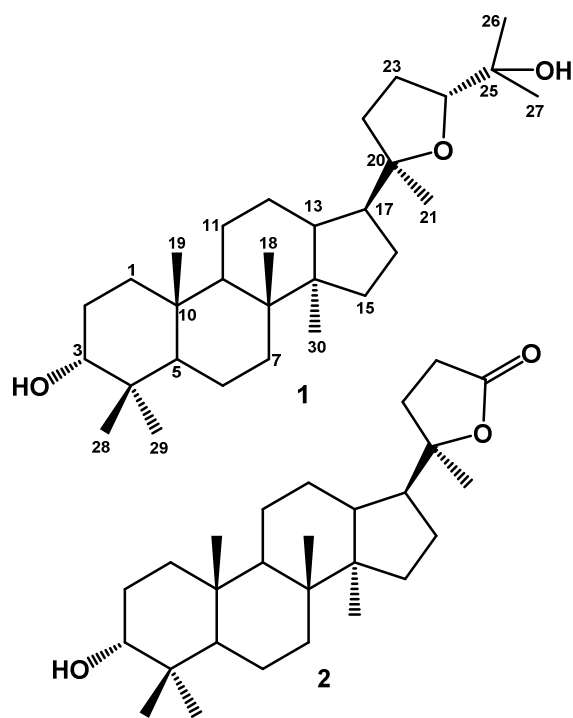


Figure 1. Chemical Structure of **1** and **2**

The relative stereochemistry of **1** was identified based on coupling constants in the 1H -NMR and biogenetic point of view occurrence of dammarane-type triterpenoid in *Chisoche-ton* genus (Supratman et al., 2019).

In comparison of **1** with literature data of a cabraleadiol (Phongmaykin et al., 2008), showed good agreement, therefore compound **1** was identified as a cabraleadiol, which shown in this plant for the first time.

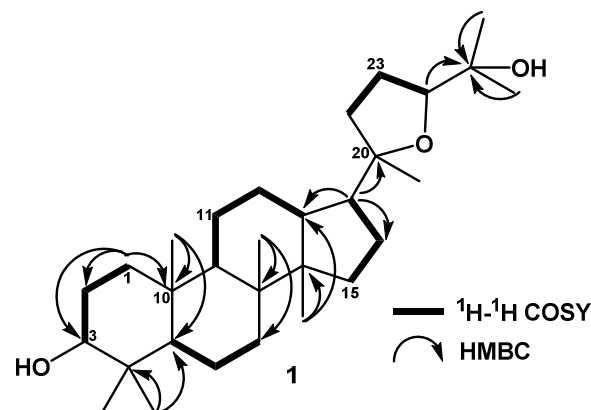


Figure 2. 1H - 1H COSY and HMBC for **1**

Compound **2** was obtained as a white amorphous powder. The IR spectra showed absorption peaks at 3550 cm^{-1} (OH), 2960 and 2865 cm^{-1} (aliphatic), 1382 and 1238 cm^{-1} (*gem*-dimethyl groups), and 1040 cm^{-1} (C-O). The NMR spectra of **2** was very similar with **1**. The main differences was the absence of an isopropyl alcohol group at [$(\delta_H$ 1.17 (3H, s) and 1.09 (3H, s), δ_C 24.1 (Me-27), 27.9 (Me-26) and 70.3 (H-25)] and the presence of carbonyl lactone signal at δ_C 176.9, suggested that compound **2** was a lactone derivative of **1**. In the HMBC spectrum, methylene signal at δ_H 2.52 was correlated to carbonyl lactone at δ_C 176.9, indicated that a carbonyl lactone was located at C-24 to make a lactone ring as a side chain ring.

The relative stereochemistry of **2** was identified based on coupling constants in the 1H -NMR and biogenetic point of view occurrence of dammarane-type triterpenoid in *Chisoche-ton* genus (Supratman et al., 2019). In comparison of **1** with literature data of a cabraleahydroxylactone (Phongmaykin et al., 2008), showed good agreement, therefore compound **1** was identified as a cabraleahydroxylactone, which shown in this plant for the first time.

