Phytochemistry, 1972, Vol. 11, pp. 811 to 814. Pergamon Press. Printed in England.

SHORT COMMUNICATION

# LIGNANS FROM *MACHILUS EDULIS**

P. L. MAJUMDER and A. CHATTERJEE
Department of Pure Chemistry, University College of Science, Calcutta-9, India

and

G. C. SENGUPTA
Department of Chemistry, Hoogly Mohsin College, Hooghly, India

(*Received* 28 *May* 1971)

**Abstract**—(+)-Guaiacin, a *trans*(2,3)-*trans*(3,4) phenyltetralin lignan, has been isolated for the first time from the stem bark of *Machilus edulis* King (Lauraceae) together with dihydroguaiaretic acid. Contrary to earlier observations, (+)-guaiacin has been converted by two different routes to (+)-galbulin, a new optical antipode of (−)-galbulin. Physical properties including NMR and MS data as well as chemical degradation reactions are reported for the first time for these lignans.

IN RECENT years the chemistry of naturally occurring lignans[1] has evoked considerable interest for their antitumor activities.[2] As a part of our research programme on the chemistry of Indian Medicinal plants, we have recently isolated two such compounds, A and B from the bark of the lauraceous species, *Machilus edulis* King. The present communication deals with the identification of these compounds.

*Machilus edulis*,[3] locally known as 'Lapche kawla' is a timber tree common in Birch Hill and Senchal area of Darjeeling. The seeds of this plant are edible and enjoy a local reputation as an effective antioxidant for butter. Compound A, $C_{20}H_{24}O_4$ ($M^+$ 328), m.p. 198–200°, $[\alpha]_D^{25}$ + 46° ($CHCl_3$) shows UV absorption at $\lambda_{max}^{EtOH}$ 285 and 240 (sh) nm (log $\epsilon$, 3·80 and 2·80) and produces a deep purple colour with conc. $H_2SO_4$, characteristic[4] of a phenyltetralin lignan. Functional group analysis of A indicates the presence of two O—$CH_3$, two C—$CH_3$ and two active hydrogen atoms. The IR spectrum indicates that it is aromatic and it shows an intense hydroxyl band at 3400 $cm^{-1}$. The presence of two phenolic hydroxyl groups was confirmed by the formation of a diacetate, $C_{24}H_{28}O_6$ ($M^+$ 412), m.p. 116° with acetic anhydride and pyridine and a dimethyl ether, $C_{22}H_{28}O_4$ ($M^+$ 356), m.p. 130–31°, $[\alpha]_D^{25}$ + 8° ($CHCl_3$) with either diazomethane or dimethyl sulphate and alkali. The diethyl ether, $C_{24}H_{32}O_4$ ($M^+$ 384) m.p. 114°, was also prepared.

The 60 Mc PMR spectrum of A in $CDCl_3$ [$\delta$, 6·69 (s), 6·61 (s), 6·21 (s), each for 1 Ar–H; 6·50–6·68 (2 Ar–H); 3·80 (2 Ar–$OCH_3$, s), 2·65 (3-benzylic protons, m), 1·53 (2H, m), 0·84 (1 C—$CH_3$, d, $J$ = 6 counts/sec), 1·05 (1 C—$CH_3$, d, $J$ = 6 counts/sec)] is comparable

* A preliminary report of the work was presented in the 58th Annual Session of Indian Science Congress, Abs. Part III, p. 193 (1970).

