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LACTONES, FLAVONOIDS AND BENZOPHENONES FROM *GARCINIA CONRAUANA* AND *GARCINIA MANNII*

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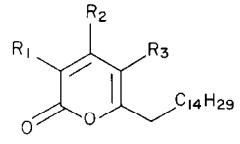
Key Word Index—*Garcinia conrauana*; *G. mannii*; Guttiferae; lactones; 3-(3″,3″-dimethylallyl)-conrauanalactone; 3-α-hydroxy-5-(heptadec-8′-enyl)-tetrahydrofuran-2-one; biflavonoids.

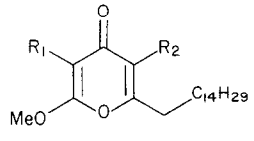
Abstract—Two novel lactones have been isolated from the stem barks of *Garcinia conrauana* and *G. mannii*. The major component of the bark of *G. conrauana* was identified as 3-(3″, 3″-dimethylallyl)-conrauanalactone [4-hydroxy-3-(3″, 3″-dimethylallyl)-6-pentadecylpyran-2-one] by comparison of spectral data of the isolated compound and two methyl ethers with that obtained for the previously isolated conrauanalactone. A minor component of the bark of *G. mannii* was tentatively identified as 3-α-hydroxy-5-(heptadec-8′-enyl)-tetrahydrofuran-2-one on the basis of spectral data from the isolated compound and its monoacetate. The distributions of biflavonoids and related compounds and benzophenones in the stem bark, heartwood, seeds and leaves of the two species are reported.

INTRODUCTION

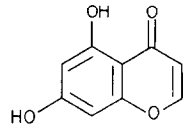
The genus *Garcinia* L. (Guttiferae) is well known as a source of xanthones, biflavonoids and benzophenones [1–3]. Recent studies on the stem bark of *G. mannii* Oliv. [4], *G. ovalifolia* Oliv. [5] and *G. densivenia* Engl. [6], collected in the Douala-Edea Forest Reserve of West Cameroun, have yielded compounds of all three types. A fourth sympatric species, *G. conrauana* Engl. proved atypical [7] by producing large amounts of conrauanalactone (**1**) from the stem bark, the first time a compound of this type has been recorded in the family.



	R_1	R_2	R_3
1	H	OH	H
2	$CH_2CH{=}C(Me)_2$	OH	H
3	H	OH	$CH_2CH{=}C(Me)_2$
6	$CH_2CH{=}C(Me)_2$	OMe	H
7	H	OMe	$CH_2CH{=}C(Me)_2$



4 $R_1 = CH_2CH{=}C(Me)_2$, $R_2 = H$
5 $R_1 = H$, $R_2 = CH_2CH{=}C(Me)_2$



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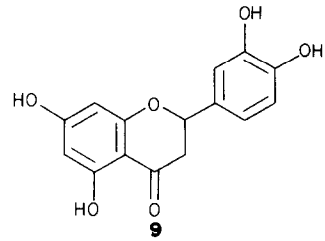

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We now report the occurrence of two further lactones in the stem barks of *G. conrauana* and *G. mannii* from collections made further north in Cameroun, in the Korup Forest Reserve. The distribution of compounds in other parts of these two species, based on Douala-Edea material, is also reported.

RESULTS AND DISCUSSION

Concentration of a petrol extract of the stem bark of *G. conrauana* from Korup gave a single compound (yield 0.8%) which analysed for $C_{25}H_{42}O_3$. The UV spectrum showed a single maximum at 290 nm, typical of an α-pyrone, and bands in the IR indicated hydroxyl and lactonic carbonyl. EIMS revealed three important features: (1) fragments for loss of $C_4H_8^{\cdot}$ and $C_4H_9^{\cdot}$ indicative of a prenyl unit, (2) fragments for loss of $C_{14}H_{28}^{\cdot}$ and $C_{15}H_{31}^{\cdot}$, and (3) ions at *m/z* 139 ($C_7H_7O_3$) and 126 ($C_6H_6O_3$). Features (2) and (3) are identical to those noted previously [7, 8] for the fragmentation of **1**. The ^1H NMR spectrum showed resonances for a pentadecyl side chain identical to **1** [7]. A sharp singlet at δ 6.00 (1H) was typical of H-3 or H-5 of a 4-hydroxy-2-pyrone and signals at δ 1.63 (3H), 1.71 (3H), 3.06 (2H) and 5.28 (1H) indicative of a 3, 3-dimethylyallyl unit. Signals at δ 163.0, 164.8 and

