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Research Article

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Chemical constituents of the bark of *Aleurites moluccana* L. Willd.

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ABSTRACT

*The dichloromethane extract of the bark of Aleurites moluccana L. Willd. afforded 12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one (**1**), spruceanol (**2**), 3-acetylaleuritolic acid (**3**), polyprenols, triglycerides and a mixture of β-sitosterol and stigmasterol in a 4:1 ratio. The structures of **1**-**3** were elucidated by extensive 1D and 2D NMR spectroscopy.*

Keywords *Aleurites moluccana* L. Willd., Euphorbiaceae, lumbang bark, 12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one, spruceanol, 3-acetylaleuritolic acid

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INTRODUCTION

Aleurites moluccana [L] Willd. of the family Euphorbiaceae, commonly known as lumbang is an indigenous tree in Southeast Asia which grows naturally in forest areas and abandoned lands at low and medium altitudes. Many parts of the tree including the seeds, leaves, flowers, and bark are frequently used in traditional medicine. Decoction of the leaves and bark are usually administered to treat headaches, fever, hypocholesterolemia, tumors, diarrhea and dysentery [1]. The flowers and sap of *A. moluccana* are commonly used in maintaining dental hygiene [2], while the seed oil is a laxative stimulant and sometimes used like castor oil [3]. An earlier study reported the isolation of swertisin from the leaves, and acetyl aleuritolic acid from the bark which were found to exhibit antimicrobial properties [4]. Moluccanin, a coumarinolignoid [5] from *A. moluccana* exhibited anti-bacterial [6] and antiviral activities [7]. Another study reported the isolation of three novel 3,4-seco-podocarpane-type trinorditerpenoids: moluccanic acid, moluccanic acid methyl ester and 6,7-dehydromoluccanic acid from the twigs and leaves of the tree which were found to possess cytotoxicity effects [8]. Furthermore, four new podocarpane-type trinorditerpenes were isolated from the twigs and leaves of *A. moluccana* [9]. Recently, five megastigmanes namely, (6S,9R)-roseoside, vomifoliol-9-O-β-apiofuranosyl-β-glucopyranoside, 3-oxo-α-ionol-O-β-apiofuranosyl-β-glucopyranoside, 3-oxo-α-ionol-O-β-glucopyranoside, and debiloside were isolated from the leaves of *A. moluccana* [10].

In this study, the isolation and structure elucidation of 12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one (**1**), spruceanol (**2**), and 3-acetylaleuritolic acid (**3**) (Fig. 1), along with polyprenols, triglycerides, β-sitosterol and stigmasterol from the CH_2Cl_2 extract of the air-dried bark of *Aleurites moluccana* [L] Willd. are reported.

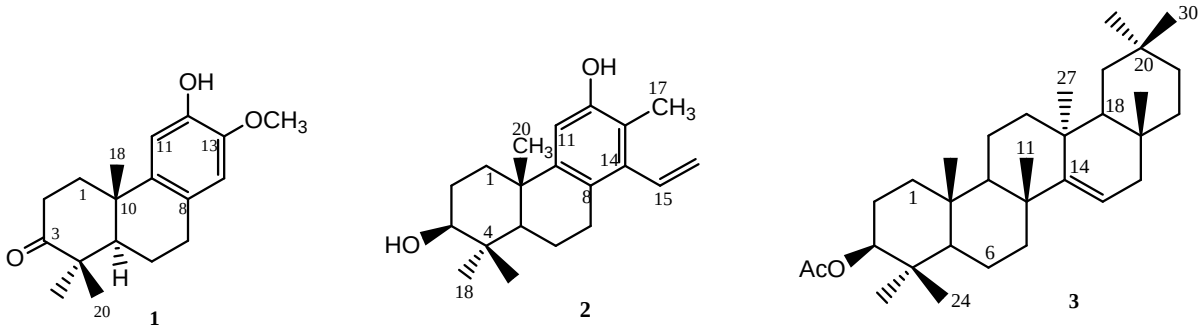

Fig. 1. Chemical constituents of *Aleurites moluccana* bark: 12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one (1), spruceanol (2), and 3-acetylaleuritolic acid (3)

EXPERIMENTAL SECTION

General Experimental Procedures

NMR spectra were recorded on a Varian VNMRS spectometer in $CDCl_3$ at 600 MHz for ^1H NMR and 150 MHz for ^{13}C NMR spectra. Column chromatography was performed with silica gel 60 (70-230 mesh). Thin layer chromtography was performed wih plastic backed plates coated with silica gel F_{254} and the plates were visualized by spraying with vanillin/H_2SO_4 followed by warming.

Sample Collection

The bark sample was collected at the Cavite State University, Indang, Cavite in September, 2011. The bark was identified as *Aleurites moluccana* [L] Willd. at the Philippine National Museum and stored at the Cavite State University Research Center Laboratory.