[1] W. M. HEARON and W. S. MACGREGOR, *Chem. Rev.* **55**, 958 (1955).
[2] M. G. KELLY and J. L. HARTWELL, *J. Natl. Cancer Inst.* **14**, 967 (1954).
[3] A. M. COWAN and J. M. COWAN, *The Trees of Northern Bengal including Shrubs, Woody Climbers, Bamboos, Palms and Tree Ferns.*
[4] G. K. HUGHES and E. RITCHIE, *Austral. J. Chem.* **7**, 104 (1934).

to that of galbulin[5] [$\delta$, 6·78 (2 Ar–H, broad S), 6·60 (2 Ar–H, broad s), 6·20 (1 Ar–H, s); 3·80 (s), 3·76 (s), 3·73 (s) and 3·50 (s), each for 1 Ar–$OCH_3$; 2·60 (3-benzylic protons, m), 1·53 (2H, m), 0·82 (1 C–$CH_3$, d, $J = 6$ counts/sec) and 1·04 (1 C—$CH_3$, d, $J = 6$ counts/sec)]. The PMR spectrum of the dimethyl ether of A is also superimposable upon that of galbulin.

The above observations suggest the identity of A with guaiacin (I), a phenyltetralin lignan, the diethyl ether of which is known. Contrary to earlier observations,[6] A readily formed a dimethyl ether, with either diazomethane or dimethyl sulphate and alkali, which has finally been identified as a hitherto unknown optical antipode of (—)-galbulin[4, 7] (II). In conformity with this observation, the dimethyl ether of A on dehydrogenation with 10% Pd/C in ethanol afforded a compound, $C_{22}H_{24}O_4$, ($M^+$ 352), m.p. 178–179° which was found to be identical (mixed m.p. superimposable IR spectra) with dehydroguaiaretic acid dimethyl ether[6, 8] (III). Further, the dimethyl ether of A on oxidation with alkaline $KMnO_4$, furnished two crystalline products. One of these was characterized as *o*-veratroylveratric acid (IV), $C_{18}H_{18}O_7$ ($M^+$ 346), m.p. 220° (lit. m.p. 221–222°),[4, 9] while the other (from its IR and MS data) appears to have the structure (V), $C_{18}H_{18}O_6$ ($M^+$ 330), m.p. 178–179°,[10] formed by the lactonization of the intermediate diphenylcarbinol acid (VI), which gives rise to the *o*-veratroylveratric acid (Chart 1). The phthalide (V) was not obtained by the earlier workers[4] from galbulin.

Finally, because of the non-availability of authentic diethyl guaiacin, the problem of assignment of the relative positions of the hydroxyl and methoxyl groups in A had to be resolved and this has been accomplished by establishing the identity (mixed m.p. and superimposable IR spectra) of the acid (VII) $C_{20}H_{22}O_7$ ($M^+$ 374), m.p. 205–206°, obtained by oxidation of diethyl ether of A (VIII) with potassium dichromate in acetic acid[6] and that of the diethyl ether of $\alpha$-conidendrin (IX) of known structure by the same method as well as by alkaline $KMnO_4$.[11]

A is, therefore, identical with guaiacin (I), also designated as 2,3-dimethyl-6-hydroxy-7-methoxy-4- (4′-hydroxy-3′-methoxy phenyl)-1,2,3,4-tetrahydronaphthalene, a *trans*(2,3)-*trans*(3,4)-phenyltetralin lignan,[7, 12] first isolated from the bark of *Machilus edulis*. The structure (I) for A is also consistent with its MS fragmentation which shows important peaks at *m/e* 328 ($M^+$), 272, 271, 255, 241, 240, 204, 189, 137. The MS fragmentations of dimethyl ether of A are the same as those reported for galbulin.[13]

Careful chromatographic resolution of the crude benzene extract of *M. edulis* resulted in the isolation of a second lignan, B in extremely poor yield. This compound appears to be dihydroguaiaretic acid[14] (X), $C_{20}H_{26}O_4$, m.p. 98° from its IR ($\nu_{max}^{Nujol}$ 3300 $cm^{-1}$) and mass spectral data [*m/e* 330 ($M^+$, 56·1), 149 (19·5), 138 (26·8), 137 (100)] which are in conformity with the structure (X).

