

Chemical Investigation on *Finlaysonia obovata* Wall.

B. P. PRADHAN* and M. M. MUKHOPADHYAY

Department of Chemistry, University of North Bengal,
Raja Rammohanpur-734 4 0

Manuscript received 4 April 1984, revised 8 March 1985,
accepted 31 July 1985

FINLAYSONIA obovata Wall.¹ (Fam.: Asclepiadaceae) is a lofty glabrous climber which is found from the Sunderbunds to Tenasserim and Malacca.

Bark: The dried and powdered bark^a (5 kg) of *Finlaysonia obovata* Wall. was extracted with petrol (b.p. 60-80°) for 30 h. It was further extracted with benzene for another 24 h.

The residue from the petrol extract was directly chromatographed over deactivated alumina. The petrol eluate on crystallisation from CHCl_3 -MeOH mixture afforded a crystalline solid (0.5 g), m.p. 218-19°, $\text{C}_{32}\text{H}_{52}\text{O}_2$ (M^+ 468); ν_{max} (KBr) 1725, 1245, 872 and 898 cm^{-1} , found to be identical with lupeol acetate⁴ (m.m.p. and co-ir).

The benzene extract was separated into neutral and acid portions by the usual method. The neutral part was chromatographed over deactivated alumina. Petrol: benzene (3:2) eluate gave crystals of β -sitosterol $\text{C}_{27}\text{H}_{48}\text{O}$ (0.4 g), m.p. 136-37°; $[\alpha]_D - 34^\circ$.

As the acid portion from the benzene extract was difficult to crystallise, acetylation with Ac_2O -Py afforded a well crystallisable solid (0.6 g), m.p. 294-95°, $\text{C}_{32}\text{H}_{50}\text{O}_4$ (M^+ 498), ν_{max} (KBr) 1700 (COOH), 1720, 1240 ($-\text{OCOCH}_3$), 998, 825 and 807 cm^{-1} ($\text{C}=\text{CH}-$). The acetate on hydrolysis with methanolic KOH furnished a hydroxy acid $\text{C}_{30}\text{H}_{48}\text{O}_3$ (M^+ 456), m.p. 287-88°; $[\alpha]_D + 72^\circ$; ν_{max} (KBr) 3440-3500 (OH), 1690 (COOH), 997, 827 and 807 cm^{-1} ; and on esterification with diazomethane in ether it formed an ester acetate $\text{C}_{32}\text{H}_{52}\text{O}_4$, m.p. 240-41°, $[\alpha]_D + 40^\circ$, identical (m.p. and ir) with an authentic specimen of acetyl methylursolate⁵.

Leaves: The dried leaves^a (3 kg) of *Finlaysonia obovata* Wall. were extracted with petrol at room temperature for 7 days and subsequently with benzene and ethanol in a Soxhlet apparatus for 24 h each.

The residue (5 g) from the petrol extract was directly chromatographed over active alumina. The petrol eluate on crystallisation from CHCl_3 -MeOH yielded two compounds of m.p. 223-24°, $[\alpha]_D + 90^\circ$ and of m.p. 235-36°, $[\alpha]_D + 80^\circ$, having the same molecular formula $\text{C}_{32}\text{H}_{52}\text{O}_2$ (M^+ 468). The ir and nmr spectra of both the compounds showed the presence of an acetyl group and a trisubstituted double bond, ν_{max} (KBr) 1725, 1240 and 825 cm^{-1} ; δ 2.05 and 5.25. Hydrolysis of the acetate with 5% methanolic KOH furnished alcohols of molecular formula $\text{C}_{30}\text{H}_{50}\text{O}$, m.p. 180-82°, $[\alpha]_D + 89$, and 190-92°, $[\alpha]_D + 85^\circ$, respectively. The parent compounds were subsequently identified as α -amyrin acetate⁶ and β -amyrin acetate⁷, respectively by

m.m.p., co-tlc and ir comparison with authentic samples.

The benzene extract left some heavy crystalline solid (8 g) which was characterised as sodium chloride. The residue from the filtrate was extracted with ether and separated into neutral and acid parts. The neutral part was purified by chromatography and crystallisation, and was identified as β -sitosterol. The acid part was esterified with diazomethane in ether solution and purified by chromatography and crystallisation to give ester $\text{C}_{32}\text{H}_{50}\text{O}_3$, m.p. 170-71°, $[\alpha]_D + 58^\circ$. On acetylation, it gave an ester-acetate $\text{C}_{34}\text{H}_{54}\text{O}_4$, m.p. 139-40°, $[\alpha]_D + 42^\circ$ which was found identical with acetyl methylursolate (m.m.p. and co-ir).

