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Chemical Constituents of *Duranta erecta* L. Flowers

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ABSTRACT

*Chemical investigation of the dichloromethane extract of the flowers of Duranta erecta L. has led to the isolation of oleanolic acid (**1**), a mixture of α-amyrin (**2a**) and β-amyrin (**2b**) in a 3:1 ratio, phytyl fatty acid esters (**3**), and triacylglycerols (**4**), The structures of **1**-**4b** were identified by comparison of their NMR data with literature data.*

Keywords: Duranta erecta, Verbenaceae, oleanolic acid, phytyl fatty acid esters, α-amyrin, β-amyrin, triacylglycerols

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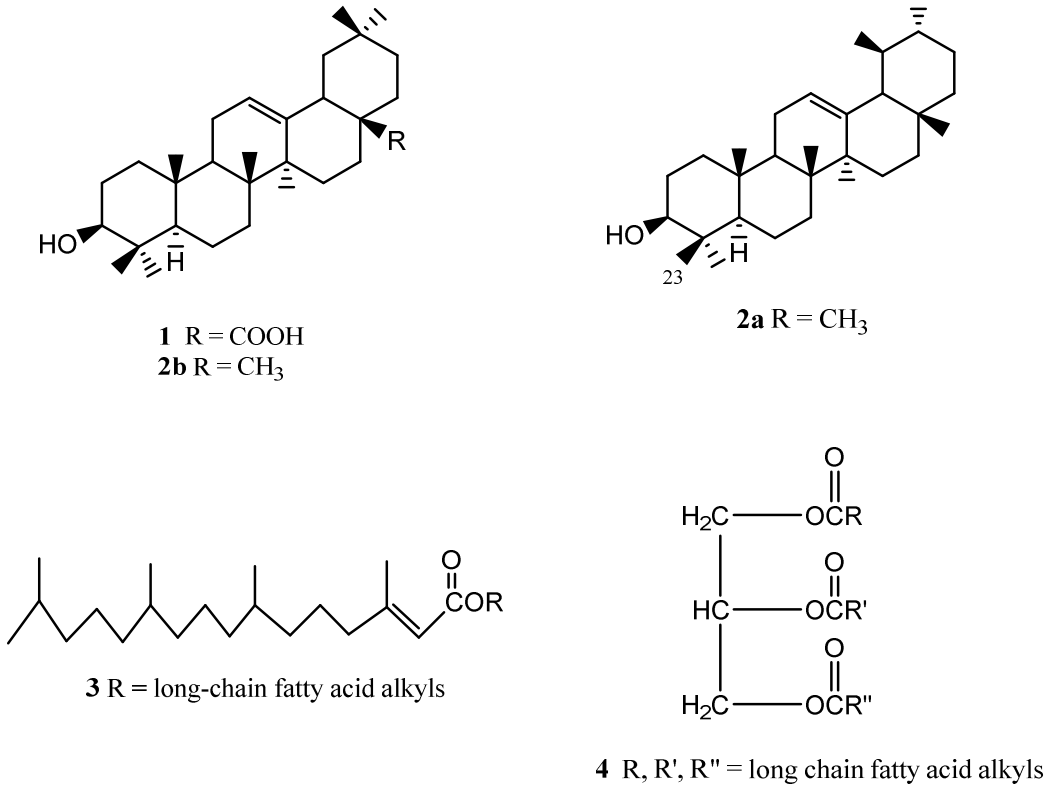
INTRODUCTION

Duranta erecta L. (syn. *D. repens* L.) of the family Verbenaceae is an ornamental plant which originated fom South America [1,2]. In Chinese medicine, the fruits were used for treatment of malaria and the leaves are employed for the treatment of abscesses [3]. The leaves of *D. erecta* yielded iridoid glucosides, duranterectosides A, B, C and D, durantosides I and II, lamiide, lamiidoside and verbascoside. Duranterectoside A durantosides I, II and III, and lamiidoside were also isolated from the stems [4]. The leaves of *D. repens* yielded durantoside IV, durantoside V, oleanolic acid, ursolic acid, (E)-cinnamic acid, β-sitosteryl-3-O-β-D-glucopyranoside, (E)-p-methoxycinnamic acid, kusaginin, glucose, durantoside I, and durantoside II [5]. The chloroform extract of stem and ethanol extract of fruits exhibited potent antishigellosis activity and moderate activity against some pathogenic bacteria and fungi. A mixture of β-amyrin and 12-oleanene 3β, 21β-diol from these extracts showed mild to moderate antimicrobial ativity [6]. Phenylethanoid glycoside acteoside, the iridoid lamiide and the saponin pseudo-ginsenoside-RT1 were isolated from the methanol extract of *D. repens*. Acteoside showed an IC_{50} of 3.05 +/- 0.09 µg/mL in the DPPH assay, while lamiide and pseudo- ginsenoside-RT1 were inactive [7]. The leaves of D. repens yielded durantanin IV and V, a bidesmosidic saponin, oleanolic acid, three phenylethanoids and five flavonoids [8]. Another study reported the isolation of the flavonoids 3,7,4′-trihydroxy-3′-(4-hydroxy-3-methylbutyl)-5,6-dimethoxyflavone,3,7-dihydroxy-3′-(4-hydroxy-3-methylbuty l)-5,6,4′-trimetho xyflavone and diterpenes 6β-hydroxy-15,16-epoxy-5β,8β,9β,10α-cleroda-3,13(16),14-trien-18-oic acid and 2β-hydroxy-15,16-epoxy-5β,8β,9β,10α-cleroda-3,13,14-trien-18-oic acid from D. repens [9]. Furthermore, 24-ethylcholest-5-en-3-α-ol, naringenin, 3,4-dihydroxy-β-phenethyl-O-α-

