Medicinal Plants of Southern Africa. Part 1. Dimeric Chalcone-based Pigments from *Brackenridgea zanguebarica*

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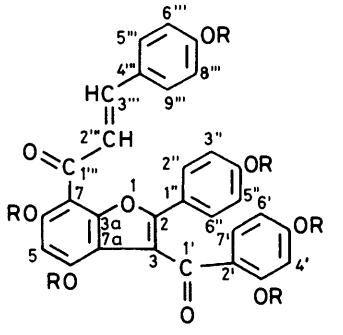
From the bark of *Brackenridgea zanguebarica* four novel biflavonoids have been identified. These include a chalcone-benzofuran dimer, its dihydrobenzofuran derivative, a known dimeric flavanone, and a chalcone-flavanone dimer.

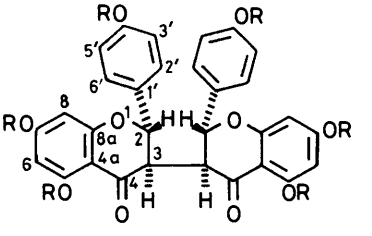
Brackenridgea zanguebarica (Ochnaceae) is a small tree occurring in the Northern Transvaal and Zambia. Various magical powers are ascribed to it by the indigenous Venda people to whom it is known as 'mutavhatsindi'. For example, no adult is allowed to dig up the roots of the tree since this can lead to sterility.[1]

It was the bright yellow colour of the root bark coupled with the peculiar properties ascribed to it, which first aroused our interest. We have now found that the bark contains the α,α′-linked dimeric dihydrochalcone, brackenin,[2] together with several other chalcone-based pigments and these findings have been published in a preliminary communication.[3] The compounds described by us are very different to those isolated from the leaves of the same tree by Bombardelli and co-workers.[4] These authors found vitexin, iso-orientin, and sequojaflavone to be present.

Of the four compounds isolated by us, three are new. They comprise 3-(2,4-dihydroxybenzoyl)-4,6-dihydroxy-2-(4-hydroxyphenyl-7-(4-hydroxycinnamoyl)benzofuran (**1**), commonly referred to as 'orange pigment', its dihydro derivative (**5**), the known[5,6] α,α′-linked biflavanone isochamaejasmin (**6**) and the chalcone-flavanone dimer (**9**). Orange pigment is the major constituent present in the bark and our efforts were concentrated on this pigment.



(**1**) R = H
(**2**) R = H, Dihydrochalcone Derivative
(**3**) R = Ac
(**4**) R = Me
(**5**) R = H 2,3 Dihydro Derivative
(**5'**) R = Me 2,3 Dihydro Derivative



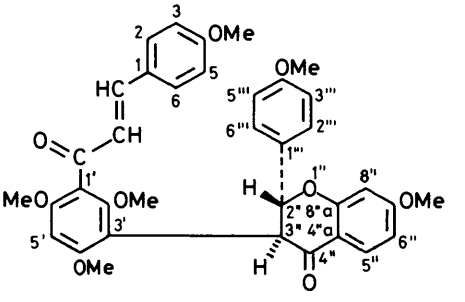
(**6**) R = H
(**7**) R = Ac
(**8**) R = Me

Results and Discussion

Separation of the bark constituents proved to be a formidable problem which was solved by an initial separation by gel chromatography (Sephadex LH 20) followed by flash column chromatography (Kieselgel 60).

Structure Elucidation of the Orange Pigment (**1**).—On two-dimensional paper chromatograms this compound, which had a low R_F in aqueous solvent, possessed a bright orange-yellow colour which deepened on fuming with ammonia, a reaction typical of chalcones. Microscale alkali degradation indicated the presence of 4-hydroxybenzoic acid, 2,4,-dihydroxybenzoic acid, and traces of resorcinol and phenol.

^{1}H N.m.r. and ^{13}C n.m.r. spectra of the deep red rosettes proved very helpful. ^{1}H N.m.r. spectra at 80 and 300 MHz (Figure) established the following. (i) The presence of six phenolic protons of which two are strongly hydrogen-bonded (δ 12.6—14.3). (ii) The presence of the partial structures (**10**), (**11**), (**12**), and (**13**), giving a total of 14 non-hydroxylic protons. (iii) The prominent doublet of doublets at low field (*J* 15.4 Hz) due to a vinyl AB (chalcone) system with *trans*-configuration. Hydrogenation with PtO_2/H_2 eliminated these protons and gave rise to two CH_2 triplets at δ 3.62 and 3.01, typical of a dihydrochalcone.[7,8]



(**9**)

