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ANTHRAQUINONES FROM *CASSIA SOPHERA* HEARTWOOD

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Key Word Index—*Cassia sophera*; Leguminosae; heartwood; 1,2,7-trihydroxy-6,8-dimethoxy-3-methylanthraquinone; 1,2,6-trihydroxy-7,8-dimethoxy-3-methylanthraquinone.

Abstract—From the heartwood of *Cassia sophera* two new isomeric anthraquinones, 1,2,7-trihydroxy-6,8-dimethoxy-3-methyl- and 1,2,6-trihydroxy-7,8-dimethoxy-3-methylanthraquinone have been isolated along with 1-octadecanol and quercetin.

INTRODUCTION

Cassia sophera is well known for its high medicinal values [1] and the roots and flowers of the plant have been investigated in the past [2–4]. The present report describes the isolation and characterization of anthraquinones obtained from the heartwood of this plant.

RESULTS AND DISCUSSION

Dried heartwood shavings were extracted with boiling ethanol and the concentrated ethanolic extract fractionated into benzene- and ethyl acetate-soluble fractions. From the benzene fraction four anthraquinones have been isolated and studied [5]. The ethyl acetate extract yielded 1-octadecanol and quercetin identified by direct comparison with authentic samples along with two anthraquinones, **1** and **2**, mp 258° and 234° (decomposition), respectively.

The mixture of the two anthraquinones on methylation yielded a single Me ether **3** thereby suggesting that the two anthraquinones were isomeric with similar substitution patterns.

A characteristic peak at 1180 cm^{-1} in the IR spectrum of the Me ether and the absence of free OH absorption indicated that methylation was complete. In the ^1H NMR the signal at δ 3.8 [*s*(*br*), 15 H] for OMe protons indicated the presence of five such groups, which was further corroborated by quantitative estimation (Zeisel's method). The presence of a C-Me group at the β-position was indicated by the signal at δ 2.25 (*s*, 3 H). The signal at δ 7.3 (*s*, 2 H) for aromatic protons suggested the highly substituted nature of the Me ether with only two free positions at H-4 and H-5. Based on this evidence, the compound could be assigned the structure 1,2,6,7,8-pentamethoxy-3-methylanthraquinone **3**, further substantiated by its elemental analysis and mp 130° (lit. mp 132–133°) [6, 7].

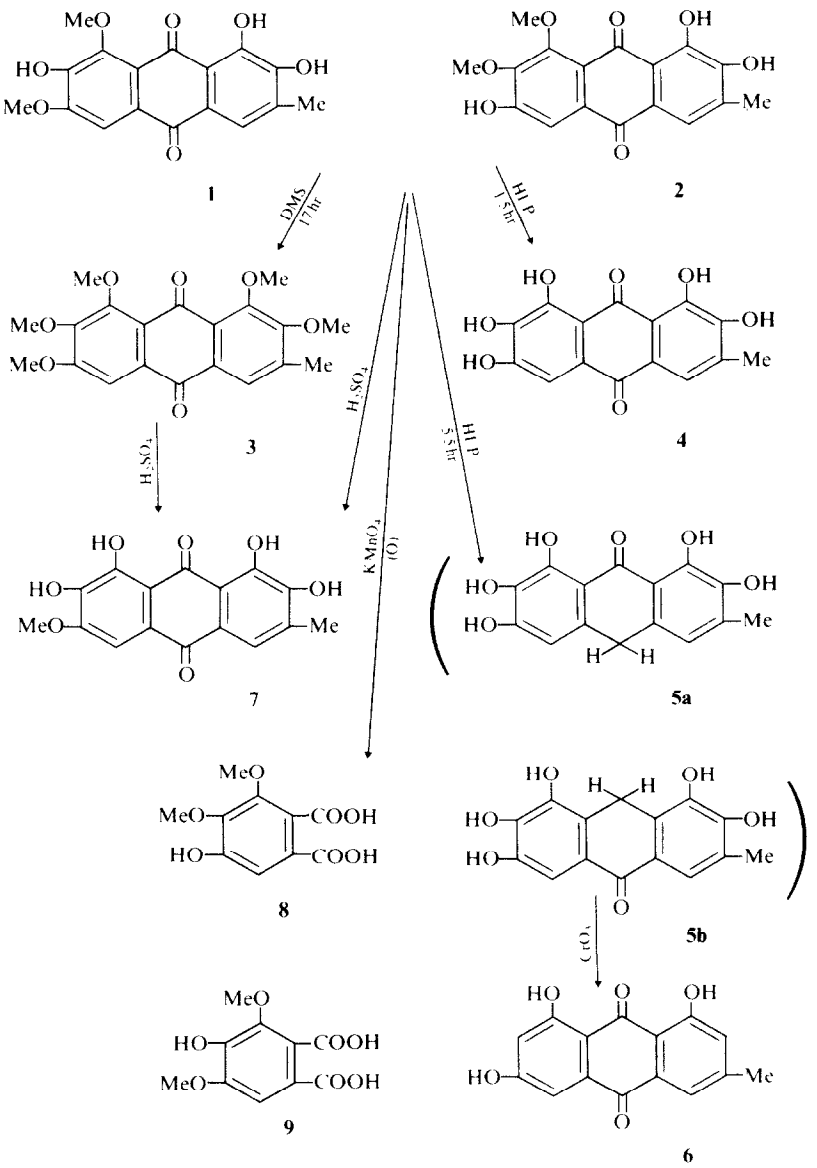
Demethylation of the mixture of the two anthraquinones by refluxing with HI and red P for 1.5 hr also gave one product **4**, confirming further that both these isomeric pigments have similar hydroxylation patterns and differ in the relative positions of OH and OMe groups. **4** was identified as 2,7-dihydroxyemodin on the basis of colour reactions [10–14], mp 320° (lit. mp 320°) [6], undepressed mmp and direct comparison with a synthetic sample [15].