[5] The PMR spectral data of galbulin have not been reported earlier. The spectrum of an authentic sample, kindly supplied by Professor W. C. TAYLOR, Dept. of Organic Chemistry, University of Sydney, Australia, was recorded by us for the purpose of comparison.
[6] F. E. KING and J. G. WILSON, *J. Chem. Soc.* 4011 (1964).
[7] A. W. SCHRECKER and J. L. HARTWELL, *J. Am. Chem. Soc.* **77**, 432 (1955).
[8] R. D. HAWORTH, C. R. MAVIN and G. SHELDRICK, *J. Chem. Soc.* 1423 (1934).
[9] R. D. HAWORTH and C. R. MAVIN, *J. Chem. Soc.* 1363 (1931).
[10] B. L. VANZETTI and A. OLIVERO, *Gazz. Chim. Ital.* **60**, 620 (1930).
[11] H. ERDTMAN, *Annalen* **513**, 234 (1934).
[12] A. J. BIRCH, B. MILLIGAN, E. SMITH and R. N. SPEAKE, *J. Chem. Soc.* 4471 (1958).
[13] A. PELTER, *J. Chem. Soc.* 74 (1968).
[14] B. CARNMALM, *Acta Chem. Scand.* **8**, 806 (1954).

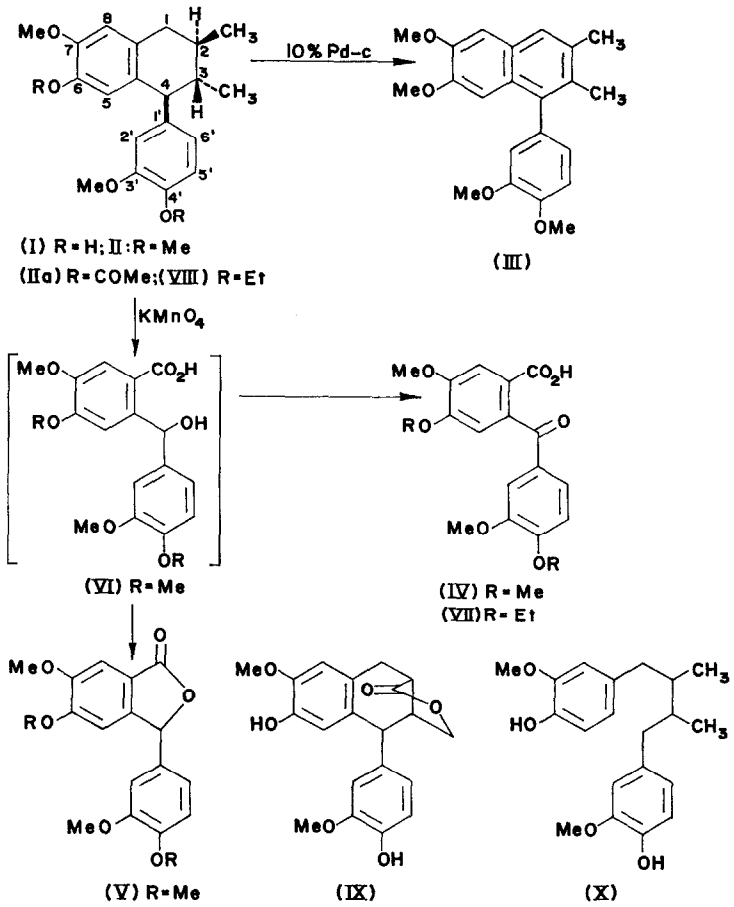


CHART 1.

## EXPERIMENTAL

*General.* M.ps were determined on the Kofler block and are uncorrected. The UV spectra were measured in 95% EtOH (aldehyde-free) and the IR spectra in Nujol mull. The analytical samples were routinely dried over $P_2O_5$ for 24 hr *in vacuo*. Anhydrous $Na_2SO_4$ was used for drying the organic solvents. Brockmann alumina and silica gel G were used for column chromatography throughout.

*Isolation of guaiacin and dihydroguaiaretic acid.* Powdered air-dried stem barks (1 kg) of *Machilus edulis* were soxhleted for 72 hr with benzene. The concentrated extract (40 g) was chromatographed over silica gel G (800 g). The petrol–benzene (1:1) eluate on concentration gave a brownish oily mass which on crystallization from petrol afforded crystals of dihydroguaiaretic acid (X, 0·002%), $C_{20}H_{26}O_4$ ($M^+$ 330), m.p. 98°.

