Racemochrysone, a Dihydroanthracenone from *Senna racemosa*

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Senna, Anthraquinones

From the hexane extract of the bark of the stems of *Senna racemosa* (syn. *Cassia racemosa*) a new dihydroanthracenone derivative, named racemochrysone, was isolated. Its structure was established as 8,9-dihydroxy-3-methoxy-2,2,6-trimethyl-(2H)-anthracen-1-one based on spectroscopical data, mainly 1D and 2D NMR experiments. In addition β-sitosterol, stigmasterol, chrysophanol and physcion were obtained. From the leaves extracts the piperidine alkaloid cassine and the hexitol pinitol were obtained.

Introduction

The genus *Cassia* (Leguminosae, subfamily Caesalpiniacee) has been recently divided into two genera, *Cassia* and *Senna*, and most of the old *Cassia* species changed to *Senna* (Irwing and Rupert, 1982). Several anthraquinones and some hydroanthracene derivatives have been reported as characteristic constituents of these plants. In particular two interesting tetrahydroanthracene derivatives, germichrysone (Takahashi *et al.*, 1976) and torosachrysone (Takido *et al.*, 1977) have been isolated from *Cassia torosa.* This paper describes the isolation and identification of a novel dihydroanthracenone derivative named racemochrysone (**1**), from *Senna racemosa* (syn. *Cassia racemosa*).

Results and Discussion

As part of our search for bioactive constituents of the Leguminosae family we have undertaken the study of *Senna racemosa* (syn. *Cassia racemosa*). This is a widely distributed species in the Yucatán peninsula in México, which is used in traditional indigenous medicine against diarrhea and eye infections.

From the hexane extract of the bark, two known anthraquinones, chrysophanol and physcion, the ubiquitous sterols β-sitosterol, stigmasterol and ergosterol were isolated. In addition a new dihydroanthracen-1-one derivative with an unusual alkylation pattern, named racemochrysone (**1**), was also isolated.

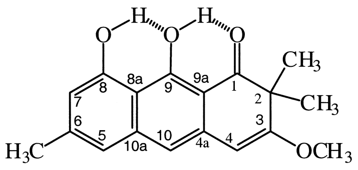

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Fig. 1.

Compound **1**, was obtained as deep orange prisms, mp 127–128°, and characterized as 8,9-dihydroxy-3-methoxy-2,2,6-trimethyl-(2H)-anthracen-1-one based on spectroscopical data, mainly 1D and 2D NMR. The high resolution mass spectrum showed the molecular formula $C_{18}H_{18}O_4$ and major fragment ions at m/z 283, 255, 240, 211 and 165. The presence of two phenolic hydroxyl protons, one of them strongly deshielded (δ 16.61 and 9.75), three aromatic protons (δ 6.61, 6.72 and 6.88), and one vinyl proton (δ 5.66) in the ^{1}H NMR spectrum together with characteristic absorptions at 260, 272, 282 and 312 nm in the ultraviolet spectrum and at 3380, 1645, 1632 and 1578 cm^{-1} in the infrared spectrum, strongly suggested an 8,9-dihydroxy-(2H)-anthracen-1-one structure. The ^{1}H NMR spectrum also showed a gem-dimethyl group (δ 1.44), an aromatic methyl group (δ 2.40) and a methoxyl group (δ 3.77).

The 2D COSY spectrum and double resonance experiments showed that two coupled aromatic protons, the broad singlet at δ 6.88 coupled with the broad doublet at δ 6.61 (J = 1.0 Hz) were both further coupled with the methyl broad singlet at δ 2.40, indicating that the aromatic methyl group must be placed at C-6. The third aromatic proton signal (δ 6.72) was assigned to H-10 and the signal at δ 5.66 to the vinyl proton H-4, because they showed to be coupled. The vinyl proton (H-4) was further coupled with the methoxyl protons, indicating that the methoxyl group must be placed at C-3 and the gem-dimethyl group at C-2. All these assignments were confirmed by 1D double resonance and 2D NOESY experiments.

