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Structure and stereochemistry of dimeric proteracacinidins possessing the rare C-4(C) → C-5(D) interflavanyl linkage☆

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Abstract

The rare series of (4→5)-linked proteracacinidins is extended by identification of oritin-(4α→5)-epioritin-4β-ol, *ent*-epioritin-(4α→5)-epioritin-4β-ol, epioritin-(4β→5)-epioritin-4α-ol and *ent*-oritin-(4β→5)-epioritin-4α-ol from the heartwoods of *Acacia galpinii* and *Acacia caffra*.

Keywords: *Acacia galpinii*; *Acacia caffra*; Leguminosae; Biflavanoids; Proteracacinidins; Absolute configuration

1. Introduction

Proteracacinidins with their 4′,7,8-trihydroxy phenolic functionality represent a relatively rare group of proanthocyanidins. Their natural occurrence is hitherto confined to the heartwoods of *Acacia galpinii* (Malan and Sireeparsad, 1995; Coetzee et al., 1998a,b; Bennie et al., 2001a) and *Acacia caffra* (Malan, 1995; Malan et al., 1994; Bennie et al., 2000, 2001a, 2001b). In these sources the leucoteracacinidins i.e. oritin- and epioritin-4-ols as incipient electrophiles for proteracacinidin biosynthesis, co-exist with a variety of monomeric flavonoids as potential nucleophiles (Malan, 1995). These monomers, however, invariably exhibit C-4 oxygenation which reduces the nucleophilicity of their A-rings compared to that of the corresponding functionality in the C-4 deoxy compounds causing alternative and often unexpected centers to participate in interflavanyl bond formation (Foo, 1989). Proanthocyanidins with flavan-3-ol or flavan-3,4-diol terminating units possessing pyrogallol-type A-rings predominantly exhibit (4→6)-interflavanyl linkages (Foo, 1985, 1986; Young et al., 1985). Only one example of a (4→5)-linked analog, *ent*-oritin-(4β→5)-epioritin-4β-ol, from the heartwood of *A. caffra* has so far been identified (Malan, 1995). Here we report the structure elucidation of four new (4→5)-coupled proteracacinidins **1**, **5** and **7** from the heartwood of *A. galpinii* and **3** from the heartwood of *A. caffra,* which were identified as the permethylaryl ether acetate derivatives **2**, **6**, **8** and **4**, respectively.

2. Results and discussion

The methanol extracts of the heartwoods of *A. galpinii* and *A. caffra* contain complex mixtures of mono-, di- and trimeric pro-/leucoanthocyanidins (Bennie et al., 2001a, and references cited therein). These compounds are accompanied by four new (4→5)-linked proteracacinidins, oritin-(4α→5)-epioritin-4β-ol **1**, epioritin-(4β→5)-epioritin-4α-ol **5**, *ent*-oritin-(4β→5)-epioritin-4α-ol **7** in *A. galpinii* and *ent*-epioritin-(4α→5)-epioritin-4β-ol **3**, from *A. caffra*. Despite extensive efforts to resolve the free phenolic mixture by partition and gel separation techniques, the dimers could only be purified as their permethylaryl ether acetate derivatives **2**, **6**, **8** and **4**, respectively. Derivatization also provided useful ^{1}H NMR reference signals facilitating unequivocal structure elucidation.

☆ Part 35 in the series 'Oligomeric flavanoids'. Part 34 (Bennie et al., 2001).

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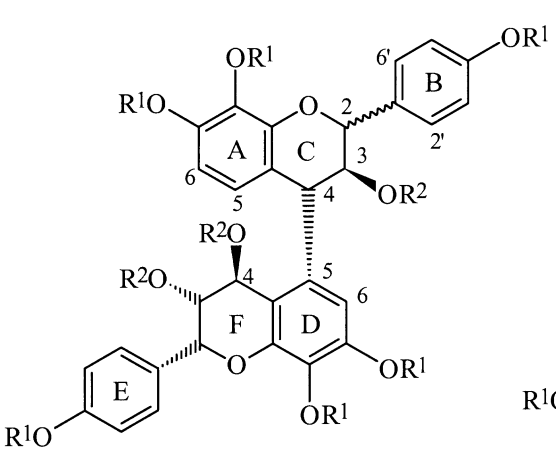