Prolonged reduction of the same mixture of anthraquinones with HI/P gave a product (λ_{max}^{EtOH} 360 nm) which on chromic acid oxidation gave a yellow compound mp 255° (λ_{max}^{EtOH} 435 nm), identified as emodin **6** by direct comparison with an authentic sample. Prolonged reduction of anthraquinones with HI/P followed by chromic acid oxidation proceeds via anthrone formation resulting in an anthraquinone with fewer β-OH groups [8, 9] (unusual elimination of β-OH in conjugation with α-OH occurs). Hence, **5** could be the corresponding anthrone (**5a** or **5b**).

Partial demethylation of **3** with sulphuric acid gave a product different from **4** and it was identified as 2,7-dihydroxyphyscion **7** by colour reactions [10–14] and mp 270° (lit. mp 273°) [6]. Position 6 is known to be resistant to demethylation with sulphuric acid [7, 9]. Demethylation of the original mixture of two anthraquinones resulted in two products, which were found to be similar to **4** and **7**. This clearly indicated the presence of a free OH at C-6 in one anthraquinone, while it is substituted as OMe in the other.

The anthraquinones **1** and **2** both analysed for two OMe groups. By usual colour tests [10–14] each was found to contain only a 1,2-dihydroxy system thus fixing the position of one OMe group at position 8 in both compounds. Therefore, it could be concluded that the difference must be in the substitution with OMe at either C-6 or C-7.

Permanganate oxidation of the mixture of **1** and **2** yielded three products. Two were identified as 3,4-dimethoxy-5-hydroxyphthalic acid **8** and 3,5-dimethoxy-4-hydroxyphthalic acid **9** by direct comparison with authentic samples (by TLC only). The third compound gave a positive test for *o*-dihydroxy groups [14] and a bathochromic shift of 10 nm with borax which suggested that it could be a catechol derivative. This evidence further supported the conclusion that one ring in both **1** and **2** is the same while in the other, the difference is in the position of OMe and OH groups at positions 6 and 7. Therefore, structures **1** and **2** have been assigned to these anthraquinones.



EXPERIMENTAL

The plant material was supplied in an air-dried state by United Chemical and Allied Products, Calcutta.

Extraction. Heartwood shavings (4 kg) were extrd with EtOH (15 l.) at reflux temp. The EtOH extract was concd under red. pres. (150 ml) and fractionated into C_6H_6- and EtOAc-soluble fractions.

A white ppt. separated out from the concd EtOAc fraction on cooling, which was filtered and crystallized from EtOAc and petrol as white crystals (1-octadecanol), mp 61°, mmp 62° (lit. 63°); ν_{max}^{KBr} cm^{-1}: 3300, 2905, 1450, 1050, 1124, 730; acetate, mp 33° (lit. 35°) [16]. ν_{max}^{KBr} cm^{-1}: 2900, 2850, 1740, 1465, 1368, 1240, 732 and 722; oxidation product with $KMnO_4$, white crystals, mp 68°, mmp 69° (lit. mp 69.5°) [16].