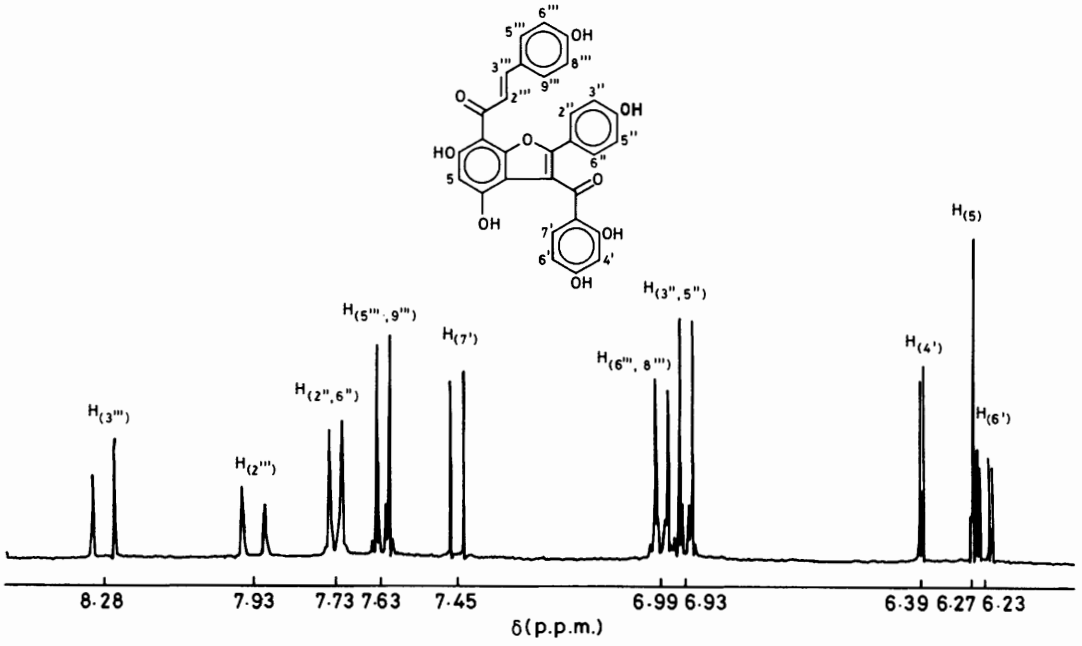
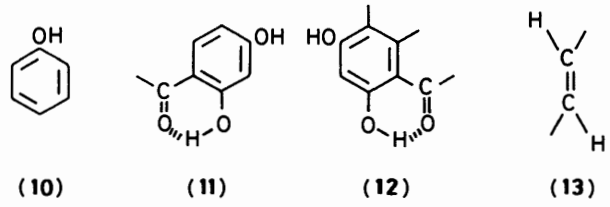


Figure. ^{1}H N.m.r. spectrum of the orange pigment (**1**) at 300 MHz in deuterioacetone



(**10**) (**11**) (**12**) (**13**)

From the above evidence the structure (**1**) was proposed for orange pigment and this was confirmed by additional spectroscopic evidence. The position of attachment of the 4-hydroxycinnamoyl residue could have been at either C-7 or C-5 but the former possibility was chosen by examining the chemical shift position (singlet δ 6.27) of the proton at C-5 and C-7 in a large number of related model compounds. This assignment was confirmed by ^{13}C n.m.r. spectroscopy and other evidence (see later).

The ^{13}C n.m.r. spectrum indicated a total of 26 signals, four of which were clearly of twice the intensity of the others, giving 30 carbons in the molecule. There were no signals below 98 p.p.m. confirming the absence of aliphatic carbons. The off-resonance spectrum indicated 16 singlets and 10 doublets. These 10 methine signals included the four of double intensity. Comparison of the carbon chemical shifts of naringenin,[9] isoliquiritigenin,[10] apigenin,[10] and 2,4,6-trihydroxyacetophenone[11] then furnished structure (**1**) which can be viewed as consisting of two linked chalcones (isoliquiritigenin and chalcononaringenin). Thirteen of the methine carbons readily fell into place but as with the proton spectrum assignment of the remaining resonance (at 99.14 p.p.m.) was less easy. However, the considerable downfield shift this carbon experiences on acetylation of the molecule, together with evidence from ^{13}C n.m.r. data collected by Chari *et al.* on related systems, leads us to believe that the above resonance belongs to C-5 and *not* C-7.