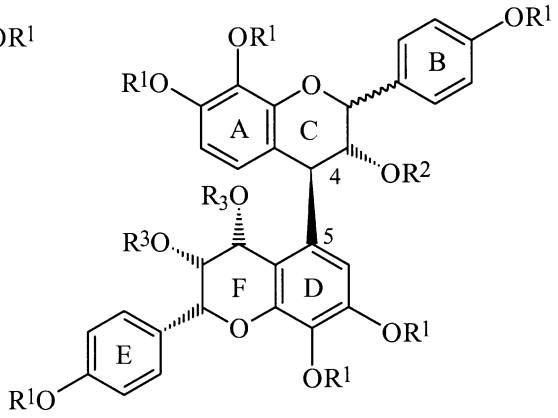

1 ≡ , $R^1=R^2=H$
2 ≡ , $R^1=Me$, $R^2=Ac$
3 ≡ , $R^1=R^2=H$
4 ≡ , $R^1=Me$, $R^2=Ac$

5 ≡ , $R^1=R^2=R^3=H$
6 ≡ , $R^1=R^2=Me$, $R^3=Ac$
7 ≡ , $R^1=R^2=R^3=H$
8 ≡ , $R^1=Me$, $R^2=R^3=Ac$

The structures and relative configurations of these derivatives were determined by analysis of MS and ^1H and ^{13}C NMR spectroscopic data (Tables 1 and 2). ^1H NMR spectral data are given for both $CDCl_3$ and C_6D_6 as solvents since poor resolution of key areas was often observed in one or the other of these solvents. Broadening of heterocyclic proton resonances with little or no sharpening at elevated temperatures was observed. Absolute stereochemistry was assessed *via* chiroptical data, while ^{13}C resonances were assigned by HMQC and HMBC experiments.

FAB–MS analyses of the permethylaryl ether acetate derivatives **2**, **4** and **8** indicated molecular formulae of $C_{42}H_{44}O_{14}$ (*m*/*z* 772) and $C_{41}H_{44}O_{13}$ (*m*/*z* 744) for the heptamethylether diacetate 6. When taken in conjunction with the number of *O*-methyl and *O*-acetyl resonances in their ^1H NMR spectra (Table 1) these formulas suggested proteracacinidin structures with carbon-carbon bonds connecting the upper oritin- and lower teracacidin-type flavanyl units. The ^1H NMR spectral data for compounds **2**, **4**, **6** and **8** indicated the presence of an AB- and two AA′BB′-spin systems as well as a one-proton singlet for aromatic protons. Protons of the heterocyclic rings resonated as two AMX-spin systems with the conspicuously deshielded 4-H(F) resonances reminiscent of the flavan-3,4-diol-type DEF lower unit (Viviers et al., 1982). Differentiation of the spin systems and the connectivities between aromatic and heterocyclic protons were effected with COSY experiments which indicated $^4J_{HH}$ coupling between the respective 2- and 2′,6′-protons. The aromatic AB-system and one-proton singlet [6-H(D)] were differentiated via the observed $^4J_{HH}$ coupling between 5-H(A) and 4-H(C). NOE associations, observed in a phase sensitive NOESY experiment, of 5-H(A) with both 4-H(C) and 6-H(D) and of 6-H(D) with both 5-H(A) and 7-OMe(D), together with the conspicuous absence of association between the 'residual' D-ring proton and 4-H(F) which characterizes proteracacinidins with (4→6)-interflavanyl linkages (Bennie et al., 2001a,b), suggested (4→5)-bonds for compounds **2**, **4**, **6** and **8**. This was unequivocally confirmed by the three bond correlations between 6-H(D) and 4-C(C) in the HMBC spectra of all four derivatives.

