Phytochemistry, Vol. 26, No. 3, pp. 813–817, 1987.
Printed in Great Britain.

0031-9422/87 $3.00 + 0.00

CONFIGURATION AND CONFORMATION OF DIHYDROFLAVONOLS FROM *ACACIA MELANOXYLON*

LAI YEAP FOO

Chemistry Division, Department of Scientific and Industrial Research, Private Bag, Petone, New Zealand

(*Revised received* 23 *July* 1986)

Key Word Index—*Acacia melanoxylon*; Leguminosae; heartwood; 2,3-*cis*-3',4',7,8-tetrahydroxydihydroflavonol; 2,3-*trans*-3',4',7,8,-tetrahydroxydihydroflavonol; 2,3-*trans*-3',4',7-trihydroxy-5-methoxy-dihydroflavonol; configuration; conformation; biogenesis.

Abstract—The isolation of two novel dihydroflavonols, namely (−)2,3-*cis*-3',4',7,8-tetrahydroxydihydroflavonol and (±)2,3-*trans*-3',4',7-trihydroxy-5-methoxydihydroflavonol together with the known (±)2,3-*trans*-3',4',7,8-tetrahydroxydihydroflavonol from *Acacia melanoxylon* is described. ^{1}H NMR study shows the aryl substituent at C-2 in the 2,3-*trans* compounds is in the normal equatorial conformation but the 2,3-*cis* isomer, in contrast, has the aryl substituent in the unusual axial conformation. The elucidation of the 2,3-*cis* configuration of the dihydroflavonol and its co-occurrence with the metabolically related 2,3-*cis*-leucoanthocyanidins or flavan-3,4-diols provide the first evidence that the biosynthesis of flavan-3-ols and proanthocyanidins with the 2,3-*cis* configuration occur in a parallel pathway to that of the 2,3-*trans* compounds.

INTRODUCTION

The heartwood of *Acacia melanoxylon*, a medium size tree native to Australia, has been the subject of a number of chemical investigations and its flavonoids shown to consist of the pyrogallol A-ring and the catechol B-ring hydroxylation pattern [1–4]. The constituents of the wood are of interest because the unreactive nature of the pyrogallol A-ring, with respect to nucleophilic reactions [5, 6], has enabled compounds normally of transient existence such as the dihydroflavonols and flavan-3,4-diols to accumulate in the wood. Such components are absent or occur at low levels in plants where the flavonoid A-ring is based on the more reactive resorcinol and especially the phloroglucinol systems and, in these plants, the predominant constituents are the proanthocyanidins. A recent study [7] of the wood of *A. melanoxylon* revealed the presence of four isomeric leucoanthocyanidins, all possessing the 2,3-*cis* configuration, which contrasts markedly to the reported 2,3-*trans* configuration of its dihydroflavonol precursor [1, 6, 8]. This anomaly prompted the present reinvestigation of the flavonoid constituents of this heartwood.

RESULTS AND DISCUSSION

Previous studies by Clark-Lewis and others [9–11] have demonstrated the relative instability of dihydroflavonols with the 2,3-*cis* configuration as compared to their 2,3-*trans* isomers. This finding may partly account for the lack of detection of the natural 2,3-*cis*-dihydroflavonols [1, 6, 8, 12] in plants which on biogenetic considerations should have compounds with such a configuration as precursors. Thus, to minimize possible chemical transformation during isolation, the extraction and work-up described here were performed at ambient temperature where possible.

