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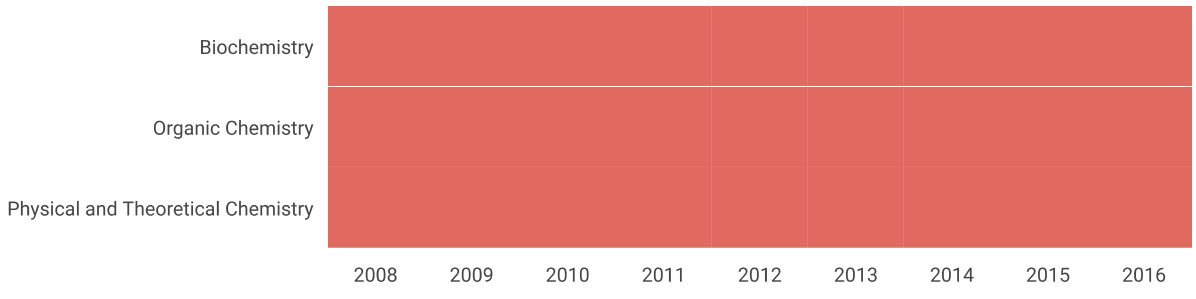

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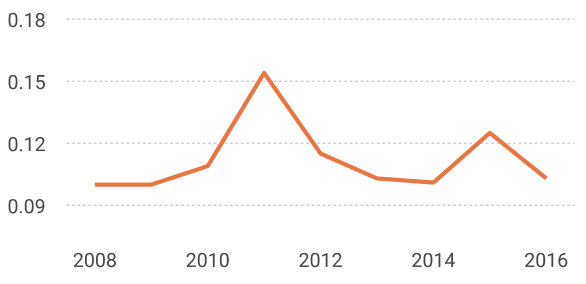

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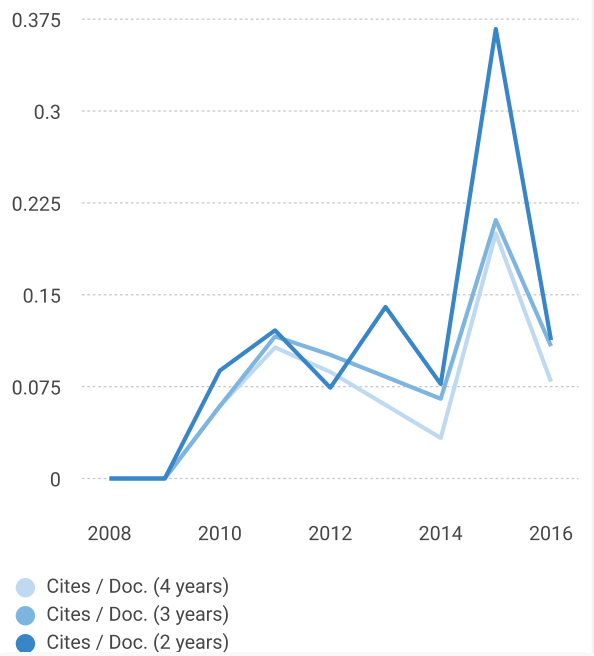