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165.0 in the ^{13}C NMR spectrum confirmed that alternate carbons of the pyrone (C-2, C-4, C-6) are directly attached to oxygen. C-3 and C-5 were represented by a doublet at δ 101.0 and a singlet at 99.32.

The above data suggest that the isolated compound is conrauanalactone substituted with a 3,3-dimethylallyl unit at either C-3 (**2**) or C-5 (**3**). Methylation with CH_2N_2 gave a mixture of two compounds which were separated by prep. TLC. Both were yellow oils and analysed for $C_{26}H_{44}O_3$. Spectral analysis of the two compounds showed them to be almost identical except for: (1) position of UV maximum (261 vs 300 nm) and (2) position of C=O resonance in ^{13}C NMR spectra (181.0 vs 165.0). The reduced chromophore and highly deshielded carbonyl resonance are typical of a 4-pyrone allowing one of the methyl ethers to be characterized as **4** or **5**. The second methyl ether will be the corresponding 2-pyrone **6** or **7**.

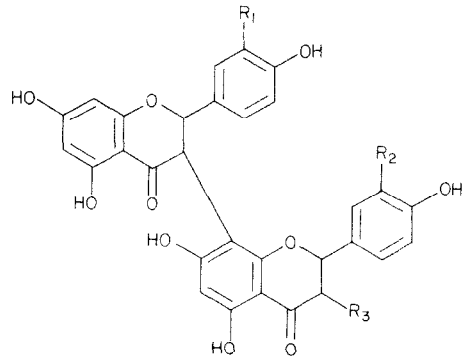
The position of the prenyl side chain was assigned on the basis of a ^1H lanthanide shift experiment on the two methyl ethers using $Eu(fod)_3$. The shifts for pyrone ring proton, H-1″ and H-2″ of the prenyl side chain, H-7′ and the methoxyl group, relative to a shift of 1.0 for the latter, are given in Table 1. As expected, in the 4-pyrone the greatest shift is shown by the pyrone ring proton with a somewhat smaller but still highly significant shift for H-1″ and H-2″. This confirms that these two substituents lie either side of C-4. In the 2-pyrone the shift for H-1″ and H-2″ remains the same but that for the pyran ring proton is reduced by 75%. This clearly requires that the prenyl substituent be placed between C-2 and C-4 and identifies the novel compound as 3-(3″, 3″-dimethylallyl)-conrauanalactone (**2**). An acetone extract yielded 5, 7-dihydroxychromone (**8**) and eriodictyol (**9**), both previously reported from the bark of this species.

Examination of the seeds of *G. conrauana* from Douala-Edea revealed **2**, **8** and **9**. No trace of **1** could be detected, although **1** was the only lactone found in stem bark from this source [7]. However, leaves from Douala-Edea yielded **1** and **2** together with **8**, **9** and manniflavanone (**10**), a biflavanone previously isolated from *G. mannii* [4]. By contrast, the heartwood of *G. conrauana* yielded only the biflavonoids morelloflavone (**11**), *O*-methylfukugetin (**12**) and a glycoside of **11**. Both **11** and **12** were characterized by comparison with material isolated from *G. densivenia* [6].