In addition to the four leucomelacacinidins [7], (−)3',4',7,8-tetrahydroxyflavanone, 3',4',7,8-tetrahydroxyflavonol and a chromatographically homogeneous fraction consistent with a dihydroflavonol constitution as shown by its UV absorption spectrum and Zn/HCl colour reaction [9] was also isolated. The expected pyrogallol A-ring and the catechol B-ring hydroxylation pattern is readily evident from both the ^{13}C and ^{1}H NMR spectral data. However, the heterocyclic carbon chemical shift region in the ^{13}C NMR spectrum of the sample exhibits clear duplication of C-2 and C-3 chemical shifts of dihydroflavonol structures. The nature of the chemical shifts and the magnitude of the proton–proton coupling constants of its ^{1}H NMR spectrum suggest a mixture of the 2,3-*cis*-and 2,3-*trans*-3',4',7,8-tetrahydroxydihydroflavonols in an approximate ratio of 2:1, respectively. Initial chromatography over a reverse phase column (MCI gel) using MeOH–H_2O and examination by ^{1}H NMR spectroscopy of the eluants indicates some resolution of the mixture, the 2,3-*cis* compound being slightly more mobile than the 2,3-*trans* product. Complete separation of the isomers was finally achieved by repeated chromatography using this system and a preliminary report of part of this work has been published [14]. The present report deals more fully with the chemical structures and conformations of the dihydroflavonols and provides a chemical basis for the relative instability of the 2,3-*cis* configuration.

Conformation of dihydroflavonols

The most distinguishing features in the NMR data between the two isomeric dihydroflavonols are their respective H-2 and C-2 chemical shifts and the H-2 and H-3 coupling constants. The normal magnitude of the coupling constants ($J_{2,3}$ = 11.8 Hz) in the 2,3-*trans* com-

pound confirms the diaxial orientation [15] of the H-2 and H-3 protons and hence the equatorial conformation of the aryl substituent or catechol ring at C-2. The *cis* isomer in contrast has $J_{2,3}$ = 3.4 Hz. This value is unusually large compared with the coupling constants of the analogous 3-methoxyflavanones with 2,3-*cis* configuration [8] and may be explained on the basis of conformational differences between the 3-methoxyflavanones and the dihydroflavonol. The relatively small value of $J_{2,3}$ for the 2,3-*cis*-3-methoxyflavanones is consistent with the C-3 substituents in the axial and hence the C-2 aryl substituent in the expected equatorial conformation [15]. This conformation is also supported by $J_{2,3}$ values calculated from the equation derived by Altona *et al.* [16, 17] using the 4-substituent approach and taking into account the electronegative effects and the orientation of the oxygen substituents. Measurement of the proton–proton torsion angles of 2,3-*cis*-dihydroflavonol using Dreiding models give values of 310° for C-2 aryl equatorial and 60° for C-2 aryl axial conformations. Calculations based on the 4-substituent equation give $J_{2,3}$ = 1.9 Hz, a value consistent with the observed coupling constants [8, 10, 15] for the C-2 aryl equatorial conformation and $J_{2,3}$ = 3.7 Hz when the aryl substituent is in the less common axial conformation. This latter $J_{2,3}$ value is in agreement with the observed $J_{2,3}$ = 3.4 Hz for the 2,3-*cis*-3',4',7,8-tetrahydroxydihydroflavonol.

Further corroboration of the axial conformation may also be obtained by consideration of the H-2 chemical shifts. In assuming that the aryl substituent in 2,3-*cis*-dihydroflavonol is in the axial conformation, the H-2 will, by necessity, be in the equatorial position which cause it to be under the deshielding influence of the aromatic A-ring. Hence the equatorially orientated H-2 (δ5.49) in the *cis* compound is expected to be more downfield than the axially orientated H-2 (δ5.00) in the *trans* isomer.