rhamnopyranosyl-4-*O*-caffeoyl-α-D-glucopyranosid, lamiide, α-glucopyranosyl-α-fructopyrano side, and α-galactopyranosyl-α-glucopyranosyl-fructopyranoside were isolated from the ethanol extract of *D. repens*. The petroleum ether extract afforded a mixture of hydrocarbons ranging from C15 - C27, fatty acids methyl esters and fatty acids with palmitic acid as the main component (46%) [10]. Moreover, *trans*-cinnamic acid was isolated from the methanol extract of the fruits of *D. repens* [11].

In this study, we report the isolation of oleanolic acid (**1**), a mixture of α-amyrin (**2a**) and β-amyrin (**2b**), phytyl fatty acid esters (**3**), and triacylglycerols (**4**) from the flowers of *D. erecta*. The structures of **1**-**4** are presented in Fig. 1.



1 R = COOH
2b R = CH_3

2a R = CH_3

3 R = long-chain fatty acid alkyls

4 R, R', R" = long chain fatty acid alkyls

Fig. 1. Chemical structures of oleanolic acid (1), α-amyrin (2a) and β-amyrin (2b), phytyl fatty acid esters (3), and triacylglycerols (4) from the flowers of *D. erecta*

MATERIALS AND METHODS

General Experimental Procedure

NMR spectra were recorded on a Varian VNMRS spectrometer in $CDCl_3$ at 600 MHz for ^{1}H NMR and 150 MHz for ^{13}C 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 F_{254} and the plates were visualized by spraying with vanillin/H_2SO_4 solution followed by warming.

Sample Collection

Samples of the flowers of *Duranta erecta* were collected from Sta Rosa, Laguna, Plippines in January 2015. The samples were authenticated at the Botany Division, Philippine National Museum.

General Isolation Procedure

A glass column 12 inches in height and 0.5 inch internal diameter was used for the chromatography. The crude extracts were fractionated by silica gel chromatography using increasing proportions of acetone in CH_2Cl_2 at 10% increment by volume as eluents. Five milliliter fractions were collected. All fractions were monitored by thin layer chromatography. Fractions with spots of the same R_f 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.

Isolation of the chemical constituents from the Flowers of Duranta erecta

The air-dried *D. erecta* flowers (70 g) were ground in a blender, soaked in CH_2Cl_2 for 3 days and then filtered. The solvent was evaporated under vacuum to afford a crude extract (1.86 g) which was chromatographed using increasing proportions of acetone in CH_2Cl_2 in 10% increments by volume. The 20% acetone in CH_2Cl_2 fraction was rechromatographed using 7.5% EtOAc in petroleum ether. The less polar fractions were combined and rechromatographed using 7.5% EtOAc in petroleum ether to afford **3** (4 mg). The more polar fractions were combined and rechromatographed using 7.5% EtOAc in petroleum ether to afford **4** (5 mg). The 30% acetone in CH_2Cl_2 fraction was rechromatographed (2 ×) using 15% EtOAc in petroleum ether to afford a mixture of **2a** and **2b** (6 mg) after washing with petroleum ether. The 90% acetone in CH_2Cl_2 fraction was rechromatographed (2 ×) using CH_3CN:Et_2O:CH_2Cl_2 (0.5:0.5:9, v/v) to afford **1** (3 mg) after washing with petroleum ether.

RESULTS AND DISCUSSION

Silica gel chromatography of the dichloromethane extract of the leaves of *D. erecta* yielded **1**–**4b**. The *N*MR spectra of **1** are in accordance with data reported in the literature for olenolic acid [12]; **2a** for α-amyrin [13]; and **2b** for β-amyrin [13]; **3** for phytyl fatty acid esters [14]; and **4** for triacylglycerols [15]. The 3:1 ratio of the mixture of α-amyrin (**2a**) and β-amyrin (**2b**) was deduced from the integrations and intensities of the ^{1}H NMR resonances for the olefinic protons of **2a** at δ 5.24 (t, *J* = 3.6 Hz, H-12) and **2b** at δ 5.27 (t, *J* = 3.6 Hz, H-12) [13].

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