The benzene–$CHCl_3$ (1:1) eluate, on removal of solvent gave a crude solid (0·17%) which on crystallization from benzene furnished colourless shining plates (0·15%) of guaiacin (I), $C_{20}H_{24}O_4$ ($M^+$ 328), m.p. 198–200°, $[\alpha]_D^{25}$ + 46° ($CHCl_3$). (Found: C, 73·58; H, 7·23%. Calc. for $C_{20}H_{24}O_4$: C, 73·17; H, 7·31%.)

*Acetylation of guaiacin.* Guaiacin was treated with $Ac_2O$/pyridine to yield a product, which crystallized from MeOH to give diacetylguaiacin (IIa), $C_{24}H_{28}O_6$ ($M^+$ 412), m.p. 116°; IR: $\nu_{max}^{Nujol}$ 1750 and 1260 $cm^{-1}$ ($OCOCH_3$); NMR: δ, 2·18 (3H, s, $OCOCH_3$); 2·28 (3H, s, $OCOCH_3$); 6·34 (s), 6·88 (s), each for 1 Ar–H; 6·55–6·81 (broad s, 3 Ar–H); 3·81 (6H, s, 2 Ar–$OCH_3$); 2·70 (m, 3 benzylic protons); 1·55 (2H, m); 1·02 (3H, d, $J$ = 6 counts/sec, C–$CH_3$); 0·84 (3H, d, $J$ = 6 counts/sec, C–$CH_3$).

*Methylation of guaiacin.* Guaiacin, on treatment with $CH_2N_2$ in $Et_2O$ or with $Me_2SO_4$ in NaOH, yielded dimethylguaiacin (II) crystallized from MeOH, $C_{22}H_{28}O_4$ ($M^+$ 356), m.p. 130–131°, $[\alpha]_D^{25}$ + 8·0° ($CHCl_3$). (Found: C, 74·16; H, 7·72%. Calc. for $C_{22}H_{28}O_4$: C, 74·15; H, 7·86%.) The identity of the compound with authentic galbulin was established by mixed m.p., TLC and superimposable IR spectra.

*Dehydrogenation of dimethylguaiacin with* Pd/C. Dimethylguaiacin (200 mg) was mixed with 100 mg of

10% Pd/C and 10 ml EtOH. The mixture was heated in a sealed tube for 5 hr in an oil bath at 200°. The product (180 mg) was crystallized from MeOH to give dehydroguaiaretic acid dimethyl ether (III), $C_{22}H_{24}O_4$ ($M^+$ 352), m.p. 178–180°; (Lit.[6,8] m.p. 178–179°) UV: $\lambda_{max}^{EtOH}$ 237, 285, 314 and 330 nm (log $\epsilon$, 5·0, 4·15, 3·61 and 3·75).

*Oxidation of dimethylguaiacin with alkaline potassium permanganate.* Dimethylguaiacin (500 mg), 2 g $KMnO_4$, 7 ml of 5% NaOH and 70 ml $H_2O$ were heated under reflux for 6 hr, after which the pink colour of permanganate was destroyed with EtOH. After the usual workup the product was taken into $CHCl_3$ and chromatographed over silica gel. The petrol–benzene (1:1) eluate on evaporation of solvent gave a solid (10 mg) which was crystallized from MeOH to give the phthalide (V), $C_{18}H_{18}O_6$, m.p.[10] 177–178°. [Mass: *m/e* 330 ($M^+$ 91·8), 290 (19·7), 271 (13·1), 255 (14·8), 166 (13·1), 165 (100), 164 (21·3).] The solid (15 mg) from the benzene eluate on removal of solvent was crystallized from MeOH to give *o*-veratroylveratric acid (IV), $C_{18}H_{18}O_7$, m.p. 220° (Lit. m.p.[11] 221–222°). [Mass: *m/e* 346 ($M^+$ 48·7), 271 (21·8), 209 (21·8), 183 (12·7), 182 (100), 167 (20), 165 (60).