Stability of the axial conformation

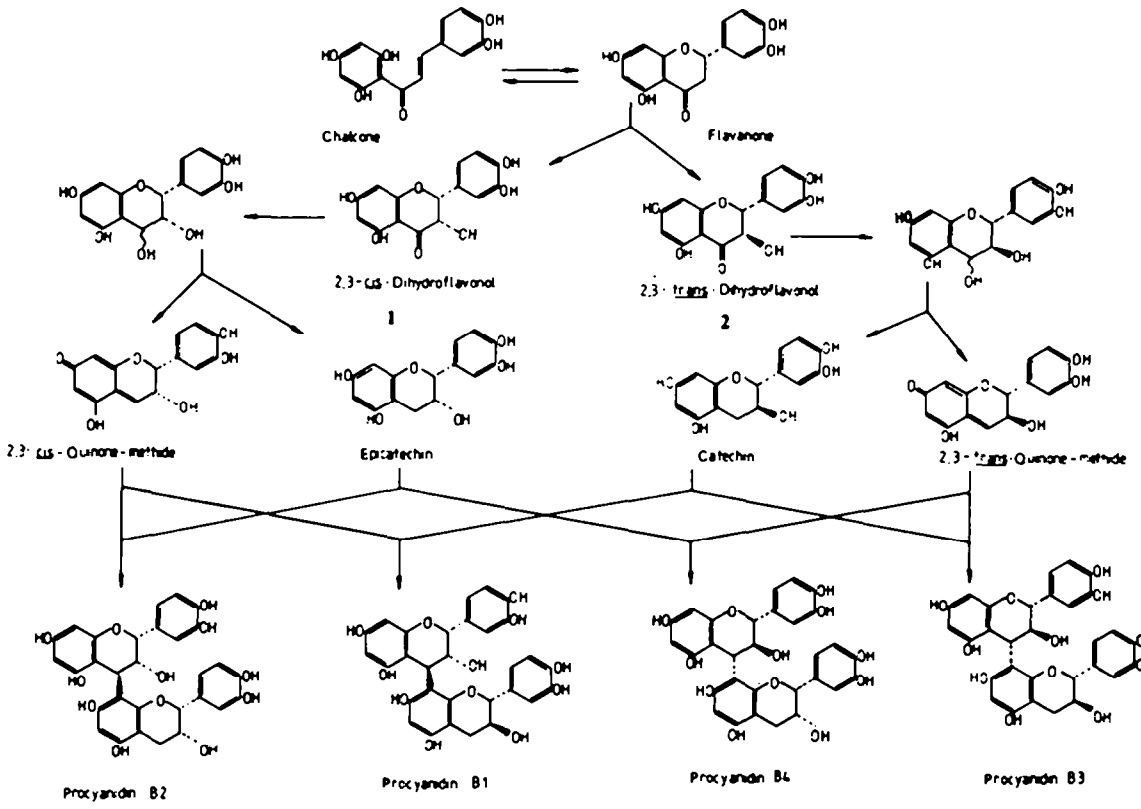
The unusual preference for the C-2 aryl axial conformation in the *cis*-dihydroflavonol is probably a result of stabilization through hydrogen-bonding interactions. A study based on Dreiding models shows that a more planar configuration between the C-3 hydroxyl and the C-4 carbonyl may be obtained with the C-2 aryl substituent in the axial conformation and hence promote more effective hydrogen-bonding between the two groups. In contrast, such planar configuration in the *trans* isomer is derived when the C-2 aryl is in the more normal equatorial conformation. This type of hydrogen-bonding interaction is supported by consideration of the published ^{1}H NMR data of 3-*O*-methyldihydroflavonols where the C-2 aryl substituents are in the equatorial conformation for both the *cis* and *trans* isomers. The C-2 axial conformation in the methylated *cis* compounds are not favoured because they can no longer be stabilized by hydrogen-bonding as in the non-methylated systems. In such cases where both the *cis* and *trans* compounds have preferred C-2 aryl equatorial conformations, the protons at C-2 will necessarily be expected to be in the axial orientation and not be affected by the anisotropy of the A-ring. Thus, the protons at C-2 are in a similar magnetic environment in these methylated compounds and the observed H-2 chemical shifts, as anticipated, are of comparable values for both *cis* and *trans* isomers [8, 10, 15].