The two quaternary carbons,[12] and the eight oxygen-bearing carbons were readily assigned, thus leaving six singlet carbons unassigned. Observed chemical shifts of related compounds allowed assignment of the six remaining singlets as follows: 127.02 p.p.m. to C-4''' and 101.81 to C7 (on the basis of hydrogenation of the cinnamoyl side chain of orange pigment and observing the changes in chemical shift); 121.01 to C-1'' and 141.70 to C-2'. From literature on benzofuran models the last two singlets were assigned as follows: 112.94 to C-3 and 111.10 to C9. The assignments shown in Table 1 were confirmed by observing ^{13}C-^{1}H couplings in the fully-coupled ^{13}C spectrum of (**1**). Thus C-8 appeared as a sharp singlet at both 20 and 125 MHz. This indicates conclusively that the 4-hydroxycinnamoyl

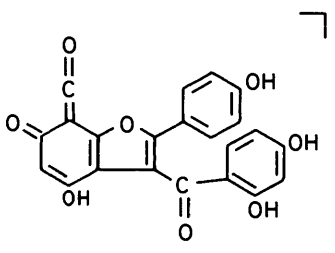
Table 1. Comparison of the ^{13}C n.m.r. chemical shifts (p.p.m.) for compounds (**1**) and (**5**). For the latter, oxygen-bearing carbons are excluded.

Carbon no.	(1)	(5)
2	152.79	90.39
3	112.94	53.99
4	165.85	—
5	99.14	96.48
6	158.96	—
7	101.81	101.86
3a	154.29	—
7a	111.10	104.87
1′	196.15	202.41
2′	114.70	113.07
3′	165.85	—
4′	102.80	102.98
5′	166.93	—
6′	108.56	108.36
7′	136.16	133.62
1″	121.01	131.23
2″,6″	128.64	127.67
3″,5″	116.11	115.91
4″	158.27	—
1‴	189.87	191.02
2‴	122.23	122.70
3‴	144.42	143.77
4‴	127.02	127.03
5‴,9‴	130.89	130.66
6‴,8‴	116.32	116.12
7‴	160.54	—

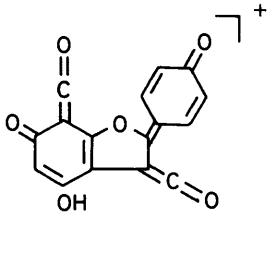
group is attached to the benzofuran at C-7. Attachment at C-5 would leave a proton at C-7, to which the C-8 could then couple. The same technique was used to distinguish C-3 from C-9. At 125 MHz C-3 clearly appears as a singlet, indicating no long range ^{13}C-^{1}H coupling. By contrast C-9 is a doublet indicating a typical 3 bond coupling.

Effect of Derivatisation on ^{13}C N.m.r. Spectrum of the Orange Pigment (**1**).—Studies by Pelter[12] and by Markham[9] have shown that acetylation of phenolic hydroxy groups shifts the signal of the acetylated carbon upfield by 6—16 p.p.m. Calculations based on substituent effects predict that a carbon *ortho* to two hydroxy groups will shift more than one *ortho* to one hydroxy and *para* to another on acetylation. In the case of (**1**) the shift is from 99.14 to 113.8 p.p.m., in keeping with attachment of the coumaroyl substituent to C-7 rather than to C-5.[13]

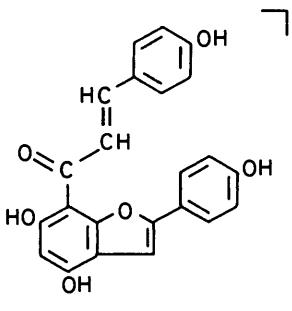
Mass Spectral Analysis of the Orange Pigment (**1**).—In considering the mass spectral fragmentation of (**1**), the earlier studies on chalcones[10,14,15] and benzofurans[14,16,17] proved useful. Under electron impact (e.i.) the molecular ion was observed as a prominent peak at 524 and using fast atom bombardment (f.a.b.) it was at 525. There were two clear fragmentation pathways. (i) This involved isomerisation of the chalcone to the corresponding flavanone followed by loss of the coumaroyl side chain by a retro Diels–Alder reaction to afford an ion at *m/z* 404, proved to be $C_{22}H_{12}O_8$ by high resolution mass spectrometry. For this ion the structure (**14**) is proposed. Typical β-cleavage of (**14**) then gave (**15**), *m/z* 294 $C_{16}H_6O_6$,



(**14**)



(**15**)

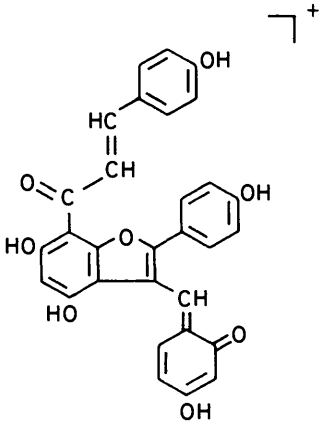


(**16**)

which was also the base peak. (ii) Prominent in this pathway is the ion (**16**) *m/z* 388 resulting from cleavage of the substituent on C-3. The 2,4-dihydroxybenzoyl ion, *m/z* 137, is also present as a major peak.