An outstanding feature without a precedent is the coupling between the two hydroxyl protons, whose signals appeared as doublets (J ca. 1.0 Hz). The coupling was evident from the 2D COSY and 1D double resonance experiments.

The ^{13}C NMR spectrum confirmed the presence of 18 carbon atoms in the molecule, four methyl groups, four methines and ten tetrasubstituted carbons according to DEPT experiments. Total assignments were done according to HMQC and HMBC experiments (Table I).

Experimental

General

Melting points are uncorrected. NMR spectra were recorded in a Varian-Unity Plus at 500 MHz for ^{1}H NMR and 125 MHz for ^{13}C NMR. IR spectra were recorded in a Nicolet Magna 550 FT spectrophotometer using KBr disc. UV spectra were recorded in a Hewlett Packard 8452A spectrophotometer. GCMS analysis were performed in a Hewlett Packard 5890 series II chromatograph with poly (5%-diphenyl–95%-dimethylsiloxane) column (ULTRA II, 25 m × 0.20 mm) with a Mass Selective Detector Hewlett Packard 5972 series. HRMS were recorded in a VG Micromass ZAF-2F.

Plant material

Senna racemosa Benth was collected in Xmatkuil, Mérida in the state of Yucatán (México) in April, 1996 and identified by Dr. José Salvador Flores. A voucher specimen (J. S. Flores No. 11 219) is deposited at the Herbarium "Alfredo Barrera Marin" of the University of Yucatán.

Table I. ^{1}H NMR (500 MHz) and ^{13}C NMR (125 MHz) data for racemochrysone (**1**) in CDCl$_3$.

	C	H	HMBC correlations ^2J, (^3J)
1	206.24	– –	9-OH, (2-Me$_2$)
2	46.52	– –	2-Me$_2$, (H-4)
3	163.74	– –	H-4, (OMe), (2-Me$_2$)
4	94.31	5.66 brs	(H-10)
4a	140.67	– –	– –
5	118.92	6.88 brs	(H-7), (H-10), (6-Me)
6	144.27	– –	6-Me
7	112.01	6.61 d (ca 1.0)	(H-5), (6-Me), (8-OH)
8	158.33	– –	8-OH
8a	110.12	– –	(H-5). (H-7), (H-10), (8-OH), (9-OH)
9	167.63	– –	9-OH
9a	104.60	– –	(H-4), (H-10), (9-OH)
10	113.80	6.72 s	(H-4)
10a	133.59	– –	H-5
6-Me	22.10	2.40 brs	(H-5), (H-7)
2-Me$_2$	24.58	1.44 s	– –
OMe	55.31	3.77 s	– –
8-OH	– –	9.75 d (ca 0.8)	C-8, (C-7), (C-8a)
9-OH	– –	16.61 d (ca 0.8)	C-9, C-1, (C-8a), (C-9a)

Extraction and isolation

The dried and ground bark (500 g) was successively extracted with hexane and CH_2Cl_2 for 72 h at room temperature. The extracts were distilled under reduced pressure affording 7.5 g and 7.2 g of residue, respectively. The dried and ground leaves (700 g) were successively extracted under the same conditions with hexane, CH_2Cl_2 and Me_2CO affording 7.7 g, 36.7 g and 21.2 g of residue, respectively.

The hexane extract of the bark (7 g) was chromatographed on silica gel (120 g; 70–230 mesh) using hexane, mixtures of hexane-CH_2Cl_2 (9:1, 8:2, 6:4 v/v) and CH_2Cl_2, providing 94 fractions. Fractions 4–7 eluted with hexane were combined and rechromatographed on silica gel to give 87.3 mg of racemochrysone (**1**) as deep orange needles, mp 128°. Fractions 20–23 eluted with hexane-CH_2Cl_2 (6:4 v/v) gave white needles mp 121–125° from methanol. GC/MS analysis indicated a mixture of ergosterol, β-sitosterol and stigmasterol. Fractions 46–54 eluted with CH_2Cl_2–AcOEt (8:2 v/v) were combined and rechromatographed on silica gel providing 8 fractions; fractions 6–7 (41.7 mg) were combined and separated by preparative TLC (hexane-CH_2Cl_2, 4:6, 3 ×) to give two known compounds, physcion and chrysophanol (Tamano and Koketsu, 1982).