The C-2 aryl axial conformation of the *cis*-dihydroflavonol on steric considerations may be expected to be energetically less stable than the aryl equatorial conformation of the *trans* isomer. This may partly account for its ready isomerization to the more thermodynamically stable 2,3-*trans* product. Such transformation is readily achieved by brief warming with dilute hydrochloric acid or by heating under reflux in ethanol to give racemic *trans* isomer. The racemic nature of the *trans* product implies inversion at both C-2 and C-3 of the dihydroflavonol and suggests the co-occurring racemic *trans*-dihydroflavonol is probably an artefact resulting from the epimerization of the *cis* isomer either during the experimental work-up and/or during the prolonged period of storage in the wood. The failure to detect the *cis* product in a number of past reports [6, 8, 19] could be reasonably attributed to the severe conditions used in the extraction of the plant materials.

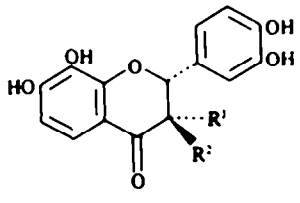
Consideration of the mechanism of the acid-catalysed inversion at C-3 suggests prior protonation at the carbonyl oxygen follows by enolization to give the thermodynamically more stable 2,3-*trans* product. Dihydroflavonols with the more common phloroglucinol A-ring hydroxylation pattern, such as dihydroquercetin, have the 5-OH most favourably placed to the C-4 carbonyl for anchimeric assistance to enolization through intramolecular hydrogen-bonding. Even in the absence of a free hydroxyl at C-5 for hydrogen-bonding, the carbonyl at C-4 is expected to be more polarized by virtue of electron donation from such *meta* oxidation pattern in the A-ring [5, 6]. These dihydroflavonols will be expected to epimerize readily to the more stable 2,3-*trans*-dihydroflavonols which could account for their predominance in extractives of plant materials.

Biogenesis of 2,3-cis-*proanthocyanidin units*

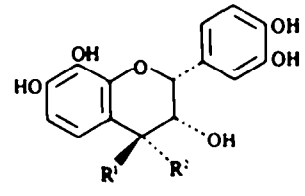
The dihydroflavonols are intermediates in the biogenesis of flavan-3-ols, proanthocyanidins [20, 21] and anthocyanidins [22], but because of the apparent absence of natural 2,3-*cis*-dihydroflavonols there has been much debate on the biogenetic routes to 2,3-*cis*-flavan-3-ols and proanthocyanidins [21, 23–25]. The isolation of 2,3-*cis*-3',4',7,8-tetrahydroxydihydroflavonol (**1**) together with its biogenetic precursor, 3',4',7,8-tetrahydroxyflavanone (**2**), as well as the co-occurring 2,3-*cis*-flavan-3,4-diols (**3**, **4**) [7] and the related proanthocyanidin dimer (**5**) [26] from *A. melanoxylon*, clearly suggests that the flavan-3-ols and proanthocyanidins with 2,3-*cis* configuration are derived in a parallel biogenetic pathway to the 2,3-*trans* analogues. The biogenesis of flavan-3-ols and proanthocyanidins of varying 2,3-stereochemistry may now be rationalized more readily by incorporating this sequence of biochemical transformation as well as the stereoisomeric quinone-methides as intermediates (Scheme 1). To accommodate the existence of both geometrical isomers, an α- or β-hydroxylase is proposed for the stereospecific hydroxylation of the flavanone at C-3 to the 2,3-*cis*- or 2,3-*trans*-dihydroflavonol, respectively. This biosynthetic scheme is consistent with isotopic tracer experiments and in particular the random loss of tritium label from C-3 during biosynthesis from labelled cinnamic acid [23]. The greater than 90% loss of the tritium label at C-3 of flavan-3-ol units cannot be accounted for by an intermediate such as the proposed flav-3-en-3-ol [23, 24]. Intermediates that lack a hydrogen at C-3 cannot conceivably pick up tritium even at low levels from an



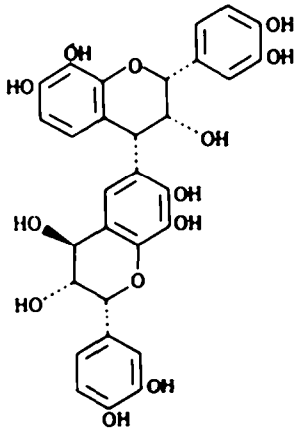
Scheme 1. Proposed biosynthetic pathways to flavan-3-ols and procyanidins.