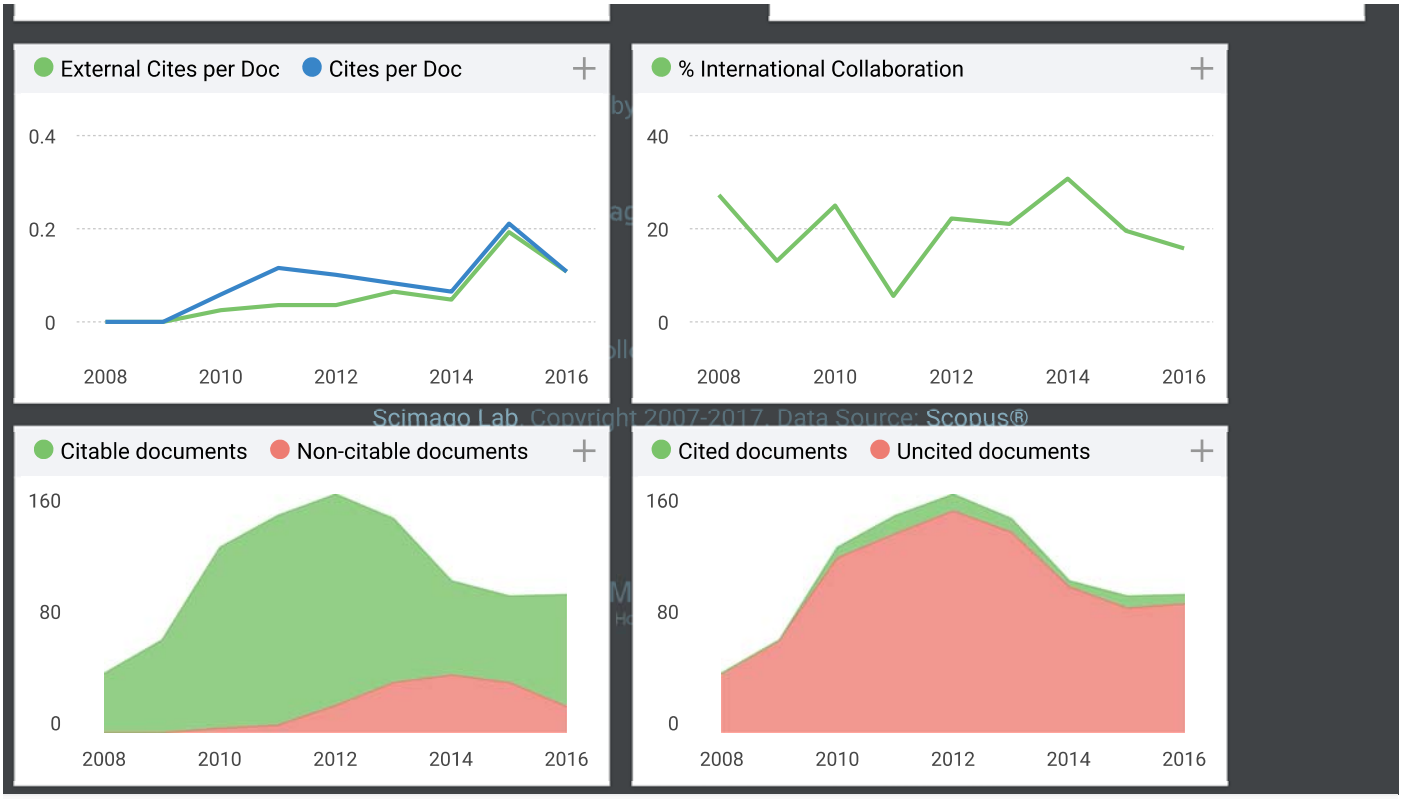

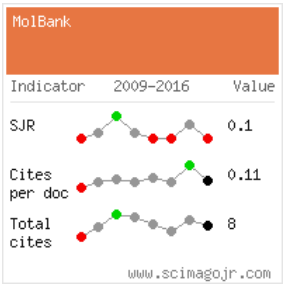

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5,9,11-Trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one from the Stem Bark of *Calophyllum tetrapterum* Miq.

by

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Abstract

A new pyranoxanthone namely 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one (**1**) was isolated from the stem bark of *Calophyllum tetrapterum* Miq. The structure of compound **1** was determined by means of spectroscopic methods including UV, IR, HRESIMS, 1D and 2D NMR. View Full-Text (/1422-8599/2017/1/M936/htm)

Keywords: *Calophyllum tetrapterum* Miq. (/search?q=Calophyllum%20tetrapterum%20Miq.); pyranoxanthone (/search?q=pyranoxanthone); natural product (/search?q=natural%20product)