The alcohol extract was hydrolysed with dilute H_2SO_4 , extracted with ether and the solvent removed. The residue on chromatography and elution with petrol-benzene (2:3) eluted a solid $\text{C}_{30}\text{H}_{48}\text{O}$, m.p. 154-56°, $[\alpha]_D - 50^\circ$. The compound formed an acetate $\text{C}_{32}\text{H}_{50}\text{O}_2$, m.p. 137-39°, $[\alpha]_D - 34^\circ$ which was identified as stigmasteryl acetate (m.m.p. and co-ir).

Acknowledgement

The authors thanks are due to Dr. B. C. Das, Gif-Sur-Yvette, France, for mass spectra, to Prof. T. R. Govindachari, CIBA Research Centre, Bombay, for the nmr spectra and to Dr. S. K. Sengupta, EIPW, Calcutta, for optical rotations. One of them (M.M.M.) is grateful to U.G.C., New Delhi, for FIP Fellowship.

References

1. J. D. HOOKER, "Flora of British India", Vol. IV, p. 7.
2. B. P. PRADHAN and M. M. MUKHOPADHYAY, *North Bengal Univ. Rev. (Sci. Technol.)*, 1982, 3, 43.
3. B. P. PRADHAN and M. M. MUKHOPADHYAY, *North Bengal Univ. Rev. (Sci. Technol.)*, 1980, 1, 145.
4. J. SIMONSEN and W. C. J. ROSS, "The Terpenes", Cambridge University Press, 1957, Vol. 4, p. 367.
5. J. SIMONSEN and W. C. J. ROSS, "The Terpenes", Cambridge University Press, 1957, Vol. 5, p. 116.
6. J. SIMONSEN and W. C. J. ROSS, "The Terpenes", Cambridge University Press, 1957, Vol. 4, p. 148.
7. J. SIMONSEN and W. C. J. ROSS, "The Terpenes", Cambridge University Press, 1957, Vol. 4, p. 244.

Pmr and Mass Spectral Behaviour of 12 α -Hydroxyrotenoids

M. KRISHNAMURTI* and MANGAL NATH**

Department of Chemistry, University of Delhi,
Delhi-110 007

Manuscript received 9 November 1979, revised
10 September 1981, accepted 31 July 1985

DETAILED and diagnostic pmr^{1,2} and mass^{3,4} spectral studies have been made for rotenoids. The number of 12 α -hydroxyrotenoids occurring

** Present address: Department of Chemistry, Hans Raj College, University of Delhi, Delhi-110 007.

in nature are not many. Their pmr⁸ and mass⁶⁻⁸ spectra have not been subjected to detailed study.

We isolated certain rotenoids⁹ for a study of their structure-activity relationship as inhibitors of respiration *in vitro*¹⁰. A scrutiny of the pmr and mass spectral behaviour of 12_a-hydroxyrotenoids has led to certain important observations and their stereochemical implications.

Pmr features: The signal for C-1 proton is diagnostic for B/C ring stereochemistry in rotenoids^{1,2}. This signal for 12_a-hydroxyrotenoids compared with the one for the corresponding rotenoids was found shifted upfield by about δ 0.20-0.25 (Table 1). The signal for C-4 appeared at the same value at around δ 6.4.

TABLE 1—SIGNALS FOR THE PROTONS OF RING A
Compd. δ (CDCl₃)

	H-1	H-4
(+)-Dolineone (1)	6.73	6.41
cis-(+)-12 _a -Hydroxydolineone (2)	6.48	6.43
(-)-Rotenone (3)	6.75	6.42
cis-(-)-12 _a -Hydroxyrotenone (4)	6.53	6.44
(+)-Erosone (5)	6.74	6.45
cis-(+)-12 _a -Hydroxyerosone (6)	6.52	6.46

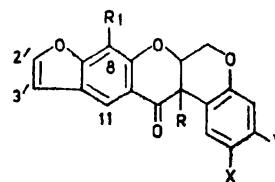
This shift in C-1 proton signal implies a slight conformational change at the B/C ring juncture. This conformational change brought about by the introduction of 12_a-OH group led to loss in potency as inhibitors of (NADH)-linked segment of the mitochondrial respiratory chain, particularly in the initial stages of inhibition process¹⁰.