Derivatives **2** and **4** exhibited coupling constants reminiscent of 2,3-*cis*-3,4-*trans* ($^3J_{2,3}=1.5$ Hz for **2** and **4**; $^3J_{3,4}=3.0$, 3.5 Hz for **2** and **4**, respectively) relative configuration (Bennie et al., 2001a) of their F-rings. The same rings in derivatives **6** and **8** revealed coupling constants ($^3J_{2,3}=3.2$, 2.8 Hz and $^3J_{3,4}=4.8$, 5.0 Hz for **6** and **8**, respectively) that were characteristic of 2,3-*cis*-3,4-*cis* relative configuration (Bennie et al., 2001a,b). Such all-*cis* configurations were confirmed by NOE associations between 2- and 4-H which indicated that these protons are cofacial. The relative 2,3-*trans*-3,4-*trans* configurations of the C-rings of compounds **2** and **8** were evident from the ^1H NMR coupling constants ($^3J_{2,3}=^3J_{3,4}=10.0$ Hz for both **2** and **8**). Heterocyclic proton coupling constants for the C-rings of derivative **4** and **6** indicated their 2,3-*cis*-3,4-*trans* ($^3J_{2,3}=1,5$, 2.5 Hz; $^3J_{3,4}=2.0$, 3.5 Hz for **4** and **6**, respectively) relative configurations. These 2,3-*cis*-3,4-*trans* C-ring configurations were confirmed by the NOE association (6.8%) between 6-H(D) and 2-H(C) for both derivatives **4** and **6**. The chemical shifts of the C-2 (C-ring) resonances in the ^{13}C NMR spectra of derivatives **2**, **4**, **6** and **8**

Table 1
^{1}H NMR spectral peaks (δ_H) of compounds **2**, **4**, **6** and **8** at 300 MHz. Splitting patterns and *J*-values (Hz) are given parentheses