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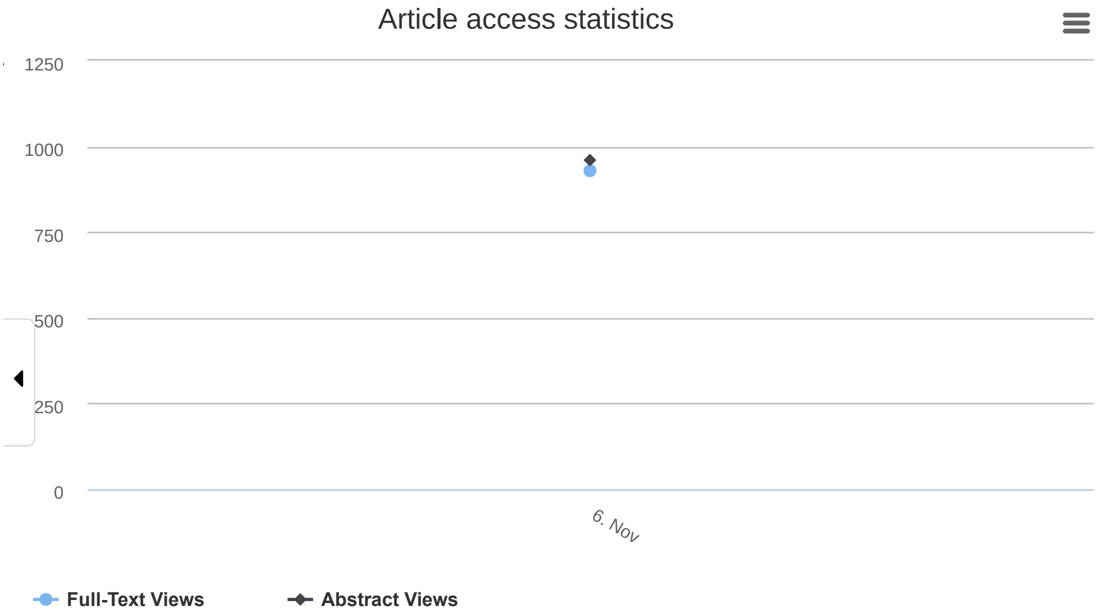

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Short Note

5,9,11-Trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one from the Stem Bark of *Calophyllum tetrapterum* Miq.

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Abstract: A new pyranoxanthone namely 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl) pyrano[2,3-a]xanthen-12(2*H*)-one (**1**) was isolated from the stem bark of *Calophyllum tetrapterum* Miq. The structure of compound **1** was determined by means of spectroscopic methods including UV, IR, HRESIMS, 1D and 2D NMR.

Keywords: *Calophyllum tetrapterum* Miq.; pyranoxanthone; natural product

1. Introduction

The genus *Calophyllum* (Clusiaceae) comprises over 200 species of trees and shrubs native to tropical Asia, East Africa and Australia. This genus is well known to be a rich source of bioactive xanthones [1–4], coumarins [5–7], chromanone acids [8–11], and flavonoids [12]. Some these were reported to exhibit of biological activities including anti-HIV, anticancer, antimalarial and antimicrobial [13,14].

In this paper, we report the chemical constituents of the stem bark of *Calophyllum tetrapterum* Miq. with the isolation of a new pyranoxanthone, 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl) pyrano[2,3-a]xanthen-12(2*H*)-one (Figure 1). The anti-HIV activity of isolated compound from this plant is also reported.

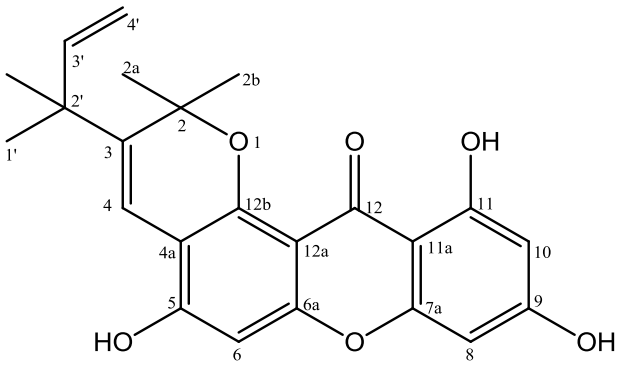

Figure 1. Structures of 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one (**1**).