1 R^1 = OH, R^2 = H
2 R^1 = H, R^2 = H



3 R^1 = H, R^2 = OH
4 R^1 = OH, R^2 = H



5

environment essentially devoid of tritium. The small level of tritium retention on this position is more satisfactorily explained by the loss of the tritium in labelled cinnamic acid during the three stages of the biochemical transformation leading to the formation of flavan-3-ols and proanthocyanidins. These steps which are clear from Scheme 1 are (i) the chalcone-flavanone interconversion, (ii) enolization of the flavanone and (iii) the enzymic hydroxylation of the flavanone to the dihydroflavonol. An additional process by which further tritium loss could take place is through enolization of the dihydroflavonol. However, this last process requires the presence of an enzyme system in the plant which allows for the retention of configuration at C-3.

5-O-*Methyldihydroquercetin*

The 7,8-hydroxylation pattern of the A-ring of the flavonoid constituents in *A. melanoxylon* has been apparent from the major compounds isolated here and elsewhere [1–4, 7]. Thus, the identification of a minor racemic 3,3',4',7-tetrahydroxy-5-methoxy-dihydroflavonol, or 5-*O*-methyldihydroquercetin, a compound not previously described, is an interesting anomaly. The dihydroflavonol structure is readily recognized from its ^{1}H and ^{13}C NMR spectra by the chemical shift values of the respective heterocyclic C-ring protons and carbons. The ^{1}H NMR data with aromatic ring protons at δ6.83 (H-2'), 6.85 (H-6') and 7.03 (H-5') characteristic of the catechol B-ring pattern and the two sets of doublets identifiable with the H-6 (δ6.14, J = 2.1 Hz) and H-8 (δ5.98, J = 2.1 Hz) of a phloroglucinol A-ring together with diaxial couplings ($J_{2,3}$ = 12.1 Hz) clearly suggest a very similar constitution to dihydroquercetin. A methoxyl group is also indicated by the proton chemical shift at δ3.81 and the corresponding carbon chemical shift at δ55.8, the latter resonance is associated with an aromatic methoxy group attached to the A-ring [27]. A monomethylated dihydroquercetin constitution is also supported by its mass spectrum with the $[M]^+$ at m/z 318. The presence of the methoxyl in the A-ring is indicated by the presence of the base ion (m/z 167) which arises from a retro-Diels–Alder fission of the C-ring of the dihydroflavonol [28]. The assignment of the methoxy group to C-5 is based on carbon chemical shift considerations. Firstly, there is an upfield chemical shift ($\sim$ 7 ppm) of the carbonyl carbon relative to dihydroquercetin caused by the absence of hydrogen bonding interactions between the hydroxyl at C-5 and the carbonyl function [27]. This assignment is also supported by consideration of the chemical shifts of the unsubstituted A-ring carbons. Methylation at the 7-hydroxyl would be expected to cause an upfield shift to both the C-6 and C-8 resonances while methylation at the 5-hydroxyl would be limited to the chemical shift of C-6 only. Comparison of these chemical shifts with those of dihydroquercetin shows that only the C-6 chemical shift is shifted upfield ($\sim$ 3 ppm). UV absorption studies also unambiguously confirm this assignment. The presence of a 7-hydroxyl is clear from the bathochromic shift from MeOH max. 288 to 324 nm induced by the addition of NaOAc solution to a methanolic solution of the compound. Also the bathochromatic shift (+ 26 nm) caused by the formation of a aluminium–flavonoid complex is readily destroyed by the addition of dilute hydrochloric acid indicating the absence of a 5-hydroxyl [29].