Ring	Proton	**2** (C_6D_6)	**2** ($CDCl_3$)	**4** (C_6D_6)	**4** ($CDCl_3$)	**6** (C_6D_6)	**6** ($CDCl_3$)	**8** (C_6D_6)	**8** ($CDCl_3$)
A	5	6.65 (*d*, 8.5)	6.15 (*d*, 8.5)	7.03 (*d*, 9.0)	6.65 (*d*, 9.0)	6.67 (*d*, 9.0)	6.19 (*d*, 9.0)	6.92 (*d*, 9.0)	6.46 (*br.s*)
	6	6.89 (*d*, 8.5)	6.39 (*d*, 8.5)	6.56 (*d*, 9.0)	6.59 (*d*, 9.0)	6.48 (*d*, 9.0)	6.43 (*d*, 9.0)	6.43 (*d*, 9.0)	6.46 (*br.s*)
B	2′, 6′	7.58 (*d*, 9.0)	7.43 (*d*, 90)	7.41 (*d*, 9.0)	7.27 (*d*, 9.0)	7.21 (*d*, 9.0)	7.14 (*d*, 9.0)	7.47 (*d*, 9.0)	7.42 (*d*, 9.0)
	3′, 5′	6.90 (*d*, 9.0)	6.92 (*d*, 9.0)	6.85 (*d*, 9.0)	6.84 (*d*, 9.0)	6.82 (*d*, 9.0)	6.84 (*d*, 9.0)	6.86 (*d*, 9.0)	6.92 (*d*, 9.0)
C	2	5.27 (*d*, 10.0)	5.03 (*d*, 10.0)	5.74 (*br.s*, 1.5)	5.22 (*br.s*, 1.5)	5.68 (*d*, 2.5)	5.64 (*d*, 2.5)	5.25 (*d*, 10.0)	4.96 (*d*, 10.0)
	3	6.39 (*t*, 10.0)	5.77 (*dd*, 10.0, 10.0)	5.80 (*dd*, 1.5, 2.0)	5.16 (*dd*, 1.5, 2.0)	3.74 (*dd*, 2.5, 3.5)	3.71 (*dd*, 2.5, 3.5)	6.22 (*t*, 10.0)	5.53 (*t*, 10.0)
	4	5.09 (*d*, 10.0)	4.50 (broadened)	5.13 (*d*, 2.0)	4.48 (*d*, 2.0)	4.49 (*d*, 3.5)	4.97 (*d*, 3.5)	4.89 (*d*, 10.0)	4.24 (*d*, 10.0)
D	6	6.41 (*s*)	6.46 (*s*)	6.62 (*br.s*)	6.16 (*br.s*)	7.38 (*br.s*)	7.06 (*br.s*)	6.80 (*br.s*)	6.36 (*br.s*)
E	2′, 6′	7.66 (*d*, 9.0)	7.43 (*d*, 9.0)	7.65 (*d*, 9.0)	7.42 (*d*, 9.0)	7.42 (*d*, 9.0)	7.28 (*d*, 9.0)	7.49 (*d*, 9.0)	7.37 (*d*, 9.0)
	3′, 5′	6.98 (*d*, 9.0)	6.95 (*d*, 9.0)	6.96 (*d*, 9.0)	6.93 (*d*, 9.0)	6.83 (*d*, 9.0)	6.85 (*d*, 9.0)	6.89 (*d*, 9.0)	6.91 (*d*, 9.0)
F	2	5.68 (*br.s*, 1.5)	5.36 (*br.s*, 1.5)	5.64 (*br.s*, 1.5)	5.45 (*br.s*, 1.5)	5.04 (*d*, 3.2)	5.33 (*d*, 3.2)	5.18 (*d*, 2.8)	5.29 (*d*, 2.8)
	3	5.95 (*dd*, 1.5, 3.0)	5.32 (*dd*, 1.5, 3.0)	6.06 (*dd*, 1.5, 3.5)	5.44 (*dd*, 1.5, 3.5)	5.93 (*dd*, 3.2, 4.8)	5.61 (*dd*, 3.2, 4.8)	6.08 (*dd*, 2.8, 5.0)	6.68 (*dd*, 2.8, 5.0)
	4	6.82 (*d*, 3.0)	6.13 (*d*, 3.0)	6.99 (*d*, 3.5)	6.29 (*d*, 3.5)	6.54 (*d*, 4.8)	6.10 (*d*, 4.8)	7.14 (*d*, 5.0)	6.54 (*d*, 5.0)
OMe		4.13, 3.96, 3.54, 3.41, 3.31, 3.29 (each *s*)	3.93, 3.87, 3.85, 3.84, 3.83, 3.79 (each *s*)	4.15, 3.98, 3.52, 3.39, 3.34, 3.26 (each *s*)	4.00, 3.94, 3.91, 3.84, 3.80, 3.70 (each *s*)	3.14, 3.37, 3.44, 3.52, 3.53, 4.05, 4.08 (each *s*)	3.47, 3.80 (×2), 3.81, 3.85, 3.90, 3.95 (each *s*)	3.33, 3.37, 3.40, 3.47, 3.97, 3.99 (each *s*)	3.77, 3.83, 3.84, 3.85, 3.88, 3.94 (each *s*)
OAc		1.97, 1.62, 1.39 (each *s*)	2.25, 1.94, 1.64 (*s*) (each *s*)	1.84, 1.83, 1.76 (each *s*)	2.16, 1.92, 1.82 (each *s*)	1.37, 1.66 (each *s*)	1.15, 1.96 (each *s*)	1.60, 1.73, 1.80 (each *s*)	1.58, 1.67, 2.07 (each *s*)

Table 2
^{13}C NMR spectral peaks (δ_C) for compounds **2**, **4**, **6** and **8**

Ring	Carbon	**2**	**4**	**6**	**8**
A	5	123.7	125.4	124.1	124.9
	6	106.8	107.1	105.5	106.4
B	2′, 6′	128.5	128.5	128.5	128.5
	3′, 5′	114.2	114.1	113.8	114.0
C	2	81.3	74.7	77.7	81.5
	3	72.9	73.5	80.7	74.5
	4	44.4	42.9	36.9	45.6
D	5	136.3	140.0	136.8	136.8
	6	106.3	108.2	111.0	107.2
E	2′, 6′	129.7	129.1	130.1	129.6
	3′, 5′	114.4	114.4	115.1	114.2
F	2	74.2	74.2	75.8	76.0
	3	68.9	67.8	68.7	68.8
	4	64.5	64.1	65.6	65.2

(Table 2) fully supported these relative configurations. Those compounds with a 2,4-*trans* configuration (**4** and **6**) displayed shielded 2-C(C) signals (ca. 4–7 ppm) compared to the chemical shifts of these carbons in derivatives with a 2,4-*cis* configuration due to the γ-gauche effect (Fletcher et al., 1977). A less prominent NOE association (3.2%) between 4-H(C) and 2′,6′-H(B) indicated a significant contribution of the A-conformation to the C-ring conformational itinerary (Porter et al., 1986; Steynberg et al., 1991) which may also explain the relatively large $^3J_{3,4}$ values via an increase in the relevant dihedral angle of 3- and 4-H(C). Alternatively, these relatively large coupling constants may be attributable to a distorted C-ring conformation, probably a sofa instead of the more familiar half-chair.