2. Result and Discussion

5,9,11-Trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one (**1**) was isolated as yellow solid, m.p. 156–158 °C. The HRESIMS displayed a negative molecular ion peak at *m*/*z* 393.1352 indicating a molecular formula of $C_{23}H_{21}O_6$ and 13 degrees of unsaturation (See supporting information, Figure S8). The UV spectrum exhibited four absorption bands characteristic of a xanthone chromophore at λ_{max} 249, 260, 320 and 335 nm [3]. The IR spectrum showed absorptions bands at ν_{max} 3436, 1652, and 1510 cm^{-1} indicating the presence of a hydroxyl, conjugated carbonyl and aromatic groups, respectively. The ^{1}H-NMR (Table 1) spectrum showed the presence of the proton signals of a pair of doublets (*J* = 2.2 Hz) in the aromatic region at δ_H 6.20 and 6.33 (each 1H) and a singlet at δ_H 6.80, suggest that compound **1** is a typical for a xanthone with five substituents. The presence of a chelated hydroxyl group at δ_H 13.48 assignable to the signal of 11-OH. The ^{1}H-NMR revealed the presence of a 2,2-dimethylpyrano group [δ_H 1.50 (6H, *s*, H2a/H-2b), 8.17 (1H, *s*, H-4) and 1,1-dimethylallyl group [δ_H 1.41 (6H, *s*, H-1′, 2′-CH_3), 5.08 (1H, *dd*, *J* = 1.1; 10.6 Hz, H-4′a), 5.16 (1H, dd, *J* = 1.1; 17.5 Hz, H-4′b), 6.02 (1H, dd, *J* = 10.6; 17.5 Hz, H-3′)]. See supporting information in Figures S1 and S2. The ^{13}C-NMR spectrum (APT experiment, Table 1) of **1** showed 21 carbon signals representing for 23 carbon atoms were observed. See Figures S3 and S4, supporting material.

Table 1. Data NMR of 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl)pyrano[2,3-a]xanthen-12(2*H*)-one in acetone-d_6.

No. C	δ_H (Mult, *J* Hz)	δ_C	HMBC
2	-	80.4	-
2a	1.50 (*s*, 3H)	27.3	C-2; C-2b
2b	1.50 (*s*, 3H)	27.3	C-2; C-2a
3	-	137.8	-
4	8.17 (*s*, 1H)	118.7	C-2; C-3; C-4a; C-2′
4a	-	108.3	-
5	-	155.6	-
6	6.80 (*s*, 1H)	103.0	C-4a; C-5; C-6a
6a	-	153.6	-
7a	-	158.1	-
8	6.33 (*d*, 2.2, 1H)	93.9	C-7a; C-9; C-10; C-11a
9	-	165.4	-
10	6.20 (*d*, 2.2, 1H)	98.7	C-8, C-11, C-11a
11	-	164.7	-
11a	-	103.9	-
12	-	183.1	-
12a	-	122.8	-
12b	-	149.9	-
1′	1.41 (*s*)	28.6	C-2′; C-3′, 2′-CH_3
2′	-	42.7	-
2′-CH_3	1.41 (*s*)	28.6	C-1′; C-2′, C-3′
3′	6.02 (*dd*, 10.6; 17.6, 1H)	147.9	C-1′; C-2′, 2′-CH_3
4′	5.16 (*dd*, 1.1; 17.5, 1H) 5.08 (*dd*, 1.1; 10.6, 1H)	112.3	C-2′, C-3′
11-OH	13.48 (*s*, 1H)	-	C-10; C-11; C-11a

The presence of long-range correlations in the HMBC spectrum of **1** between the proton signal of a chelated hydroxyl group (δ_H 13.48, 11-OH) was correlated with two quaternary carbons (δ_C 164.7; 103.9) and a methine (δ_C 98.7) carbon signals, showing that C-10 is unsubstituted. One of them, proton signal of aromatic region at δ_H 6.33 (*J* = 2.2 Hz) that showed long-range correlations with two oxyaril carbon signals (δ_C 165.4 (C-9); 158.1 (C-7a)), a quarternary (δ_C 103.9) and a methine carbon signals (δ_C 98.7), showing that δ_H 6.33 at C-8. Furthermore, proton signal of isolated aromatic (δ_H 6.80, *s*, H-6) has correlation with two oxyaril carbons [δ_C, 155.6 (C-5), 153.6 (C-6a)] and a quaternary

carbon signals (δ_C 108.3, C-4a), which showed that 2,2-dimethylpyrano group were fused at C-4a and C-12b. The methine signal of vinyl group at δ_H 8.17 (H-4) on the 2,2-dimethylpyrano group showed long-range correlations with four quarternary carbons [δ_C 137.8 (C-3), 108.3 (C-4a), 80.4 (C-2), 42.7 (C-2′)]. This showed that C-3 bonded with 1,1-dimethylallyl group. This HMBC correlation is similar to the previous reported of xanthone in *Calphyllum pseudomole* [3]. Long-range correlations in HMBC consistent with the structure **1** are shown in Figure 2 and supporting information in Figure S5–S7. Based on the above spectral evidence, 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl) pyrano[2,3-a]xanthen-12(2*H*)-one was established to have structure **1** which is a novel compound and. had not been reported yet.

