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2-ETHOXYCARBONYL-1-HYDROXYANTHRAQUINONE FROM *RUBIA AKANE*

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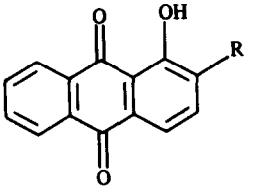
Key Word Index—*Rubia akane*; Rubiaceae; roots; 1-hydroxy-2-methylanthraquinone; 2-ethoxycarbonyl-1-hydroxyanthraquinone.

Abstract—From the roots of *Rubia akane*, 1-hydroxy-2-methylanthraquinone and a new anthraquinone, 2-ethoxycarbonyl-1-hydroxyanthraquinone were isolated. The structure of the new compound was elucidated by spectroscopy and synthesis.

INTRODUCTION

The roots of *Rubia akane* Nakai (=*R. cordifolia* L. var. *mungista* Mig., Rubiaceae) have been used in herbal medicine and as a natural colorant in Japan. The Rubiaceae family is a rich source of anthraquinones [1]. During analysis of natural colorants by HPLC with a diode array detector, we found an unknown anthraquinone in the chloroform extract of *R. akane*. The present paper describes the isolation and identification of a new anthraquinone (**1**).



1 R = CO_2Et
2 R = Me

RESULTS AND DISCUSSION

The purification of the chloroform extract by medium pressure chromatography and preparative TLC gave two anthraquinones, one of which was directly identified by HPLC, UV and ^1H NMR comparison with an authentic sample of 1-hydroxy-2-methylanthraquinone (**2**).

The UV spectrum of the other compound (**1**) was similar to that of **2**, and the IR spectrum of **1** included chelated (1633 cm^{-1}) and non-chelated carbonyl groups (1670 cm^{-1}) of a quinone. A combination of ^1H and ^{13}C NMR counts with the mass spectrum led to the molecular formula $C_{17}H_{12}O_5$ for **1**. The compound **1** seemed to be an anthraquinone which had two substituents, a hydrogen-bonded hydroxy (δ13.53 in the ^1H NMR) and an ethoxycarbonyl groups (OEt at δ4.46 and 1.44 in the ^1H NMR, and the ester carbonyl at 1725 cm^{-1} in the IR). The ^1H NMR revealed six aromatic protons, two *ortho*-coupled (δ7.87 and 8.23) and four *ortho*-substituted phenyl (δ7.85 × 2H, 8.31 and 8.35). These data suggested that (**1**) is 2-ethoxycarbonyl-1-hydroxyanthraquinone.

This structure was confirmed by synthesis. Nitration of 2-methylanthraquinone [2] gave 2-methyl-1-nitroanthraquinone, which was converted to 1-amino-2-carboxyanthraquinone by Scholl's method [3]. The amino group was substituted for hydroxy through the diazo intermediate to give 2-carboxy-1-hydroxyanthraquinone. After esterification, the synthetic 2-ethoxycarbonyl-1-hydroxyanthraquinone was found to be identical with the natural material by comparison of HPLC, UV and ^1H NMR.

EXPERIMENTAL

Isolation. The plants were collected in Tsukuba, Japan by Dr M. Satake during January 1987. Dried roots (28.5 g) were extracted with $CHCl_3$ (500 ml). After solvent evapn, the residue (*ca* 230 mg) was chromatographed under medium pressure by Kusano C.I.G. column CPS-HS-221-1 (100 × 22 mm). Elution with hexane–EtOAc (40:1) afforded a pure yellow solid (4 mg), which was identified as 1-hydroxy-2-methylanthraquinone (**2**) by HPLC, UV and ^1H NMR. The other fraction eluted with C_6H_6–Me_2CO (20:1) was further purified by rechromatography and prep. TLC [Kieselgel 60F_{254}, 0.25 mm, C_6H_6–Me_2CO (30:1)] to give **1** (3 mg) as an orange solid; mp 126° (reddish orange needles from MeOH); EIMS (probe) 70 eV, *m/z* (rel. int.): 296 $[M]^+$ (100), 251 $[M-EtO]^+$ (98), 224 $[M-C_3H_4O_2]^+$ (81), 194 (17), 167 (15), 139 (53); UV λ_{max}^{EtOH} nm: 224sh, 228, 252, 279sh, 335, 403.5; +NaOH 245, 278, 306, 517; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1725, 1670, 1633, 1593, 1258, 1126; ^1H NMR (400 MHz, $CDCl_3$): δ1.44 (3H, *t*, *J* = 7.2 Hz, Me), 4.46 (2H, *q*, *J* = 7.2 Hz, CH_2), 7.84–7.86 (2H, *m*, H-6 and H-7), 7.87 (1H, *d*, *J* = 8.1 Hz, H-3), 8.23 (1H, *d*, *J* = 8.1 Hz, H-4), 8.31, 8.34–8.36 (2H, *m*, H-5 and H-8), 13.53 (1H, *s*, OH); ^{13}C NMR (100 MHz, $CDCl_3$): δ14.28 (Me), 61.73 (CH_2), 117.23 (C-9a), 118.15 (C-4), 125.18 (C-2), 127.25, 127.49 (C-5 and

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C-8), 133.23, 133.30 (C-8a and C-10a), 134.56, 134.93 (C-6 and C-7), 136.10 (C-4a), 138.57 (C-3), 162.46 (C-1), 164.74 (CO_2), 182.00 (C-10), 188.43 (C-9).