Structure Elucidation of 2,3-Dihydro Orange Pigment (**5**).—This compound was bright yellow and on paper chromatograms the colour deepened when treated with fuming ammonia. It eluted from the Sephadex column immediately ahead of orange pigment to which it bore a close similarity. The material could not be obtained pure in the free phenolic form, being contaminated with isochamaejasmin (**6**). However, when examined at 300 MHz it was relatively easy to deduce the proposed structure which was confirmed by analysis of the pure hexamethyl ether. In all respects the ^{1}H n.m.r. spectrum followed closely that of orange pigment with the one difference that the protons on C-2 and C-3 of the benzofuran ring stood out clearly as a doublet of doublets at δ 5.34 and 5.82 (*J* 5.7 Hz).

The ^{13}C n.m.r. spectrum of compound (**5**) was also readily analysed by analogy with (**1**). In Table 1 the carbon shifts for these two compounds are compared. The molecular ion of (**5**) was obtained only under f.a.b. conditions. The ion observed at *m/z* 527 analysed for $C_{30}H_{12}O_9H^+$, corresponding to the protonated molecular ion. Under e.i. conditions the highest mass peak was at *m/z* 508. It is suggested that this ion has structure (**17**), a stabilised benzofuran system, resulting from

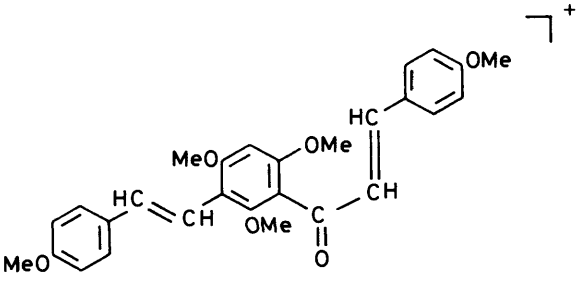


(**17**)

loss of H_2O from the molecular ion *via* enolisation of the benzoyl group at C-3. Other fragments were analogous to those found in (**1**).

Structure Elucidation of the Chalcone-Flavanone Dimer (**9**).—Of the three pigments isolated this is the only conventional 'biflavonoid'. The compound was obtained pure as its hexamethyl ether derivative. When recorded at 25 °C in $CDCl_3$ the ^{1}H n.m.r. spectrum was poorly resolved, even at 500 MHz. Increasing the temperature to 47 °C improved the appearance of the spectrum dramatically. Under these conditions a full assignment of peaks was readily made (see Experimental section). The chalcone vinyl protons appeared as a doublet of doublets (δ 7.04 and 6.74), each having a characteristic *trans*-coupling of 15.9 Hz. The other doublets were at δ 5.88 and 4.64 (*J* 12.1 Hz) indicating a *trans* diaxial coupling typical of dihydroflavonols.[10]

The mass spectrum of compound (**9**) contained fragments of both chalcone and flavanone moieties, as expected. An intense molecular ion occurred at *m/z* 610 ($C_{36}H_{34}O_9$) and the base peak was a t *m/z* 460. The latter forms by a retro Diels–Alder fragmentation and probably has the structure (**18**).



(**18**)

Structure Determination of 3,3″-*Linked Naringenin Dimer* (*Isochamaejasmin*) (**6**).—This compound was not obtained pure in the free phenolic form but was contaminated with dihydro orange pigment (**5**). None the less, its n.m.r. spectrum was relatively simple to analyse. Our n.m.r. data (in deuterioacetone) agree closely with values published by Chen and his co-workers[6] for isochamaejasmin. Compound (**6**) was obtained pure as its hexamethyl ether (Chen[6] quotes spectral data for the tetramethyl ether) and also as its hexa-acetate derivative. Isochamaejasmin and several related isomers have previously been isolated from *Thymeliacea* spp. by Chang[5] and Chen.[6]

The mass spectrum of compound (**6**) produced a molecular ion under f.a.b. conditions only. Under e.i. conditions easily recognisable fragments occurred at *m/z* 390 (retro Diels–Alder), at *m/z*), at *m/z* 271 (flavanone monomer), and at *m/z* 153, 152, 120, and 121 (further retro Diels–Alder fragments).