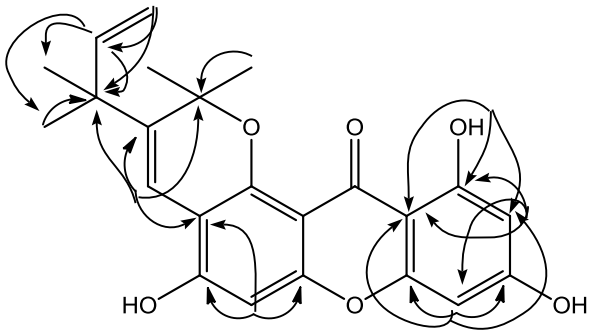

Figure 2. Selected HMBC correlations for **1**.

On inhibition of human immunodeficiency virus type-1 reverse transcriptase (HIV-1 RT) against human lymphocytes in vitro, compound **1** exhibited IC_{50} values of 84.60 μg/mL. Those anti-HIV-1 RT data suggested that compound **1** has weak activity.

3. Experiment Section

3.1. General

Melting points were obtained on a Thermo Scientific Fisher-Johns Melting Point Apparatus 220 VAC (Waltham, MA, USA). NMR spectra were recorded on a JEOL 400 ECA spectrophotometer (Tokyo, Japan) in acetone-d_6 at 400 (^{1}H), 100 (^{13}C) MHz using APT experiment with TMS as the internal standard. The UV spectra was measured with Shimadzu series 1800 spectrophotometer (Kyoto, Japan). The IR spectra was recorded with Perkin-Elmer spectrum-100 FT-IR (Waltham, MA, USA). The mass spectra was recorded using a Waters LCT Premier XE (Santa Clara, CA, USA). Coloumn chromatography and radial chromatography were carried out using silica gel 60 G Cat. No. 1.07734.1000 and Si gel 60 PF_{254} Cat. No. 1.07749.1000 (Merck, Darmstadt, Germany).

3.2. Plant Material

The stem bark of *C. tetrapterum* Miq. was collected in October 2015 from Lungkut Layang Village, District Kapuas, West Kalimantan, Indonesia. The sample was identified by Mr. Ismail Rachman, Herbarium Bogoriense, Center of Biological Research and Development, National Institute of Science, Bogor, Indonesia.

3.3. Extraction and Isolation

The dried stem bark of *C. tetrapterum* Miq. (2.0 kg) were macerated in 10 L methanol twice for 2 days each. After evaporating of the solvent in a rotary evaporator, it was obtained 260 g of pale brown semi-solid. Further, the methanol extract were partitioned first with *n*-hexane (1:1 *v*/*v*). The methanol extract was added with water (10% *v*/*v*) to increase the polarity and then partitioned with ethyl acetate (1:1 *v*/*v*). The ethyl acetate extract (35 g) was subjected to coloumn chromatography over silica gel

and eluted with *n*-hexane-ethyl acetate (from 9:1 to 1:1) to give fractions A–C. Fraction B was then subjected further to coloumn chromatography and eluted with *n*-hexane-ethyl acetate (from 9:1 to 1:1) to produce subfractions B_1–B_3. Subfraction B_3 was purified by planar radial chromatography using *n*-hexane-chloroform (from 3:7 to 7:3), chloroform and chloroform-ethyl acetate 9:1 to yielded compound **1** (10 mg).

3.4. Anti-HIV Reverse Transcriptase Activity

The anti-HIV-1 RT inhibition of compound **1** was evaluated at Institute of Tropical Desease, Universitas Airlangga by a non-radioactive immunocolorimetric assay [13].

4. Conclusions

A new pyranoxanthone compound, 5,9,11-trihydroxy-2,2-dimethyl-3-(2-methylbut-3-en-2-yl) pyrano[2,3-a]xanthen-12(2*H*)-one (**1**) was isolated from the stem bark of *Calophyllum tetrapterum* Miq. This compound showed inactive toward anti-HIV-1 RT.

Supplementary Materials: ^{1}H-NMR, ^{13}C-NMR, HMQC, HMBC and HRESIMS spectra are reported in the supplementary materials as Figures S1–S8 and structure refinement parameters as Table 1.

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