The ^{1}H NMR spectrum of derivative **6** indicated only two acetoxy signals, seven *O*-methyl resonances including a shielded signal (δ 3.47 in $CDCl_3$) reminiscent of the chemical shift of a non-phenolic *O*-methyl group. When taken in conjunction with the chemical shift of 3-H(C) (δ 3.74 vs 6.22 for **8**, both in C_6D_6) and the observed coupling between this proton and the shielded *O*-methyl proton in a COSY experiment, these data collectively indicated that 3-OH(C) had been methylated during the derivatization with diazomethane (Botha et al., 1981).

Derivatives **2** and **4** displayed high-amplitude negative Cotton effects ($[\theta]_{247.2} = -1.54\times10^4$, $[\theta]_{245.5} = -1.94\times10^4$, respectively) near 240 nm in their CD spectra. This indicate a 4α-orientation of the DEF-flavanyl unit at C-4 (C-ring), and in conjunction with ^{1}H NMR coupling constants of the protons of this ring, defined 2*R*,3*S*,4*S* and 2*S*,3*S*,4*S* absolute configuration for **2** and **4**, respectively. Positive high-amplitude Cotton effects in the CD spectra of derivatives **6** and **8** ($[\theta]_{240.6} = 9.08\times10^3$, $[\theta]_{244.4} = 2.47\times10^4$, respectively) similarly confirmed a 4β (C-ring) DEF- flavanyl moiety and hence 2*R*,3*R*,4*R* and 2*S*,3*R*,4*R* absolute stereochemistry for the stereocenters of these rings in **6** and **8**, respectively. Both epioritin-4α-

ol and epioritin-4β-ol, as the likely biogenetic precursors to the DEF flavanyl moieties, occur abundantly in both *A. galpinii* and *A. caffra* (Malan, 1995). Hence, these units possess 2*R*,3*R*,4*S* and 2*R*,3*R*,4*R* absolute configuration in **2** and **4**, and **6** and **8**, respectively.

We were intrigued by the fact that moderate temperature increases during accumulation of ^{1}H NMR spectral data did not lead to significant sharpening of the broadened resonances in especially the heterocyclic region. Inspection of Dreiding models indicated severe restrictions to rotation about the interflavanyl bond via steric interaction between 6-H(D) and 4-OAc(F) of the lower unit and 5-H(A) and 3-OAc(C) of the upper moiety. This would lead to a fairly rigid interflavanyl conformation in which the process of rotation is being replaced by a librating action. Such a notion is supported by the fact that the characteristic duplication of signals reminiscent of interchange between the preferred and a non-preferred interflavanyl bond conformation (Steynberg et al., 1995) was hardly discernable in the ^{1}H NMR spectra.

In compound **2** NOE association of 6-H(D) with 3- and 4-H(C) but not with 5-H(A), and association of 4-H(F) with 3-H(C) but not with 5-H(A) and 4-H(C) indicated a preferred conformation where the DEF-unit is approximately perpendicular to the plane of the ABC-moiety and the E/F-ring portion folding backwards underneath the top unit [see Steynberg et al., 1995, for presentations of the more crowded (compressed) and less crowded conformations]. An orthogonal arrangement is presumably compromised in order to maximize an attracting π-alkyl interaction between the A-ring and the methyl group of 4-OAc(F) (Hunter and Saunders, 1990). This would also explain the selective NOE association of 6-H(D) with 3- and 4-H(C) but not with 5-H(A). Such a preference for the crowded conformation was also observed for (4→8)-linked profisetinidin derivatives (Steynberg et al., 1995) and procyanidin diastereomers and presumably results from a tendency to minimize the surface area of the molecule, and hence solute-solvent contact (Foo and Porter, 1983).