Possible Structure Relationships.—The five biflavonoids found in *Brackenridgea zanguebarica*, while showing diversity, also have certain distinct features in common. The linkage through C-3 to another molecule is common to all of them; the hydroxylation pattern is also similar in all five compounds. Biosynthetic considerations have previously indicated the central role of the chalcone in the biosynthesis of flavonoid monomers but their role in biflavonoid compounds is less clear.[18] The compounds isolated in the present study provide for the first time C–C linked biflavonoids bearing a chalcone moiety. It can be speculated that the dimers of this series arise from initial chalcone dimerisation followed by further elaboration.

Experimental

M.p.s were determined on a Kofler micro melting point apparatus and are uncorrected. I.r. spectra were recorded on a Perkin-Elmer 283 spectrophotometer as Nujol mulls or in dichloromethane solvent. N.m.r. spectra were recorded on the following instruments: Varian FT 80A, Varian XL 300, Bruker WM 360 and Bruker WM 500. The solvents used are indicated in the text. Mass spectra were run on the following: Varian CH 7, Varian MAT 212 and Varian MAT 212 equipped with fast atom bombardment (f.a.b.) ionisation source.

Ethanolic extracts of the bark of the tree were examined by two dimensional chromatography using a solvent system developed by Mabry.[19] Spraying with ammoniacal silver nitrate showed the presence of about 20 compounds. When the extract was examined in 'conventional' solvent systems such as butanol–acetic acid–water and 2% acetic acid, no separation was possible and the compounds remained virtually unseparated at the solvent front.

Extraction of the finely ground corky yellow bark (400 g), firstly with light petroleum, chloroform, and finally benzene afforded a total of 18 g of waxy material. Subsequent extraction of the residue (Soxhlet) with ethanol (95%) then gave a yellow brown powder (181 g).

Separation on Sephadex LH 20.—Sephadex LH 20 gel (100 g) was allowed to swell for 12 h in aqueous ethanol (47%) and a column (4 cm × 34 cm) packed in the normal way. A flow rate of 2.8 ml/min was maintained by peristaltic pump. In a typical run phenolic material (8 g) in the minimum of eluant was applied to the column and 15 ml fractions were then collected. The eluant was monitored by means of a flow-through cell and a u.v. detector set at 280 nm. This gave five pooled fractions as follows: tubes 1—100, oily fraction (0.3 g); 101—290, fraction 1 (1.8 g); 291—450 fraction 2 (1.2 g); 451—600, fraction 3 (1.4 g; 601—770, fraction 4 (1.8 g); 771—980, fraction 5 (0.7 g). Each fraction (2 g) was purified further by flash column (2 × 15 cm) chromatography using silica gel (Kieselgel 60) and benzene ethanol (9:1) as eluant.

3-(2,4-*Dihydroxybenzoyl*)-2-(4-*hydroxyphenyl*)-7-(4-*hydroxycinnamoyl*)-4,6-*dihydroxybenzofuran* (**1**), *Orange pigment.*—This compound was present in fraction 5, and after final separation on silica gel gave orange red rosettes (300 mg), m.p. 252—253 °C (acetone–light petroleum) (Found: M^+, 524.10794 $C_{30}H_{20}O_9$ requires M^+, 524.11069); $\nu_{max.}$(Nujol) 3 520—3 320, 1 622, 1 600, 1 545, and 1 509 cm^{-1}; δ[$(CD_3)_2CO$, 300 MHz] 6.24 (1 H, dd, *J* 2.3, 8.71 Hz, 6′-H), 6.27 (1 H, s, 5-H), 6.39 (1 H, d, *J* 2.3 Hz, 3′-H), 6.93 (2 H, d, *J* 8.8 Hz, 3″,5″-H), 6.99 (2 H, d, *J* 8.5 Hz, 6‴,8‴-H), 7.46 (1 H, d, *J* 8.8 Hz, 7′-H), 7.63 (2 H, d, *J* 8.5 Hz, 5‴,9‴-H), 7.74 (2 H, d, *J* 8.5 Hz, 2″,6″-H), 7.94 (1 H, d, *J* 15.4 Hz, 2‴-H), 8.28 (1 H, d, *J* 15.4 Hz, 3‴-H), 9.0 (1 H, br s, OH), 9.2 (1 H, br s, OH), 9.8 (1 H, br s, OH), 10.2 (1 H, br s, OH), 12.4 (1 H, s, 6-OH), and 14.3 (1 H, s, 3′-OH); *m/z* (e.i.) 524 (M^+, 2.4%), 404 (1.3), 388 (0.6), 294 (6.3), 284 (1.3), 268 (1.8), 266 (2.2), 256 (0.2), 242 (0.5), 240 (0.3), 149 (2.4), 142 (1.6), 137 (4.1), 121 (3.3), 110 (29.7), and 94 (100) (Found: 404.05845, 294.01286. $C_{22}H_{12}O_8$, $C_{16}H_6O_6$ require 404.05318, 294.01641 (respectively; ^{13}C n.m.r spectra data are reported in the Table.