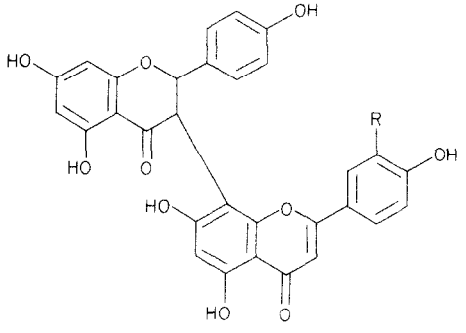
Preparative TLC of a concentrate of the petrol extract of the stem bark of *G. mannii* collected at Korup gave a single compound in low yield (0.0013%). It analysed for $C_{21}H_{38}O_3$ and showed no UV absorption. The IR spectrum gave major bands at

Table 1. Relative lanthanide induced shifts (shift for OMe = 1.0) for protons of **4** and **6**

Compound	Relative shift for:				
	OMe	H-5	H-1″	H-2″	H-7′
4	1.0	5.5	3.3	2.7	1.0
6	1.0	1.3	3.1	2.5	0.5



10 $R_1=R_2=R_3=OH$
14 $R_3=OH, R_1=R_2=H$
15 $R_2=R_3=OH, R_1=H$
16 $R_1=R_2=R_3=H$



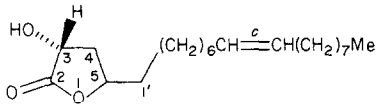
11 R = OH
12 R = OMe

3450 and 1750 cm^{-1} indicating a hydroxyl substituent and suggesting a 5-membered saturated lactone [9]. The EIMS showed a series of fragments indicative of an hydroxy-lactone with a heptadecene side chain. Major fragments for the side chain were $C_{16}H_{30}^{\cdot}$, $C_{15}H_{28}^{\cdot}$, $C_{14}H_{26}^{\cdot}$, and $C_8H_{17}^{\cdot}$, suggesting placement of the double bond at C-8. The lactone moiety gave major fragments for $C_6H_9O_3^+$ and $C_5H_7O_3^+$, both of which readily lost water and which underwent further fragmentation typical of 5-membered lactones [10].

These data suggest a tetrahydrofuranone substituted with a secondary hydroxyl and heptadec-8-en. This hypothesis was sustained by the ^1H NMR spectrum. A signal at δ 4.22 (1H) appeared as a triplet which, on acetylation, resolved into a double doublet (J = 2.5, 5.5 Hz) and was deshielded to δ 5.22. This is typical of an equatorial oxymethine proton coupling only with an adjacent CH_2 and requires the partial structure $-CO-CH(OH)-CH_2-$. A second 1H signal at δ 4.62 occurred as a pentaplet and could be assigned to C-5 where it would couple with the four equivalent protons (δ 2.80) of C-4 and C-1′. A further distinct resonance was observed at δ 5.20 as a triplet for 2H. Both the resonance position and the shape of this signal conform with published data [11] for the olefinic protons of an isolated double bond in an otherwise saturated fatty acid. On the basis of the above the isolated compound can be tentatively characterized as 3-α-hydroxy-5-(heptadec-8′-enyl)-tetrahydrofuran-2-one (**13**). This is the same com-

pound as the unidentified EGC-27, isolated in trace amounts from the stem bark of *G. mannii* from Douala-Edea [8].

An acetone extract of the stem bark yielded three biflavonoids, **10**, the major compound of the stem bark from Douala-Edea [4], and biflavanones GB-1 (**14**) and GB-2 (**15**). The latter two compounds were identified by comparison of spectral data from the parent compound and its fully methylated derivative with that published [12]. The heartwood gave four biflavanones, **10**, **14**, **15**, and GB1a (**16**) in yields of 0.42%, 0.15%, 0.22% and 0.06% respectively. Spectral characteristics of **16** and its fully methylated derivative were in close agreement with published data [12]. The leaves and seeds both yielded the benzophenone xanthochymol (**17**), identified by comparison with material isolated from *G. ovalifolia* [5], and the biflavanones **10**, **14** and **15**.



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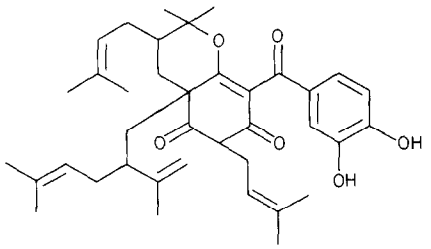

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EXPERIMENTAL

UV spectra were run in EtOH and IR spectra as KCl discs. ^{1}H NMR spectra were run at 90 MHz in $CDCl_3$, unless otherwise stated, using TMS as int. standard. ^{13}C NMR spectra were run in $CDCl_3$, unless otherwise stated, at 25.1 MHz using the FT mode. EIMS were obtained at 70 eV and elevated temps. Mps are uncorr. Petrol refers to the bp 40–60° fraction unless otherwise stated.