NOE associations of 6-H(D) with 2- and 3-H(C) but not with 4-H(C) and 5-H(A), and of 4-H(F) with both 5-H(A) and 4-H(C) but not with 2- and 3-H(C) were observed for derivative **6**. Derivative **8** similarly showed NOE association 6-H(D) with 3-H(C) but not with 4-H(C) and 5-H(A), and of 4-H(F) with both 4-H(C) and 5-H(A) but not with 3-H(C). These highly selective associations indicated a less crowded but rigid interflavanyl conformation with no ^{1}H NMR evidence of interchange with the more crowded rotamer in both derivatives **6** and **8**.

Identification of compounds **1**, **3**, **5** and **7** extends the rare series of proteracacinidins possessing the C-4(C)→C-5(D) interflavanyl linkage. This is yet another demonstration of the heterogeneity of the interflavanyl bonds among natural sources lacking C-4(C) deoxy flavanoids as powerful nucleophilic terminating units.

3. Experimental

^{1}H NMR spectra were recorded on a Bruker AVANCE DPX 300 spectrometer for solns. as indicated, with Me_4Si as internal standard. FAB mass spectra were recorded on a VG-70E instrument with a VG 11–250J data system and an iontech saddlefield FAB gun. TLC was performed on precoated Merck plastic sheets (silica gel 60 PF_{254}, 0.25 mm) and the plates were sprayed with H_2SO_4–HCHO (40:1; v/v) after development. Preparative plates (PLC) [20×22 cm, Kieselgel PF_{254} (1.0 mm)] were air dried and used without prior activation. Column chromatography was done on Sephadex LH-20 in various columns, solvent systems and flow rates (to be specified in each instance). Methylations were performed with an excess of CH_2N_2 in $MeOH/Et_2O$ over a period of 48 h at −15 °C while acetylations were conducted in Ac_2O-pyridine at ambient temperature. Evaporations were done under reduced pressure at ambient temp. in a rotary evaporator, and freeze drying of aqueous solutions on a Virtis 12 SL freezemobile.

3.1. Isolation of phenolic compounds

The extraction of the heartwoods of *A. caffra* and *A. galpinii* and column separations to give fractions A–UU and A–Z, respectively, were comprehensively described in Parts 28 (Coetzee et al., 1998a) and 32 (Bennie et al., 2000) and need not to be repeated.

3.2. Oritin-(4α→5)-epioritin-4β-ol hexa-O-methylether triacetate 2

Methylation of a portion (100 mg) of fraction M from *A. galpinii* followed by PLC in hexane–benzene–Me_2CO–MeOH (43:42:10:5; v/v) gave six bands at R_f 0.60 (3.0 mg), 0.36 (11.0 mg), 0.31 (21.0 mg), 0.20 (24.0 mg), 0.14 (7.0 mg) and 0.10 (6.0 mg). Acetylation of the R_f 0.31 band followed by PLC in hexane–benzene–Me_2CO–MeOH (47:46:5:2; v/v) afforded two main bands at R_f 0.45 (6.0 mg) and 0.42 (9.0 mg). The latter band yielded compound **2** as a white amorphous solid. (Found: M^+, 772.2732. $C_{42}H_{44}O_{14}$ requires M, 772.2731); δ_H (Table 1); ^{13}C NMR (C_6D_6, 20 °C): δ 20.2, 20.4, 21.3 [3×CH_3COO−], 44.4 [4-C(C)], 54.9, 55.0, 55.8, 56.1, 60.7, 60.8 [6x-OCH_3], 64.5 [4-C(F)], 68.9 [3-C(F)], 72.9 [3-C(C)], 74.2 [2-C(F)], 81.3 [2-C(C)], 106.3 [6-C(D)], 106.8 [6-C(A)], 112.4 [10-C(D), 114.2 (×2) [3′5′-C(B)], 114.4 (×2) [3′,5′-C(E)], 119.5 [10-C(A)], 123.7 [5-C(A)], 128.5 (×2) [2′,6′-C(B)], 129.4 [1′-C(E)], 129.7 (×2) [2′,6′-C(E)], 129.8 [1′-C(B)], 136.3 [5-C(D)], 137.5 [8-C(D)], 138.9 [8-C(A)], 149.2 [9-C(D)], 150.0 [9-C(A)], 153.5 [7-C(A)], 155.5 [7-C(D)], 160.3 [4′-C(E)], 168.6, 168.9, 170.0 [3xCH_3COO−]; CD $[\theta]_{230.8}$ 256, $[\theta]_{236.6}$ 3837, $[\theta]_{239.7}$ 36, $[\theta]_{246.3}$−11080, $[\theta]_{260.0}$ −1536, $[\theta]_{274.0}$ −3971,