Orange Pigment Dihydrochalcone (**2**).—This compound was prepared from (**1**) by catalytic hydrogenation PtO_2/H_2 and gave yellow needles, m.p. 250—251 °C (acetone–light petroleum) (Found: M^+, 526.12287. $C_{30}H_{22}O_9$ requires *M*, 526.12634; δ_H[$(CD_3)_2CO$, 80 MHz] 3.01 (2 H, t, *J* 7.0 Hz, CH_2CH_2), 3.62 (2 H, t, *J* 7 Hz, $COCH_2$), 6.25 (1 H, s, 5-H), 6.44 (1 H, d, *J* 2.2 Hz, 6′-H), 6.80 (2 H, d, *J* 8.5 Hz, 6‴,8‴-H), 6.83 (2 H, d, *J* 8.8 Hz, 3″,5″-H), 7.18 (2 H, d, *J* 8.4 Hz, 5‴,9‴-H), 7.45 (1 H, d, *J* 8.7 Hz, 7′-H), 7.49 (2 H, d, *J* 8.7 Hz, 2″6″-H), 12.43 (1 H, s, 6-OH), and 14.32 (1 H, s, 3′-OH); δ_C(20 MHz) 29.2 (C-3‴), 44.3 (C-2‴), 98.8 (C-5), 101.0 (C-7), 102.7 (C-4′), 108.6 (C-6′), 110.8 (C-9), 112.6 (C-3), 114.4 (C-2′), 115.3 (C-6‴,8‴), 115.8 (C-3″,5″), 120.6 (C-1″), 128.55 (C-2″,6″), 129.4 (C-5‴,9‴), 132.0 (C-4‴), 136.0‴ (C-7′), 152.8 (C-2), 154.4 (C-8), 155.6 (7‴), 158.6 (C-4″), 158.6 (C-6), 165.4 (C-3′), 165.7 (C-4), 165.8 (C-5′), 196.1 (C-1′), and 202.1 (C-1‴).

Orange Pigment Hexa-acetate (**3**).—Partially purified compound (**1**) (100 mg) on treatment with acetic anhydride–pyridine for 8 h at 25 °C, followed by separation on silica gel using benzene–ethyl acetate (8:2) afforded yellow needles (62 mg, methanol), m.p. 108—109 °C (Found: C, 65.3; H, 4.3; $C_{42}H_{32}O_{15}$ requires C, 65.0, H, 4.1%); δ_H[$(CD_3)_2CO$, 80 MHz] 2.22 (3 H, s, $COCH_3$), 2.23 (3 H, s, $COCH_3$), 2.26 (6 H, s, 2 × $COCH_3$), 2.36 (6 H, s, 2 × $COCH_3$), and 8.01—7.93 (14 H, m, ArH, vinyl H) (Found: M^+ − 42, 734.16317. $C_{40}H_{30}O_{14}$ requires 734.16349, 1.4%); *m/z* 692 (4.5), 650 (7.2), 608 (8.2), 566 (1.4), 524 (0.4), 446 (4.6), 387 (5.1), 294 (11.1), 268 (3.4), 146 (10.6), 137 (12.1), and 121 (9.1).

Orange Pigment Hexamethyl Ether (**4**).—Methylation of compound (**1**) (100 mg) with dimethyl sulphate–potassium carbonate in acetone gave yellow crystals (51 mg), m.p. 109—111 °C (Found: M^+, 608.20228. $C_{36}H_{32}O_9$ requires 608.20459); δ_H($CDCl_3$, 80 MHz) 3.64 (3 H, s, OCH_3), 3.67 (3 H, s, OCH_3), 3.72 (3 H, s, OCH_3), 3.78 (6 H, s, 2 × OCH_3), 3.88 (3 H, s, OCH_3), 6.36—6.48 (3 H, m, 5,4′,6′-H), 6.72—6.93 (4 H, m, 3″,5″,6‴,8‴-H), 7.26 (1 H, d, *J* 15.9 Hz, 2‴-H), and 7.47—7.76 (6 H, m, 2″,6″,3‴,5‴,9‴,7′-H); *m/z* (e.i.) 608 (M^+, 100%), 591 (4), 590 (4), 588 (5), 563 (7), 476 (3.5), 477 (3), 458 (15.5), 449 (8.6), 430 (3), 312 (6.2), 304 (8), 297 (5.1), 267 (3.4), 237 (3), 166 (29), 165 (100), 150 (7), and 121 (33).