Tabel 1. NMR data for compounds **1** and **2** (CDCl₃ 600 MHz for ¹H and 150 MHz for ¹³C)

Position of C	Compound 1		Compound 2	
	¹³ C NMR δ _c (mult.)	¹ H NMR δ _H (Integral, mult., J=Hz)	¹³ C NMR δ _c (mult.)	¹ H NMR δ _H (Integral, mult., J=Hz)
1	33.7 (t)	1.42 (1H, m) 1.34 (1H, m)	35.2 (t)	1.17 (1H, m) 1.50 (1H, m)
2	25.4 (t)	1.55 (1H, m) 1.28 (1H, m)	33.7 (t)	1.40 (1H, dd; 2,4; 9,6) 1.46 (1H, m)
3	76.4 (d)	3.38 (1H, t, 3)	76.3 (d)	3.37 (1H, s)
4	37.3 (s)	-	37.3 (s)	-
5	49.6 (d)	1.24 (1H, m)	49.4 (d)	1.95 (1H, m)
6	18.3 (t)	1.39 (1H, m) 1.30 (1H, m)	18.3 (t)	1.37 (1H, m) 1.56 (1H, m)
7	34.8 (t)	1.63 (1H, m) 1.76 (1H, m)	26.9 (t)	1.71 (1H, m) 1.82 (1H, m)
8	40.7 (s)	-	40.6 (s)	-
9	50.7 (d)	1.44 (1H, m)	50.4 (d)	1.41 (1H, dd, 2,4, 13,2)
10	37.7 (s)	-	37.7 (s)	-
11	21.7 (t)	1.53 (1H, m) 1.46 (1H, m)	25.4 (t)	1.20 (1H, m) 1.68 (1H, m)
12	27.1 (t)	1.75 (1H, m) 1.60 (1H, m)	21.3 (t)	1.49 (1H, m) 1.24 (1H, m)
13	42.8 (d)	1.62 (1H, m)	43.2 (d)	1.53 (1H, m)
14	50.2 (s)	-	50.3 (s)	-
15	31.5 (t)	1.04 (1H, m) 1.80 (1H, m)	31.2 (t)	1.90 (1H, m) 1.10 (1H, m)
16	25.9 (t)	1.51 (1H, m) 1.72 (1H, m)	25.1 (t)	1.52 (1H, m) 1.80 (1H, m)
17	49.8 (d)	1.83 (1H, m)	49.5 (d)	1.23 (1H, m)
18	16.2 (q)	0.95 (3H, s)	15.6 (q)	0.92 (3H, s)
19	16.6 (q)	0.84 (3H, s)	16.1 (q)	0.82 (3H, s)
20	86.7 (s)	-	90.3 (s)	-
21	27.3 (q)	1.13 (3H, s)	25.4 (q)	1.33 (3H, s)
22	35.3 (t)	1.22 (1H, m) 1.40 (1H, m)	31.3 (t)	1.47 (1H, m) 2.01 (1H, m)
23	26.4 (t)	1.85 (1H, m) 1.90 (1H, m)	29.3 (t)	2.52 (1H, d, 10) 2.62 (1H, d, 9,9)
24	86.3 (d)	3.62 (1H, dd, 4,8, 10,2)	176.9 (s)	-
25	70.3 (s)	-	-	-
26	27.9 (q)	1.17 (3H, s)	-	-
27	24.1 (q)	1.09 (3H, s)	-	-
28	28.4 (q)	0.92 (3H, s)	28.4 (q)	0.91 (3H, s)
29	22.2 (q)	0.82 (3H, s)	22.2 (q)	0.81 (3H, s)
30	15.6 (q)	0.87 (3H, s)	16.4 (q)	0.87 (3H, s)

CONCLUSIONS

Two dammarane-type triterpenoids have been isolated from the stem bark of *Chisocheton pentandrus* belong to Meliaceae family and identified by spectroscopic data as cabraleadiol (**1**) and cabraleahydroxylactone (**2**). The investigation of these dammarane-type triterpenoids were shown in this species

for the first time and strengthen the occurrence of this compounds in this genera.

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