Synthesis. Nitration of 2-methylanthraquinone (5.0 g) by KNO_3 (2.5 g) in conc H_2SO_4 (25 ml) afforded 2-methyl-1-nitroanthraquinone (5.2 g) [2]. ^{1}H NMR (200 MHz, *d*-DMSO): δ2.37 (3H, *s*, Me), 7.93–7.98 (2H, *m*, H-6 and H-7), 8.02 (1H, *d*, J = 8.1 Hz, H-3), 8.11–8.23 (2H, *m*, H-5 and H-8), 8.32 (1H, *d*, J = 8.1 Hz, H-4). According to Scholl's method [3], 2-methyl-1-nitroanthraquinone (3.20 g) was refluxed in 30% KOH–MeOH (70 ml) to give crude 1-amino-2-carboxyanthraquinone (0.94 g), ^{1}H NMR (400 MHz, *d*-DMSO): δ7.21 (1H, *d*, J = 7.8 Hz, H-3), 7.69, 7.76 (each 1H, *t*, J = 7.4 Hz, H-6 and H-7), 7.95, 8.02 (each 1H, *d*, J = 7.4 Hz, H-5 and H-8), 8.18 (1H, *d*, J = 7.8 Hz, H-4), 9.31 (2H, *br s*, NH_2), a part of which was converted to the ethyl ester, mp 201–202° (reddish orange needles from $CHCl_3$–MeOH): ^{1}H NMR (400 MHz, $CDCl_3$): δ1.36 (3H, *t*, J = 7.1 Hz, Me), 4.33 (2H, *q*, J = 7.1 Hz, CH_2), 7.47 (1H, *d*, J = 8.1 Hz, H-3), 7.67, 7.73 (each 1H, *m*, J = 1.3 and 7.5, H-6 and H-7), 8.17, 8.24 (each 1H, *dd*, J = 1.3 and 7.5 Hz, H-5 and H-8), 8.25 (1H, *d*, J = 8.1 Hz, H-4) 8.56, 9.76 (each 1H, *br s*, NH_2). Reaction of 1-amino-2-carboxyanthraquinone (30 mg) with $NaNO_2$ (30 mg) in 3 M H_2SO_4 (2 ml) with ice-cooling for 1 hr was followed by addition of H_2O (4 ml) and warming at 85° for 2 hr. The crude product was refluxed for 7 hr in EtOH (0.3 ml) and $CHCl_3$ (2.5 ml) containing two drops of conc H_2SO_4. After purification by prep. TLC [Kieselgel 60F_{254}, 1 mm, C_6H_6–Me_2CO (30:1)] 2-ethoxycarbonyl-1-hydroxyanthraquinone (8 mg) was obtained; mp 127–129° (reddish orange crystals from MeOH), which was identical to the natural product by HPLC [TSK-GEL ODS-120T, 250 × 4.6 mm, 40°, 10% aq. HOAc–MeOH (7:3→1:9)], UV and ^{1}H NMR.

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OCHNABIANTHRONE: A *TRANS*-9,9′-BIANTHRONE FROM *OCHNA PULCHRA**

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Key Word Index—*Ochna pulchra*; Ochnaceae; root bark; ochnabianthrone; circular dichroism.

Abstract—(−)-*trans*-2,2′-Digeranyloxy-7,7′-dimethyl-4,4′,5,5′-tetrahydroxy-9,9′-bianthrone, (−)-ochnabianthrone, was isolated from the root bark of *Ochna pulchra*. The circular dichroism curve and therefore the absolute configuration at 9,9′ (*R*,*R*′ or *S*,*S*′) is similar to that of (−)-sennidin A_1.

INTRODUCTION

Ochna pulchra Hook. f. (Ochnaceae) is a woody plant of central and southern Africa [2–4], which is commonly known in parts of Zimbabwe by the vernacular names 'umnyelenyele' or 'muparamhosva' [2, S. Sibanda and C. Nyanyira, unpublished results]. While its mature leaves are regarded as good cattle feed, the immature leaves are suspected of stock poisoning [2]. The widespread use of this plant in traditional medicine in Zimbabwe as an anti blood-parasitic agent and in the treatment of skin diseases [S. Sibanda and C. Nyanyira, unpublished results] has led to this phytochemical investigation. Previous studies on the genus resulted in the isolation of biflavonyl ethers, *C*-glycosylflavones and a furanoflavone from the aerial parts of *O. squarrosa* [5, 6], biflavones from *O. pumilia* and *O. atropurpurea* [7, 8]. In addition, glycerides had been detected in the fruits of *O. squarrosa* [9, 10] and *O. atropurpurea* [8]. However, with regards to *O. pulchra*, only glycerides had been detected in the fruit [11]. In this communication we report the isolation and character-

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