trans-2,3-*Dihydro Orange Pigment* (**5**).—This yellow pigment was present in fraction 4 off the Sephadex column but

Table 2. ^{1}H N.m.r. chemical shifts (p.p.m.) for isochamaejasmin (**6**), its hexa-acetate- (7), and hexamethyl ether derivative (**8**)

Proton(s)	(**6**)[$(CD_3)_2CO$]	(7)[$CDCl_3$]	(**8**)[CD_2Cl_2]
3	3.80	3.77	3.73—3.86
2	4.90	5.08	5.10
8	5.82	6.54	5.95
6	5.96	6.62	6.04
3′,5′	6.88	7.16	6.88
2′,6′	7.13	7.16	7.13
OH	8.75, 12.00		
OMe			3.73, 3.80, 3.86
OAc		2.27, 2.29, 2.38	

was contaminated with several other components. Repeated separation on silica gel finally yielded a fraction (60 mg) which was pure enough for meaningful spectroscopic analysis. The ^{1}H n.m.r. spectrum at 500 MHz readily revealed its structure: $\delta_H[(CD_3)_2CO]$ 5.29 (1 H, d, *J* 5.83 Hz, 3-H), 6.05 (1 H, d, *J* 5.83 Hz, 2-H), 6.36 (1 H, d, *J* 2.35 Hz, 4′-H), 6.38 (1 H, dd, *J* 8.84 Hz, *J* 2.40 Hz, 6′-H), 6.83 (2 H, d, *J* 8.56 Hz, 6‴,8‴-H), 6.89 (2 H, d, *J* 8.56 Hz, 3″,5″-H), 7.36 (2 H, d, *J* 8.57 Hz, 2″,6″-H), 7.47 (2 H, d, *J* 8.62 Hz, 5‴,9‴-H), 7.76 (1 H, d, *J* 8.5 Hz, 7′-H), 7.79 (1 H, d, *J* 15.6 Hz, 2‴-H), 7.99 (1 H, d, *J* 15.6 Hz, 3‴-H), 12.75 (1 H, s, 6-OH), and 14.1 (1 H, s, 3′-OH); δ_C (20 MHz) 53.99 (C-3), 90.39 (C-2), 96.48 (C-5), 101.86 (C-7), 102.98 (C-4′), 104.87 (C-9), 108.36 (C-6′), 113.07 (C-2′), 115.91 (C-3″,5″), 116.12 (C-6‴,8‴), 122.70(C-2″,7″), 127.03(C-4‴), 127.67(C-2″,6″), 130.66(C-5‴,9‴), 131.23 (C-1″), 133.62 (C-7′), 143.77 (C-3‴), 158.17, 160.14 (C-7‴), 160.75 (C-4‴), 163.41, 165.64, 166.32, 167.70, 191.02 (C-1‴), and 202.41 (C-1″); *m/z* (f.a.b.) 527 (MH^+, 9%), 508.11767 ($M^+ - 18$, $C_{30}H_{20}O_8$ requires 508.11578.

trans-2,3-*Dihydro Orange Pigment, Hexamethyl Ether* (**5′**).—Chromatography of the crude methylated material (potassium carbonate–dimethyl sulphate) over silica gel (benzene–ethyl acetate, 8:1) afforded the hexamethyl ether as a yellow solid, m.p. 66 °C (methanol) (Found: M^+, 610.22045. $C_{36}H_{34}O_9$ requires 610.22024); δ_H($CDCl_3$, 250 MHz) 5.34 (1 H, d, *J* 5.4 Hz, 3-H), 5.82 (1 H, d, *J* 5.3 Hz, 2-H), 6.05 (1 H, s, 5-H), 6.35 (1 H, d, *J* 2.2 Hz, 4-H), 6.54 (1 H, dd, *J* 2.2 Hz, 8.7 Hz, 6′-H), 6.84 (2 H, d, *J* 8.7 Hz, 3″,5″-H), 6.85 (2 H, d, *J* 8.7 Hz, 6‴,8‴-H), 7.01 (1 H, d, *J* 15.8 Hz, 2‴-H), 7.26 (2 H, d, *J* 8.7 Hz, 2″,6″-H), 7.42 (2 H, d, *J* 8.9 Hz, 5‴,7‴-H), 7.50 (1 H, d, *J* 15.9 Hz, 3‴-H), and 7.72 (1 H, d, *J* 8.7 Hz, 7′-H).