The filtrate of the EtOAc fraction was concd and macerated with C_6H_6–$CHCl_3$ (1:2) when a yellow ppt. was obtained. This crystallized as pale yellow crystals (quercetin) from MeOH–$CHCl_3$ mp 309°, mmp 309° (lit. mp 310°); λ_{max}^{EtOH} nm: 256, 370; +NaOMe: 270, 320, 393; +$AlCl_3$: 276, 304 *sh*, 333, 430; +$AlCl_3$–HCl: 272, 303, 353, 401; +NaOAc: 272, 322 *sh*, 372; +NaOAc–H_3BO_3: 260, 300, 367 [17–19].

The remaining yellow soln on TLC gave two spots R_f 0.75 and 0.78 (EtOAc–MeOH, 5:1; spray: NH_3). These could be sepd by prep. TLC (Si gel G) and the compounds recovered from the adsorbent by elution with MeOH.

Methylation. To the anthraquinone mixture (2 g), DMS (15 ml), Me_2CO (300 ml) and K_2CO_3 (4 g) were added and the mixture refluxed for 17 hr. K_2CO_3 was filtered off, washed with Me_2CO and the combined filtrate and Me_2CO washings concd and poured into ice–H_2O. The solid was collected and crystallized from $CHCl_3$–MeOH as yellow crystals, mp 131°, yield 80%. C, 64.00; H, 5.40; –OMe, 41.10. $C_{15}H_5O_2(OMe)_5$ requires: C, 64.53; H, 5.37; –OMe, 41.66%. λ_{max}^{EtOH} nm: 250, 280, 356. ν_{max}^{KBr} cm^{-1}: 2980, 1660, 1620, 1580, 1280, 1180, 1080, 1020 and 940, etc. ^1H NMR (80 MHz, $CDCl_3$) δ: 7.3 (*s*, 2 H); 3.8 (*s* (*br*), 15 H); 2.25 (*s*, 3 H).

Demethylation. (a) Complete demethylation. The mixture of anthraquinones (0.06 g) in Ac_2O (3 ml) was refluxed with HI (6 ml) and red P for 1.5 hr. Unreacted HI was distilled off; the

clear soln decanted and extracted with Et_2O. The Et_2O soln was evapd and the residue crystallized from EtOAc–petrol, mp 320° (lit. 321°), yield 68%. (b) Partial demethylation. (1) Partial demethylation of mixture. The mixture of anthraquinones (0.06 g) was heated at 100° for 1.5 hr with H_2SO_4 (80%, 3 ml), cooled and poured into crushed ice (50 g). Extraction with Et_2O yielded two compounds which were separated on a Si gel column having mp 320° and 271°; yield 25 and 35%, respectively. (2) Partial demethylation of Me ether. Me ether (0.03 g) was heated with H_2SO_4 (80%; 1.5 ml) as above. The product crystallized from MeOH–$CHCl_3$, mp 270 (lit. mp 273–274°), yield 66%.

Prolonged reduction followed by CrO_3 *oxidation.* Me ether (0.06 g), HI (6 ml) and red P were refluxed for 5.5 hr. Unreacted HI was distilled off and the clear soln decanted. The reaction product was extracted in Et_2O (λ_{max}^{EtOH} nm: 360) and oxidized by dissolving it in Ac_2O (3 ml) and HOAc (1 ml) and refluxing with occasional shaking for 30 min during which time CrO_3 (0.3 g), dissolved in HOAc (3 ml), was added. The final reaction mixture was refluxed for 2 hr and then poured into ice–H_2O (50 ml). Extraction with Et_2O, concn and crystallization from EtOAc–petrol yielded yellow crystals, mp 253° (lit. 255°); λ_{max}^{EtOH} nm: 435 (lit. 436); yield 48%.

$KMnO_4$ *oxidation.* The anthraquinone mixture (0.05 g) in Me_2CO (50 ml) was refluxed with excess $KMnO_4$ at 100° for 5 hr. Excess Me_2CO was distilled off and MnO_2 dissolved by passing SO_2 gas through the soln. The clear soln was extracted with Et_2O, the Et_2O extract shaken with $NaHCO_3$ and then acidified with HCl. On concn it was found to contain three acids on TLC, R_f 0.40, 0.63 and 0.81 (EtOAc–C_6H_6, 1:4, spray: I_2). Two were identified by direct comparison with authentic samples, R_f 0.68 and 0.81 (EtOAc–C_6H_6, 1:4, v/v; spray: I_2) as 5-hydroxy-3,4-dimethoxyphthalic acid and 4-hydroxy-3,5-dimethoxyphthalic acid, respectively. The third acid gave colour tests for a catechol; λ_{max}^{EtOH} nm: 228 and $\lambda_{max}^{EtOH+borax}$ nm: 238 (bathochromic shift of 10 nm).

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