EXPERIMENTAL

Extraction. Heartwood (1 kg) of *A. melanoxylon* was extracted twice with Me_2CO–H_2O (7:3) by soaking the chips at room temp. for a minimum of 24 hr each time; the combined extracts were shaken with NaCl. The upper layer was concd on a rotatory evaporator at < 40° and the residue diluted with H_2O and filtered over glass wool. The filtrate was exhaustively extracted with EtOAc, the extracts dried (Na_2SO_4) and concd (< 40°) to give a solid residue (14 g). Fractionation of the residue by CC on LH20 using EtOH as solvent yielded two main fractions I and II, the latter being predominantly oligomeric proanthocyanidins.

3',4',7,8-Tetrahydroxyflavonol. Treatment of the residue from fraction I with EtOH H_2O (1:3) gave crystalline 3',4',7,8-tetrahydroxyflavonol (2.3 g) mp > 300°. λ_{max}^{MeOH} nm: 258, 370; ($AlCl_3$) 281, 468; ($AlCl_3$–HCl) 267, 432. MS (m/z): 302 ($[M]^+$, 100%), 153 (30%). ^{13}C NMR (acetone-d_6, ppm): 114.1 (C-6) 115.1 (C-4a), 115.3 (C-5'), 115.5 (C-2'), 115.9 (C-6'), 120.9 (C-5), 123.7 (C-1'), 133.1 (C-8), 137.2 (C-3), 145.2 (C-3', C-4'), 147.5 (C-2), 150.0 (C-7, C-8a) and 173.0 (C-4).

Further fractionation of the mother liquor from fraction I on Sephadex LH20 using EtOH H_2O (1:3) yielded two major flavonoid fractions IA and IB. The following dihydroflavonols were obtained by repeated chromatography of fraction IA on MCI gel using MeOH–H_2O (3:7) as eluant.

(−)*2,3-cis-3',4',7,8-Tetrahydroxydihydroflavonol.* (300 mg) $[\alpha]_D$ −49.5° (*c* 0.10, Me_2CO–H_2O) CD: $\Delta\varepsilon_{350}$ +4.54; $\Delta\varepsilon_{290}$ −9.34 (*c* 0.014, H_2O) R_f values on cellulose TLC using *t*-BuOH–HOAc–H_2O (3:1:1, solvent A), 0.68 and HOAC–H_2O (3:47, solvent B), 0.40. λ_{max}^{MeOH} nm: 292; ($AlCl_3$) 354; ($AlCl_3$–HCl), 292. MS (m/z): 304 ($[M]^+$, 15%), 286 (6), 153 (100), 123 (41). ^{13}C NMR (acetone-d_6 ppm): 73.5 (C-3), 82.7 (C-2), 110.7 (C-6), 114.0 (C-4a), 115.4 (C-2'), 115.9 (C-5'), 119.1 (C-6'), 120.2 (C-5), 128.7 (C-1'), 133.4 (C-8), 145.3 (C-4'), 145.8 (C-3'), 151.4 (C-8a), 152.6 (C-7), 191.4 (C-4). ^{1}H NMR (acetone-d_6): δ4.39 (*d*, J = 3.4 Hz, H-3), 5.49 (*d*, J = 3.4 Hz, H-2), 6.62 (*d*, J = 8.6 Hz, H-6), 6.82 (*s*, H-2'), 6.86 (*d*, J = 1.6 Hz, H-6'), 7.08 (*d*, J = 1.6 Hz, H-5') and 7.30 (*d*, J = 8.6 Hz, H-5). Treatment of this dihydroflavonol with dil. HCl at 60° for 5 min or heating at 75° for 48 hr in EtOH resulted in considerable epimerization to give a mixture of 2,3-*cis* and 2,3-*trans* compounds in a ratio of 15:85, respectively. Methylation with CH_2N_2–Et_2O gave 3,3',4',7,8-pentamethoxyflavonol as the main product [18].