Isochamaejasmin (**6**).—This dimer was isolated from fraction 4 together with the orange pigment. In view of the limited information available on the compound the spectral characteristics of the free phenolic form,[5,6] its hexamethyl ether, m.p. 86 °C and its hexa-acetate derivative, m.p. 138 °C are listed in Table 2. For the latter derivative found M^+, 692.149416. $C_{38}H_{25}O_{13}$ requires 692.15293.

Chalcone-Flavanone Dimer, Hexamethyl Ether (**9**).—This compound (30 mg) was obtained from fraction 4 after methylation (potassium carbonate–dimethyl sulphate) and separation on silica gel, m.p. 73 °C (Found: M^+, 610.22045. $C_{36}H_{34}O_9$ requires 610.220024); δ_H($CDCl_3$, 250 MHz) 3.71 (9 H, s, 3 × OCH_3), 3.83 (9 H, s, 3 × OCH_3), 4.64 (1 H, d, *J* 12.1 Hz, 3″-H), 5.88 (1 H, d, *J* 12.1 Hz, 2″-H), 6.20 (1 H, s, 5′-H), 6.62 (1 H, dd, *J* 2.2, 8.8 Hz, 6″-H), 6.74 (1 H, d, *J* 15.9 Hz, 2-H), 6.79 (2 H, d, *J* 8.6 Hz, 3‴,5‴-H), 6.88 (2 H, d, *J* 8.7 Hz, 3,5-H), 7.04 (1 H, d, *J* 15.9 Hz, β-H), 7.27 (2 H, d, *J* 8.6 Hz, 2‴,6‴-H), 7.40 (2 H, d, *J* 8.4 Hz, 2,6-H), and 7.92 (1 H, d, *J* 8.8 Hz, 5″-H); δ_C ($CDCl_3$, 20 MHz) 50.46 (C-3″), 55.12 (OCH_3), 55.31 (2 × OCH_3), 55.53 (OCH_3), 55.84 (2 × OCH_3), 82.70 (C-2″), 91.6 (C-5′), 100.91 (C-8″), 110.00 (C-1′), 110.00 (C-6″), 114.29 (C-3,5), 114.37 (C-3‴,5‴), 114.37 (C-10″), 114.79 (C-3′), 126.57 (C-α), 127.33 (C-1), 128.61 (C-2‴,6‴), 129.14 (C-5″), 130.22 (C-2,6), 130.57 (C-1‴), 145.19 (C-β), 157.85 (C-2′), 157.85 (C-6′), 159.59 (C-4′), 159.59 (C-4‴), 161.59 (C-4), 163.53 (C-9″), 165.76 (C-7″), 191.81 (C-4″), and 194.34 (chalcone CO); *m/z* (e.i.) 610 (M^+, 81.2%), 594 (12.8), 578 (61.6), 489 (9.6), 460 (100), 445 (14.3), 432 (28.7), 339 (34.4), 327 (14.9), 313 (24.6), 297 (17.6), 283 (19.3), 165 (34.1), 161 (74.6), 151 (42.7), and 121 (87.2).

Hydrogenation of the above hexamethyl ether (PtO_2–H_2) afforded a pale yellow solid, m.p. 66 °C. Its n.m.r. spectrum clearly reflected loss of the double chalcone double bond; δ_H($CDCl_3$, 80 MHz) 2.85 (2 H, m, β-CH_2), 3.65—3.82 (2 H, m, α-CH_2), 370 (6 Hz, s, 2 × OCH_3), 3.76 (6 H, s, 2 × OCH_3), 3.82 (6 H, s, 2 × OCH_3), 4.57 (1 H, d, *J* 12.4 Hz, 3‴-H), 5.86 (1 H, d, *J* 12.4 Hz, 2″-H), 6.50 (1 H, d, *J* 2.3 Hz, 8″-H), 6.62 (1 H, dd, *J* 2.3, 8.6 Hz, 6″-H), 6.73 (2 H, d, *J* 8.8 Hz, 3,5-H), 6.77 (2 H, d, *J* 8.8 Hz, 3‴,5‴-H), 7.06 (2 H, d, *J* 8.7 Hz, 2,6-H), 7.22 (2 H, d, *J* 8.7 Hz, 2‴,6‴-H), and 7.91 (1 H, d, *J* 8.5 Hz, 5″-H).

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