and $[\theta]_{288.7}$ 4. The remaining bands contain related pro-/leucoteracacinidins which were reported previously (Coetzee et al., 1998a,b).

3.3. *Ent-epioritin-(4α→5)-epioritin-4β-ol hexa-O-methylether triacetate* **4**

Methylation of a portion (200 mg) of fraction S from *A. caffra* followed by PLC in benzene–Me_2CO–EtOAc (7:2:1; v/v) gave four bands at R_f 0.43 (17.4 mg), 0.34 (13.3 mg), 0.31 (22.5 mg), and 0.21 (21.1 mg). Acetylation of the R_f 0.34 band followed by PLC in benzene–Me_2CO (9:1; v/v) afforded two main bands at R_f 0.55 (3.3 mg) and 0.40 (3.0 mg). The R_f 0.40 band yielded compound **4** as a white amorphous solid. (Found: M^+, 772.2731. $C_{42}H_{44}O_{14}$ requires M, 772.2731); δ_H (Table 1); ^{13}C NMR (C_6D_6, 20 °C): δ 20.4, 20.5, 20.9 [3×CH_3COO-], 42.9 [4-C(C)], 54.8, 54.9, 56.0, 56.1, 60.8, 61.1 [6×-OCH_3], 64.1 [4-C(F)], 67.8 [3-C(F)], 73.5 [3-C(C)], 74.2 [2-C(F)], 74.7 [2-C(C)], 107.1 [6-C(A)], 108.2 [6-C(D)], 111.3 [10-C(D)], 114.1 (×2) [3′,5′-C(B)], 114.4 (×2) [3′,5′-C(E)], 115.4 [10-C(A)], 125.4 [5-C(A)], 128.5 (×2) [2′,6′-C(B)], 129.1 (×2) [2′,6′-C(E)], 129.4 [1′-C(E)], 130.0 [1′-C(B)], 137.8 [8-C(D)], 138.9 [8-C(A)], 140.0 [5-C(D)], 150.3 [9-C(A)], 151.1 [9-C(D)], 153.3 [7-C(A)], 155.0 [7-C(D)], 160.1 [4′-C(E)], 160.3 [4′-C(B)], 168, 169, 170.3 [3×CH_3COO-]; CD $[\theta]_{237.2}$ 65, $[\theta]_{245.5}$ −19400, $[\theta]_{267.1}$ 2, $[\theta]_{273.6}$ 838, $[\theta]_{283.7}$ −1851, $[\theta]_{288.6}$ 15, and $[\theta]_{292.1}$ 872. The remaining bands contain related pro-/leucoanthocyanidins that were reported elsewhere (Bennie et al., 2000, 2001b).