The two hydrogens of the methylenedioxy group appeared as non-equivalent doublets (*J* 1.5Hz) separated by δ 0.05 and situated around δ 5.7. In closely related cooccurring non-rotenoids, viz. pachyrrhizin (10), neotenone (11) and erosone (12), the same two protons were equivalent and gave a combined singlet (Table 2). The non-equivalence of the two hydrogens of the methylenedioxy group can be accounted for on the basis of

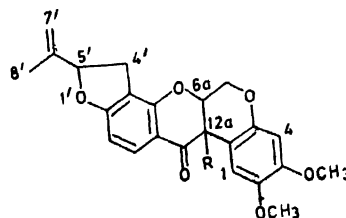
TABLE 2—PROTON SIGNALS FOR METHYLENEDIOXY GROUP OF RING A

Compd.	δ (CDCl ₃)
(+)-Dolineone (1)	5.92 (d, <i>J</i> 1.5Hz) and 5.86 (d, <i>J</i> 1.5Hz)
cis-(+)-12 _a -Hydroxydolineone (2)	5.78 (d, <i>J</i> 1.5Hz) and 5.73 (d, <i>J</i> 1.5Hz)
cis-(+)-12 _a -Hydroxypachyrrhizone (9)	5.81 (d, <i>J</i> 1.5Hz) and 5.76 (d, <i>J</i> 1.5Hz)
Pachyrrhizin (10)	5.97 (2H, s)
Neotenone (11)	5.86 (2H, s)
Erosone (12)	5.97 (2H, s)

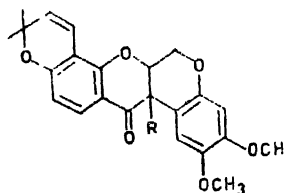
the chirality in the rotenoid skeleton about B/C ring junction and concomitant different environment for the 2 hydrogens.



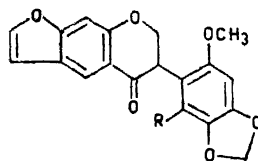
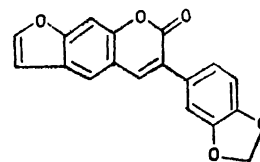
- (1) R=H, R₁=H, X, Y=OCH₃O
 (2) R=OH, R₁=H, X, Y=OCH₃O
 (5) R=H, R₁=H, X=Y=OCH₃
 (6) R=OH, R₁=H, X=Y=OCH₃
 (9) R=OH, R₁=OCH₃, X, Y=OCH₃O



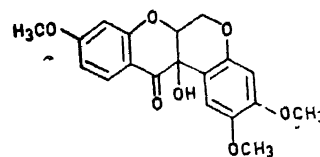
- (3) R=H
 (4) R=OH



- (7) R=H
 (8) R=OH



- (11) R=H
 (12) R=OH



(13)

Mass spectral study: Important mass spectral fragments of optically active 12_a-hydroxyrotenoids, viz. 12_a-hydroxydolineone (2), 12_a-hydroxypachyrrhizone (9), 12_a-hydroxyerosone (6), 12_a-hydroxymunduserone (13) and 12_a-hydroxyrotenone (4) are shown in Table 3. The same for *trans*-(±)-12_a-hydroxyrotenone prepared synthetically¹¹ is also given.

The R.D.A. and R.D.A. with H-transfer were major fragmentation pathways for all the compounds. The chromene fragment B₁ (from R.D.A.) formed the base peak for all the compounds except for 9 where by coincidence the ethylene and the ketene fragments from R.D.A. with H-transfer happen to have the same *m/e* value of 191, making

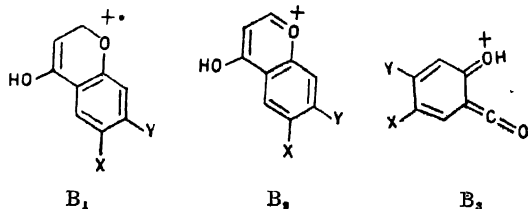
TABLE 3—IMPORTANT MASS SPECTRAL FRAGMENTS OF COMPOUNDS

	m/e (% Abundance)				
	2	9	6	13	4
M^{+}	352 (23.60)	382 (18.80)	368 (26.80)	358 (21.70)	410 (69.5)
$M^{+} - H_2O$	334 (0.51)	364 (0.94)	350 (1.23)	340 (0.40)	392 (2.44)
B_1	192 (100)	192 (72.64)	208 (100)	208 (100)	208 (100)
B_2	191 (89.50)	191 (*)	207 (40.5)	207 (42.50)	207 (63.41)
B_3	165 (16.50)	165 (13.90)	181 (7.84)	181 (6.70)	181 (5.0)
B_1-30	162 (3.16)	162 (5.16)	—	—	—
B_1-15	—	—	193 (10.20)	193 (10.0)	193 (15.20)
A_1	160 (5.55)	190 (11.43)	160 (7.8)	154 (10.0)	202 (3.2)
A_2	161 (16.67)	191 (*)	161 (9.8)	161 (5.40)	203 (9.80)

Mass spectrum *trans*-(±)-12_a-hydroxyrotenone: 410 (23.7%, M^{+}), 392 (3.12), 208 (100), 207 (39.6), 203 (15.62), 202 (2.10), 197 (13.54), 191 (12.9), 190 (4.2), 189 (8.3), 183 (18.8), 169 (17.70), 155 (15.6).