Plant material. Bark of *Garcinia conrauana* was collected in the Korup Forest Reserve in the summer of 1979 (voucher: D. W. Thomas 661 at Kew); heartwood, seeds and leaves were collected in the Douala-Edea Forest Reserve in the summer of 1976 (voucher: P. G. Waterman and D. McKey 807 at Kew). Bark of *G. mannii* was collected in the Korup Forest Reserve in the summer of 1979 (voucher: D. W. Thomas 729 at Kew); heartwood, seeds and leaves were collected in the Douala-Edea Forest Reserve in the summer of 1976 (voucher: P. G. Waterman and D. McKey 877).

General extraction procedures for all samples. Samples were milled and then extracted with petrol followed by Me_2CO. Extracts were concd under red. pres.

G. conrauana *stem bark* (160 g). On concn the petrol extract gave a ppt of **2** (1.28 g), recrystallized from petrol–EtOAc as needles, mp 86–89°. Found: $[M]^+$ 390.3155; $C_{25}H_{42}O_3$ requires 390.3134. UV λ_{max} nm: 290. IR ν_{max} cm^{-1} 3100 (OH), 1668 (C=O), 1625, 1588, 1470, 1430, 1405. ^{1}H NMR (Me_2CO-d_6) δ 0.92 (3H, *t*, 15′-Me), 1.22–1.43 (26H, *m*, 13 × CH_2), 1.63, (3H, *s*, 3″-Me), 1.71 (3H, *s*, 3″-Me), 2.42 (2H, *t*, J = 7 Hz, H-1′), 3.06 (2H, *d*, J = 6 Hz, H-1″), 5.28 (1H, *t*, J = 6 Hz, H-2″), 6.00 (1H, *s*, H-5). ^{13}C NMR δ 99.3 (*s*, C-3), 101.0 (*d*, C-5), 122.0 (*d*, C-2″), 130.7 (*s*, C-3″), 163.0, 164.8, 165.0 (3 × *s*, C-2, C-4, C-6). EIMS *m/z* (rel. int.): 390 $[M]^+$ (75), 335 $[M-C_4H_9]^+$ (55), 179 $[M-C_{15}H_{31}]^+$ (16), 139 (16), 126 (10). **2** (580 mg) in Et_2O was reacted with CH_2N_2 to give a mixture of two compounds which were separated by prep. TLC (Si gel: solvent, petrol–EtOAc, 7 : 3) to give **4** (R_f = 0.37, 192 mg) and **6** (R_f = 0.70, 143 mg). **4** was a yellow oil. Found: $[M]^+$ 404.3242; $C_{26}H_{44}O_3$ requires 404.3290. UV λ_{max} nm: 261. IR ν_{max}^{film} cm^{-1}: 1740, 1637, 1595, 1462, 1405. ^{1}H NMR δ 3.97 (3H, *s*, 2-OMe). ^{13}C NMR δ 163.3, 163.6 (2 × *s*, C-2, C-6), 181.0 (*s*, C-4). EIMS *m/z* (rel. int.): 404 $[M]^+$ (77), 389 (93). **6** was a yellow oil. Found: $[M]^+$ 404.3240; $C_{26}H_{44}O_3$ requires 404.3290. UV λ_{max} nm: 300. IR ν_{max}^{film} cm^{-1}: 1700, 1640, 1565, 1465. ^{1}H NMR δ: 3.86 (3H, *s*, 4-OMe). ^{13}C NMR δ 165.1, 165.8 (*s*, C-2, C-4, C-6). EIMS *m/z* (rel. int.): 404 $[M]^+$ (91), 389 (45). TLC of the Me_2CO concentrate (3 systems) revealed **8** and **9**, identified by comparison with authentic samples [7] but not isolated.