3.4. *Epioritin-(4β→5)-epioritin-4α-ol hepta-O-methylether diacetate* **6**

A portion (80 mg) of fraction Q from *A. galpinii* was methylated and the mixture separated by PLC in hexane–benzene–Me_2CO–MeOH (43:42:10:5; ×2; v/v) to give five bands at R_f 0.70 (2.0 mg), 0.66 (4.0 mg), 0.48 (7.0 mg), 0.38 (16.0 mg) and 0.31 (12.8 mg). Acetylation of the R_f 0.38 band followed by PLC in hexane–benzene–Me_2CO–MeOH (43:42:10:5; v/v) afforded a main band at R_f 0.42 (7.0 mg). This yielded compound 6 as a white amorphous solid. (Found: M^+, 744.2782. $C_{41}H_{44}O_{13}$ requires M, 744.2781); δ_H (Table 1); ^{13}C NMR (C_6D_6, 20 °C): δ 20.4, 20.6 [2×CH_3COO-], 36.9 [4-C(C)], 54.9, 55.1, 55.8, 56.0, 57.4, 60.6, 60.9 [7×−OCH_3], 65.6 [4-C(F)], 68.7 [3-C(F)], 75.8 [2-C(F)], 77.7 [2-C(C)], 80.7 [3-C(C)], 105.5 [6-C(A)], 111.0 [6-C(D)], 114.1 [10-C(D)], 113.8 (×2) [3′,5′-C(B)], 115.1 (×2) [3′,5′-C(E)], 119.0 [10-C(A)], 124.1 [5-C(A)], 128.5 (×2) [2′,6′-C(B)], 129.2 [1′-C(E)], 130.1 (×2) [2′,6′-C(E)], 132.3 [1′-C(B)], 136.8 [5-C(D)], 137.5 [8-C(D)], 138.6 [8-C(A)], 149.0 [9-C(A)], 149.7 [9-C(D)], 153.4 [7-C(A)], 155.2 [7-C(D)], 159.8 [4′-C(E)], 160.4 [4′-C(B)], 169.2, 170.3 [2×CH_3COO-]; CD $[\theta]_{222.9}$ 57, $[\theta]_{229.4}$ 4491, $[\theta]_{234.5}$ 2672, $[\theta]_{240.6}$ 9083, $[\theta]_{246.0}$ 4229, $[\theta]_{2250.3}$ 5861, $[\theta]_{264.4}$ 17, $[\theta]_{284.7}$ −4116 and $[\theta]_{300.7}$ 151. The remaining bands of fraction Q did not show defined compounds on TLC and were therefore not further investigated.

3.5. *Ent-oritin-(4β→5)-epioritin-4α-ol hexa-O-methylether triacetate* **8**

Methylation of a portion (100 mg) of fraction U from *A. galpinii* and PLC separation in benzene-Me_2CO (4:1; ×2; v/v) gave five bands at R_f 0.65 (20.0 mg), 0.42 (8.0 mg), 0.35 (18.0 mg), 0.26 (9.0 mg) and 0.11 (6.0 mg). Acetylation of the R_f 0.65 band followed by PLC in benzene–Me_2CO (9:1; v/v) afforded a main band at R_f 0.51 (17.0 mg). The latter band yielded compound **8** as a white amorphous solid. (Found: M^+, 772.2732. $C_{42}H_{44}O_{14}$ requires M, 772.2731); δ_H (Table 1); ^{13}C NMR (C_6D_6, 20 °C): δ 20.3, 20.4, 20.5 [3×CH_3COO-], 45.6 [4-C(C)], 54.8, 54.9, 55.8, 56.0, 60.8, 60.9 [6×−OCH_3], 65.2 [4-C(F)], 68.8 [3-C(F)], 74.6 [3-C(C)], 76.0 [2-C(F)], 81.5 [2-C(C)], 106.4 [6-C(A)], 107.2 [6-C(D)], 113.9 [10-C(D)], 114.0 (×2) [3′,5′-C(B)], 114.2 (×2) [3′,5′-C(E)], 119.3 [10-C(A)], 124.9 [5-C(A)], 128.5 (×2) [2′,6′-C(B)], 129.1 [1′-C(E)], 129.6 [2′,6′-C(E)], 129.8 [1′-C(B)], 136.8 [5-C(D), 137.5 [8-C(D)], 138.6 [8-C(A)], 149.8 [9-C(D)], 149.9 [9-C(A)], 153.5 [7-C(A)], 155.2 [7-C(D)], 159.9 [4′-C(E)], 160.6 [4′-C(B)], 168.6, 170.0, 170.4 [3×CH_3COO-]; CD $[\theta]_{224.2}$ 64, $[\theta]_{230.5}$ 7593, $[\theta]_{235.6}$ 943, $[\theta]_{244.4}$ 24650, $[\theta]_{262.0}$ 54, $[\theta]_{275.1}$ −6320 and $[\theta]_{308.3}$ −317. The remaining bands of fraction U still comprised complex mixtures and were not further investigated.

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