* A part of base peak.

it the most abundant base peak. For 9 fragment B_1 is still prominent (72.6%).



The parent ion M^{+} is extraordinarily abundant for 12_a-hydroxyrotenone (69.5%) signifying its chemical and mass spectral stability than other 12_a-hydroxyrotenoids⁹. The low abundance of $M-H_2O^{+}$ dehydrated parent ions conforms to thermodynamic stability of B/C *cis* ring fusion. The fragment B_1 loses formaldehyde (-30 amu) or CH_3 radical (-15 amu) for methylenedioxy and dimethyl compounds, respectively as usual. An important ion B_3 (B_1-27 amu) is uniformly observed for all the compounds and seems to be diagnostic of 12_a-hydroxy compounds. Such odd-electron species as B_3 are considered diagnostically more important than equally abundant even-electron fragments⁹.

Ketene fragments A_1 (R.D.A) and A_2 (R.D.A. with H-transfer) are observed in close similarity to rotenoids.

12_a-Hydroxyrotenone (*trans*-synthetic) fragmented through R.D.A. etc. in close similarity to other *cis*-compounds but for two unique fragments at m/e 190(4.2%) and 189(8.3%) which were practically absent for *cis*-(-)-12_a-hydroxyrotenone (4). These are presumably dehydration products of B_1 (m/e , 208) and B_2 (m/e , 207). Also dehydrated parent ion $M-H_2O^{+}$ was much more abundant ($M-H_2O^{+}$

= 1 : 8) as compared with 4 having the same ratio at 1 : 29. Once again comparative thermodynamic stability of *cis*-structure is vindicated.

References

1. L. CROMBIE and J. W. LOWN, *J. Chem. Soc.*, 1962, 775.
2. D. G. CARLSON, D. WEISLEDER and E. M. TALLENT, *Tetrahedron* 1973, 19, 2731.
3. R. I. REED and J. M. WILSON, *J. Chem. Soc.*, 1963, 5949.
4. H. AUDIER, *Bull. Soc. Chim. Fr.*, 1966, 9, 2892.
5. G. L. D. KRUPADANAM, P. N. SARMA, G. SRIMANNARAYANA and N. V. SUBBA RAO, *Tetrahedron Lett.*, 1977, 2125.
6. W. D. OLLIS, C. A. RHODES and I. O. SUTHERLAND, *Tetrahedron*, 1967, 23, 4741.
7. P. N. SARMA, G. SRIMANNARAYANA and N. V. SUBBA RAO, *Indian J. Chem., Sect. B*, 1976, 14, 152.
8. F. W. McLAFFERTY, "Interpretation of Mass Spectra", W. A. Benjamin Inc., Mass., 1973, pp. 86-44.
9. A. J. KALRA, M. KRISHNAMURTI and M. NATH, *Indian J. Chem., Sect. B*, 1977, 15, 1084.
10. M. NATH, T. A. VENKITASUBRAMANIAN and M. KRISHNAMURTI, *Bull. Environ. Contam. Toxicol.*, 1980, 24, 116.
11. M. E. OBERHOLZER, G. J. H. RALL and D. O. ROUX, *Tetrahedron Lett.*, 1974, 2211.

Extractive Spectrophotometric Determination of Trace Amounts of Rhodium with Diphenylcarbazide in Presence of α -Picoline

KABITA SARMAH and H. K. DAS*

Department of Chemistry, Gauhati University, Gauhati-781 014

Manuscript received 22 May 1984, revised 15 February 1985, accepted 31 July 1985

DIPHENYLCARBAZIDE has long been known as a reagent for the detection and determination of a number of metals¹. It is used as a sensitive and specific reagent for spectrophotometric determination of chromium². With a slightly acidic rhodium(III) solution, it forms a purple coloured complex on heating for a few minutes. In the present investigation, it is found that in presence of α -picoline, rhodium(III) forms with diphenylcarbazide a bluish-violet complex, extractable into methyl isobutyl ketone. Diphenylcarbazide and α -picoline make a suitable ligand pair for the determination of rhodium spectrophotometrically. This forms the basis of the method which has proved to be very sensitive and selective.

Experimental

Absorbance measurement were made with a Hilger-Uvispek spectrophotometer in matched quartz cells of 1 cm optical path.

A stock solution of rhodium(III) was prepared by dissolving rhodium trichloride (Johnson Matthey) in 1 N hydrochloric acid and standardised by known