Isolation

The air-dried bark (1.1 kg) of *Aleurites moluccana* was ground in a blender, soaked in CH_2Cl_2 for three days and then filtered. The filtrate was concentrated under vacuum to afford a crude extract (78.7 g) which was chromatograped using increasing proportions of acetone in CH_2Cl_2 at 10% increment. The 30% acetone in CH_2Cl_2 fraction was rechromatographed using 10% EtOAc in petroleum ether, followed by 12.5% EtOAc in petroleum ether, and finally 15% EtOAc in petroleum ether as eluents. The fractions eluted with 10% EtOAc in petroleum ether were combined and rechromatographed (4 ×) in the same solvent to afford triglycerides (4 mg). The fractions eluted with 12.5% EtOAc in petroleum ether were combined and rechromatographed (5 ×) in the same solvent to afford polyprenols (7 mg) after washing with petroleum ether. The fractions eluted with 15% EtOAc in petroleum ether were combined and rechromatographed (3 ×) in the same solvent to afford a mixture of β-sitosterol and stigmasterol (5 mg) after washing with petroleum ether. The 40% acetone in CH_2Cl_2 fraction was rechromatographed (10 ×) using 10% EtOAc in petroleum ether to afford **1** (6 mg).

The air-dried bark (0.90 kg) of *Aleurites moluccana* was ground in a blender, soaked in CH_2Cl_2 for three days and then filtered. The filtrate was concentrated under vacuum to afford a crude extract (52.9 g) which was chromatograped using increasing proportions of acetone in CH_2Cl_2 at 10% increment. The 10% acetone in CH_2Cl_2 fraction was rechromatographed (9 ×) using 10% EtOAc in petroleum ether to afford **3** (7 mg). The 40% and 50% acetone in CH_2Cl_2 fractions were combined and rechromatographed (11 ×) using Et_2O:CH_3CN:CH_2Cl_2 (0.5:0.5:9) to afford **2** (5 mg).

***12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one* (1):** colorless solid. ^{13}C NMR (150 MHz, $CDCl_3$): δ 37.62 (C-1), 34.65 (C-2), 217.37 (C-3), 47.28 (C-4), 50.64 (C-5), 20.38 (C-6), 30.60 (C-7), 127.35 (C-8), 140.16 (C-9), 36.94 (C-10), 110.51 (C-11), 143.78 (C-12), 144.71 (C-13), 111.22 (C-14), 26.89 (C-18), 21.04 (C-19), 24.48 (C-20), 55.81 (OCH_3).

***Spruceanol* (2):** colorless solid. ^{13}C NMR (150 MHz, $CDCl_3$): δ 37.33 (C-1), 27.98 (C-2), 78.74 (C-3), 38.96 (C-4), 49.32 (C-5), 18.93 (C-6), 29.27 (C-7), 125.20 (C-8), 147.79 (C-9), 37.57 (C-10), 109.68 (C-11), 151.98 (C-12), 119.07 (C-13), 139.20 (C-14), 135.42 (C-15), 119.55 (C-16), 12.83 (C-17), 28.04 (C-18), 15.34 (C-19), 24.81 (C-20).

***3-Acetylaleuritolic acid* (3):** colorless solid. ^{13}C NMR (150 MHz, $CDCl_3$): δ 37.37 (C-1), 23.45 (C-2), 80.87 (C-3), 37.67 (C-4), 55.58 (C-5), 18.72 (C-6), 40.75 (C-7), 39.02 (C-8), 49.05 (C-9), 37.92 (C-10), 17.30 (C-11), 33.65 (C-12), 37.31 (C-13), 160.57 (C-14), 116.78 (C-15), 31.33 (C-16), 51.42 (C-17), 41.43 (C-18), 35.32 (C-19), 29.29 (C-

20), 33.31 (C-21), 30.70 (C-22), 27.94 (C-23), 16.58 (C-24), 15.62 (C-25), 26.17 (C-26), 22.43 (C-27), 183.56 (C-28), 31.86 (C-29), 28.66 (C-30), 170.99, 21.30 (OAc).

RESULTS AND DISCUSSION

Silica gel chromatography of the dichloromethane extract of the bark of *A. moluccana* afforded 12-hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one (**1**), spruceanol (**2**), and 3-acetylaleuritolic acid (**3**). The structures of **1**-**3** were elucidated by extensive 1D and 2D NMR spectroscopy.

12-Hydroxy-13-methoxy-8,11,13-podocarpatrien-3-one (**1**), spruceanol (**2**) and 3-acetyl aleuritolic acid (**3**) were earlier reported as constituents of *A. moluccana* [11]. Diterpene **2** was first reported as a constituent of the root and bark of *Cunuria spruceana* which exhibited cytotoxic and antitumor activity [12]. The structure of **3** was confirmed by comparison of its ^{13}C NMR data with those reported in the literature for 3-acetylaleuritolic acid [13]. Triterpene **3** exhibited antimicrobial activity against *S. aureus* and *S. typhimurium* [14]; significant inhibitory activity on vitality of adult male worms of *O. gutturosa* [15]; strong inhibition of DNA topoisomerase II and high cytotoxicity against human lung carcinoma A549 cells [16]. The structures of polyprenols [17], triglycerides [18], β-sitosterol [19] and stigmasterol [19], were confirmed by comparison of their ^{13}C NMR data with those reported in the literature.

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