*Ethylation of guaiacin.* Guaiacin (500 mg) was ethylated with EtI (3 ml) in dry acetone (20 ml) and anhydrous $K_2CO_3$ (1 g) under reflux for 42 hr in the usual way[6] to furnish diethyl guaiacin (VIII, 450 mg), crystallized from MeOH, $C_{24}H_{32}O_4$ ($M^+$ 384), m.p. 114° (Lit. m.p.[6] 114–115°).

*Oxidation of diethylguaiacin with potassium dichromate and glacial acetic acid.* A mixture of diethylguaiacin (400 mg), HOAc (32 ml) and $K_2Cr_2O_7$ (1·6 g) was heated on a water bath for 6 hr and then poured in crushed ice and extracted with ether. The ether extract was shaken with $NaHCO_3$, the aqueous layer was acidified with conc. HCl and extracted with ether, dried and chromatographed over silica gel. The benzene eluate gave a solid which was crystallized from benzene–petrol mixture (1:1) to give crystals of 3-methoxy-4-ethoxy-6-(3′-methoxy-4′-ethoxy benzoyl) benzoic acid (VII, 40 mg), m.p. 205° (Lit. m.p.[6] 206–207·5°).

*Ethylation of α-conidendrin.* α-Conidendrin (2 g) was ethylated with EtI (11 ml) and anhydrous $K_2CO_3$ (5 g) in dry acetone according to standard procedure[6] to furnish diethyl-α-conidendrin (1·5 g), crystallized from MeOH, $C_{24}H_{28}O_6$ ($M^+$ 412) m.p. 177–178° (Lit. m.p.[15] 178–179°).

*Oxidation of diethyl-α-conidendrin with potassium dichromate and glacial acetic acid.* A mixture of diethyl-α-conidendrin (500 mg), HOAc (40 ml) and $K_2Cr_2O_7$ (2 g) was heated on a water bath for 7 hr and the reaction mixture was processed as in the case of diethylguaiacin. The acid (35 mg) isolated from this reaction was found to be identical (mixed m.p., TLC and superimposable IR spectra) with 3-methoxy-4-ethoxy-6-(3′-methoxy-4′-ethoxy benzoyl)benzoic acid (VII) obtained from diethylguaiacin.

*Oxidation of diethyl-α-conidendrin with potassium permanganate in acetone.* Diethyl-α-conidendrin (500 mg) in acetone (25 ml) was treated with $KMnO_4$ (2·5 g) portion-wise during a period of 56 hr at room temp. The reaction product was worked up according to standard procedure.[11] The crude acid (20 mg) obtained in this reaction was purified by chromatography over silica gel and was found to be identical with (VII).

*Acknowledgements*—The authors express their sincere thanks to Professor W. C. Taylor, Department of Organic Chemistry, University of Sydney, Australia, Drs. A. W. Schrecker, National Institutes of Health, Bethesda, Washington, for generous supply of authentic galbulin, W. L. Shilling, Crown Zellerback Corporation, Camas, Washington for large supply of authentic α-conidendrin, B. C. Das, C.N.R.S., Gif-Sur Yvette, France, for mass spectral measurements, D. N. Roy, University of Toronto, Canada, for NMR spectra and S. C. Pakrashi, I.I.E.M., Calcutta for optical rotations and IR spectra. One of the authors (G.C.S.) is highly indebted to Professors M. K. Chatterjee, Principal, Hooghly Mohsin College and S. N. Dhar, Head of the Department of Chemistry of the College for encouragement and laboratory facilities and also to the Education Department, Government of West Bengal for providing financial assistance.

[15] R. D. HAWORTH and G. SHELDRICK, *J. Chem. Soc.* 636 (1935).

*Key Word Index—Machilus edulis*; Lauraceae; lignans; guaiacin.