(±)*2,3-trans-3',4',7,8-Tetrahydroxydihydroflavonol.* (120 mg) $[\alpha]_D$ 0° (*c* 0.12, Me_2CO–H_2O), R_f values of cellulose TLC, 0.68 (A) and 0.40 (B). λ_{max}^{MeOH} nm: 290; ($AlCl_3$) 350; ($AlCl_3$–HCl) 290. MS (m/z): 304 ($[M]^+$, 10%), 286 (9), 153 (100), 138 (40), 137 (42), 123 (98). ^{13}C NMR (acetone-d_6, ppm): 74.0 (C-3), 85.0 (C-2), 111.7 (C-6), 113.7 (C-4a), 116.2 (C-2'), 116.6 (C-5'), 119.7 (C-6'), 121.2 (C-5), 129.4) (C-1'), 133.3 (C-8), 145.6 (C-3'), 146.4 (C-4'), 151.6 (C-8a), 153.4 (C-7) and 194.2 (C-4). ^{1}H NMR (acetone-d_6: δ4.56 (*d*, J = 11.8 Hz, H-3), 5.00 (*d*, J = 11.8 Hz, H-2), 6.65 (*d*, J = 8.6 Hz, H-6), 6.90 (*s*, H-2'), 6.92 (*br s*, H-6'), 7.09 (*br s*, H-5'), 7.32 (*d*, J = 8.6 Hz, H-5).

(±)*3',4',7-Trihydroxy-5-methoxy-dihydroflavonol* (5-O-*methyldihydroquercetin*). (30 mg) mp 251 253° (H_2O), $[\alpha]_D$ 0° (*c* 0.07, Me_2CO–H_2O). R_f values on cellulose TLC, 0.75 (A), 0.38 (B%). λ_{max}^{MeOH} nm: 288; ($AlCl_3$) 314; ($AlCl_3$–HCl) 288; (NaOAc) 324. MS (m/z): 318 ($[M]^+$, 6%), 300 (10), 289 (26), 196 (10), 195 (10), 180 (16), 167 (100), 152 (20), 123 (54). ^{13}C NMR (acetone-d_6 ppm): 55.8 (OMe) 72.7 (3), 82.6 (C-2), 93.4 (C-6), 95.6 (C-8) 102.6 (C-4a), 115.3 (C-2', C-5'), 119.4 (C-6'), 128.5 (C-1'), 145.0 (C-4'), 145.8 (C-3'), 162.3 (C-5), 163.8 (C-8a), 164.9 (C-7) and 190.0 (C-4). ^{1}H NMR (acetone-d_6): δ3.81 (*s*, OMe), 4.68 (*d*, J = 12.1 Hz, H-3), 4.86 (*d*, J = 12.1 Hz, H-2), 5.98 (*d*, J = 2.1 Hz, H-8), 6.14 (*d*, J = 2.1 Hz, H-6), 6.83 (H-2'), 6.85 (H-6'), 7.03 (H-5').

(−)*3',4',7,8-Tetrahydroxyflavanone.* (170 mg) Obtained from

fraction 1B using MCI gel (MeOH–H_2O, 3:7) recrystallized from H_2O, mp 127–128°, $[\alpha] - 2°$ (*c* 0.10, HeOH) (lit. [18] mp 126–127°, $[\alpha] - 11.7°$). R_f on cellulose TLC, 0.74 (t BA) and 0.20 (HOAc). λ_{max}^{MeOH} nm: 290; ($AlCl_3$) 314; ($AlCl_3$–HCl) 290. MS (*m/z*): 288 ([M]$^+$, 43%), 180 (12), 153 (100), 152 (78), 136 (340), 123 (58). ^{13}C NMR (acetone-d_6, ppm): 43.8 (3), 80.4 (2), 111.1 (C-4a, C-6), 115.2 (C-2'), 116.5 (C-5'), 119.5 (C-6'), 119.7 (C-5), 131.5 (C-1'), 133.4 (C-8), 145.6 (C-4'), 145.9 (C-3'), 152.0 (C-8a), 153.2 (C-7) and 194.5 (C-4).