G conrauana *seeds* (580 g). Identical treatment to that accorded the stem bark gave **2** (2.1 g) from the petrol concentrate and revealed **8** and **9** in the Me_2CO concentrate.

G. conrauana *leaves* (ca 1 g). Co-chromatography of the petrol concentrate with **1** and **2** (3 systems) revealed the presence of both lactones. Similar investigation of the Me_2CO concentrate revealed **8**–**10**.

G. conrauana *heartwood* (230 g). Prep. TLC of an aliquot (25%) of the Me_2CO concentrate on Si gel (solvent, C_6H_6–C_5H_5N–HCO_2H, 36 : 9 : 5) gave **12** (R_f = 0.37, 48 mg), **11** (R_f = 0.27, 340 mg) and **11**-glyc. (R_f = 0.10, 435 mg). Exhaustive methylation of **11** and **12** [6] gave heptamethyl-**11**, identified by direct comparison (UV, IR, ^{1}H NMR, ^{13}C NMR, EIMS) with an authentic sample. Hydrolysis of **11**-glyc. gave **11**, identified as above.

G. mannii *stem bark* (55 g). Prep. TLC of the petrol concentrate on Si gel (solvent, petrol–EtOAc, 7 : 3) gave **13** (R_f = 0.37, 7 mg) as an oil. Found: $[M]^+$ 338.2811; $C_{21}H_{38}O_3$ requires 338.2821. IR ν_{max}^{film} cm^{-1}: 1750 (C=O), 1460. ^{1}H NMR (Me_2CO-d_6) δ 0.94 (3H, *t*, 17′-Me), 1.27–1.76 (26H, *m*, 13 × CH_2), 2.80 (4H, *m*, H-4, H-1′), 4.22 (1H, *m*, H-3), 4.62 (1H, *pentaplet*, H-5), 5.36 (2H, *t*, J = 7 Hz, H-8′, H-9′). EIMS *m/z* (rel. int.): 338 $[M]^+$ (55), 320 $[M-H_2O]^+$ (7), 222 (10), 208 (5), 194 (6), 129 (75), 115 (5), 111 (26), 99 (1), 97 (1), 57 (100), 43 (36). Acetylation of **13** (4 mg) in C_5H_5N with Ac_2O followed by normal work-up gave **13**-monoacetate as an oil. Found: $[M]^+$ 380.2939; $C_{23}H_{40}O_4$ requires 380.2926. IR ν_{max}^{film} cm^{-1}: 1780, 1750. ^{1}H NMR (Me_2CO-d_6) δ 2.10 (3H, *s*, 3-OAc), 4.83 (1H, *pentaplet*, H-5), 5.22 (1H, *dd*, J_1 = 5.5 Hz, J_2 = 2.5 Hz, H_{eq}-3), 5.36 (2H, *t*, J = 7 Hz, H-8′, H-9′). TLC analysis of the Me_2CO concentrate revealed the presence of **10**, **14** and **15**. All were identified by comparison with material isolated from the heartwood (see below).

G. mannii *heartwood* (165 g). Prep. TLC of the Me_2CO concentrate on Si gel (solvent, $CHCl_3$–MeOH, 4 : 1) gave **10** (R_f = 0.20, 930 mg), **15** (R_f = 0.29, 480 mg), **14** (R_f = 0.42, 335 mg) and **16** (R_f = 0.53, 132 mg). Compound **10** was identified by direct comparison with material previously isolated from this species [4]. Compounds **14**–**16** were all subjected to exhaustive methylation [4] and identified by comparison of spectral data (UV [12], ^{1}H NMR [12], ^{13}C NMR [13], EIMS [12, 14]) with that published for the original compounds and their methylated derivatives.

G. mannii *leaves* (ca 1 g) *and seeds* (500 mg). Petrol concentrates of both samples were shown to contain **17** by

co-TLC (3 systems) with a previously isolated sample of **17** [5]. Similarly, both Me_2CO concentrates were shown to contain **10**, **14** and **15**.

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