The CH_2Cl_2 extract of the leaves (25 g) was fractionated by CC on 400 g of silica gel (70–230 mesh) using hexane, CH_2Cl_2, Me_2CO and mixtures of Me_2CO–MeOH (9:1, 8:2 v/v) as eluants. 40 fractions were obtained. Fractions 33–7 (Me_2CO–MeOH, 9:1 v/v) which gave positive Draggendorf reaction, were combined. The residue (2.93 g) was suspended in a 5% solution of HCl and MeOH and extracted with CH_2Cl_2. The aqueous phase was neutralized with NH_4OH and extracted with CH_2Cl_2 affording the crude alkaloid cassine. Further purification of 100 mg of crude cassine by CC on acidic aluminum oxide (1 g) using CH_2Cl_2–MeOH (8:2 v/v) as eluent afforded 58 mg of pure cassine, mp 57–58° (lit. 57–58.5° Highet, 1964).

The acetone extract of the leaves precipitated spontaneously. The extract was filtered affording 3.0 g of pinitol, mp 185–186° (lit. 185–188° Calle *et al.*, 1987). The residue (17.5 g) obtained after distillation of the solvent was fractionated on 200 g of silica gel (70–230 mesh) as described above. Fractions 16–18 (11.85 g) eluted with Me_2CO–MeOH (9:1) were combined and chromatographed on 100 g of silica gel using CH_2Cl_2 and mixtures of CH_2Cl_2–MeOH as eluants. Fractions 4–5 eluted with CH_2Cl_2–MeOH (95:5) afforded 2.0 g of crude cassine.

Racemochrysone (**1**)

Deep orange prisms, mp 127–128°. HR-EIMS, 70 eV, *m/z* (rel. Int.): 298.1195 $[M]^+$ (65) (calcd. 298.1205 for $C_{18}H_{18}O_4$), 288 $[M\text{-}Me]^+$ (100), 267 $[M\text{-}MeO]^+$ (3), 255 $[M\text{-}Me\text{-}CO]^+$ (19), 240 $[M\text{-}2 \times Me\text{-}CO]^+$ (9), 211 $[M\text{-}C_4H_6O]^+$ (6), 165 $[C_{13}H_9]^+$ (9); IR ν_{max}, cm^{-1} (KBr): 3383, 2994, 2965, 2939, 1645, 1632, 1602, 1578, 1395; RMN ^1H and RMN ^{13}C (see Table I).

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Calle J., Rivera A. and Joseph-Nathan P. (1987), Pinitol from the leaves of *Gliricida sepium.* Planta Med. **3**, 303.

Highet R. J. (1964), Alkaloids of *Cassia* species. I. Cassine. J. Org. Chem. **29**, 471–474.

Irwing H. S. and Rupert C. B. (1982). The American Casiinae a synoptical revision of the leguminosae tribe Cassiae subtribe Cassinae in the new world. Memoirs of the New York Botanical Garden **35**, Part I, 454.

Takahashi S., Takido M., Sankawa U. and Shibata S. (1976), Germichrysone, a hydroanthracene derivative from seedlings of *Cassia torosa.* Phytochemistry **15**, 1295–1296.

Takido S., Takahashi S., Masuda K. and Yusukawa K. (1977), Torosachrysone, a new tetrahydroanthracene derivative from the seeds of *Cassia torosa.* Lloydia **40**, 191–194.

Tamano M. and Koketsu, J. (1982), Isolation of hydroxyanthrones from the roots of *Rumex acetosa* Linn. Agric. Biol. Chem. **46**, 1913–1914.