Acknowledgements—The author wishes to acknowledge the assistance of John Lloyd of the Forest Research Institute for the sample of *A. melanoxylon* heartwood, Dr Herbert Wong for the NMR data and Dr Dick Girven for the MS data.

REFERENCES

1. Clark-Lewis, J. W. and Mortimer, P. I. (1960) *J. Chem. Soc.* 4106.
2. King, F. E. and Bottomley, W. (1953) *Chem. Ind.* 1368.
3. King, F. E. and Bottomley, W. (1954) *J. Chem. Soc.* 1399.
4. Clark-Lewis, J. W. and Porter, L. J. (1972) *Aust. J. Chem.* **25**, 1943.
5. Malan, E. and Roux, D. G. (1975) *Phytochemistry* **14**, 1835.
6. Fourie, T. G., Du Preez, I. C. and Roux, D. G. (1972) *Phytochemistry* **11**, 1763.
7. Foo, L. Y. and Wong, H. (1986) *Phytochemistry* **25**, 1961.
8. Clark-Lewis, J. W. (1968) *Aust. J. Chem.* **21**, 2059.
9. Pew, J. C. (1948) *J. Am. Chem. Soc.* **70**, 3031.
10. Clark-Lewis, J. W., Jemison, R. W. and Nair, V. (1968) *Aust. J. Chem.* 3015.
11. Clark-Lewis, J. W. and Korytnyk, W. (1958) *J. Chem. Soc.* 2367.
12. Keogh, E. J., Philbin, E. M., Ushioda, S. and Wheeler, T. S. (1963) *J. Chem. Soc.* 3162.
13. Bohm, B. A. (1975) in *The Flavonoids Advances in Research* (Harborne, J. B., Mabry, T. J. and Mabry, H., eds) p. 597. Chapman & Hall, London.
14. Foo, L. Y. (1986) *J. Chem. Soc. Chem. Commun.* 675.
15. Clark-Lewis, J. W., Jackman, L. M. and Spotswood, T. M. (1964) *Aust. J. Chem.* **17**, 632.
16. Haasnoot, C. A. G., de Leeuw, F. A. A. M. and Altona, C. (1980) *Tetrahedron* **36**, 2783.
17. Porter, L. J., Wong, R. Y., Benson, M. and Chan, B. G. (1986) *J. Chem. Res.* 86.
18. Clark-Lewis, J. W. and Nair, V. (1964) *Aust. J. Chem.* **17**, 1164.
19. Candy, H. A., Brookes, K. B., Bull, J. R., McGarry, E. J. and McGarry, J. M. (1978) *Phytochemistry* **17**, 1681.
20. Roux, D. G. and Ferreira, D. (1974) *Phytochemistry* **13**, 2039.
21. Platt, R. V., Opie, C. T. and Haslam, E. (1984) *Phytochemistry* **23**, 2211.
22. Heller, W., Britsch, L., Forkmann, G. and Grisebach, H. (1985) *Planta* **163**, 191.
23. Jacques, D., Opie, C. T., Porter, L. J. and Haslam, E. (1977) *J. Chem. Soc. Perkin Trans.* 1 1637.
24. Hemingway, R. W. and Laks, P. E. (1985) *J. Chem. Soc. Chem. Commun.* 746.
25. Stafford, H. A. (1983) *Phytochemistry* **22**, 2643.
26. Foo, L. Y. (1986) *J. Chem. Soc. Chem. Commun.* 236.
27. Roitman, J. N. and James, L. F. (1985) *Phytochemistry* **24**, 835.
28. Clark-Lewis, J. W. (1968) *Aust. J. Chem.* **21**, 3025.
29. Porter, L. J. and Markham, K. R. (1972) *Phytochemistry* **11**, 1477.