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ROTENOIDS, ISOFLAVONES AND CHALCONES FROM THE STEM BARK OF *MILLETTIA USARAMENSIS* SUBSPECIES *USARAMENSIS*

ABIY YENESEW, JACOB O. MIDIWO and PETER G. WATERMAN*†

Department of Chemistry, University of Nairobi, P.O. Box 30197, Nairobi, Kenya; *Phytochemistry Research Laboratories; Department of Pharmaceutical Sciences, University of Strathclyde, Glasgow G1 1XW, Scotland, U.K.

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Key Word Index—*Millettia usaramensis* subsp. *usaramensis*; Leguminosae; stem bark; *trans*-12a-hydroxyrotenoids; α-hydroxydihydrochalcone; isoflavones; cinnamoyl acetate; (+)-12a-epimillettosin; (+)-usararotenoid-A; (+)-12-dihydrousararotenoid-A; (+)-usararotenoid-B; α,4,2′-trihydroxy-4′-*O*-geranyldihydrochalcone; norisojamicin; 4-*O*-geranylcinnamyl acetate; NMR.

Abstract—From the stem bark of *Millettia usaramensis* subsp. *usaramensis* four new 12a-hydroxyrotenoids with the unusual *trans* B/C ring junction ((+)-12a-epimillettosin, (+)-usararotenoid-A, (+)-12-dihydro-usararotenoid-A, and (+)-usararotenoid-B), a new α-hydroxydihydrochalcone (α,4,2′-trihydroxy-4′-*O*-geranyldihydrochalcone), a new isoflavone, (norisojamicin), and a new cinnamyl alcohol derivative (4-*O*-geranylcinnamyl acetate) have been isolated and characterized. In addition, the known compounds 4′-*O*-geranylisoliquiritigenin, isoliquiritigenin, barbigerone, jamaicin and maximaisoflavone-G were identified. The structures were determined on the basis of spectroscopic evidence and chemical transformations. © 1997 Elsevier Science Ltd

INTRODUCTION

In our investigations on the phytochemistry of *Millettia* species of Kenya, we have previously reported new isoflavones from *M. dura* [1, 2]. Here we report the results of our investigation on the stem bark of another species, *Millettia usaramensis* subsp. *usaramensis*.

Millettia usaramensis Taub. is a shrub or small tree 3–7 m tall with small rounded buds. Two subspecies are known; subspecies *usaramensis* and subspecies *australis*. Subspecies *usaramensis* occurs in Kenya and Tanzania while subspecies *australis* is found in Mozambique, Malawi and Zimbabwe [3]. In East Africa the roots of *M. usaramensis* are supposedly used as a remedy for snake bite [4].

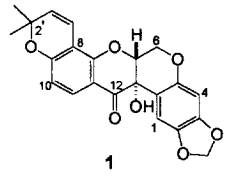
RESULTS AND DISCUSSION

The dried and ground stem bark of *Millettia usaramensis* subsp. *usaramensis* was extracted with ethyl acetate. Silica gel chromatography of the extract using gradient elution from hexane to ethyl acetate resulted in the isolation of 12 compounds of which seven are new.

Compound **1** was isolated as needles. The HREI mass spectrum showed a $[M]^+$ peak at m/z 394.0991 corresponding to molecular formula of $C_{22}H_{18}O_7$. The ^1H NMR (Table 1) and ^{13}C NMR (Table 2) spectra showed this compound to have a 12a-hydroxyrotenoid skeleton (quaternary sp^3 carbon at 66.6 ppm for C-12a). The presence of 2,2-dimethylpyrano and methylenedioxy substituents was evident from ^1H NMR and their placement at C-8/C-9 (159.8 ppm for C-9) and at C-2/C-3 (142.5, 149.5 ppm) was indicated by the ^{13}C NMR data.

The chemical shift value for H-1 (δ 7.67) was strongly deshielded when compared to the values observed for rotenoids with a *cis*-B/C ring junctions (δ 6.4–6.8), indicating that the B/C ring junction has a *trans* stereochemistry [5–8]. However, there are two possible conformers with a *trans*-B/C ring junction, which are defined by the relative configuration of H-6a to the two H-6 protons, in which H-6a can be described as equatorial or axial. In this case analysis of the spin-coupling of protons at C-6 and C-6a showed the presence of one large coupling constant ($J \approx 11.2$ Hz) between H-6a and one of the C-6 protons; thus requiring the presence of a 1,2-*trans*-diaxial relationship [9]. The CD spectrum of **1** (Fig. 1) showed a positive

† Author to whom correspondence should be addressed.



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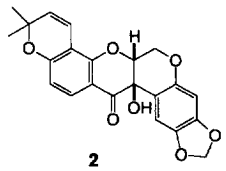

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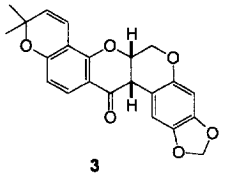

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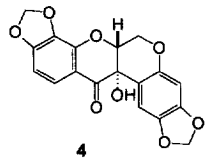

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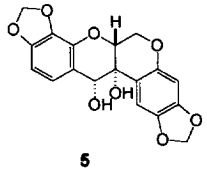

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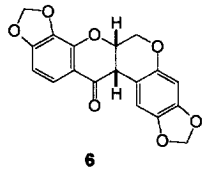

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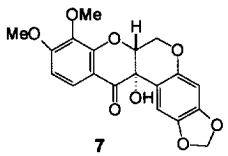

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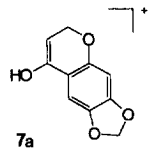
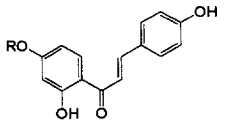
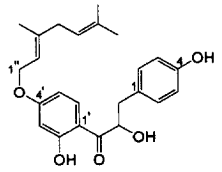


7a



8 R = geranyl

9 R = H



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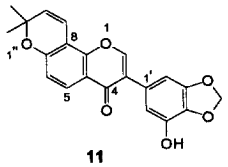

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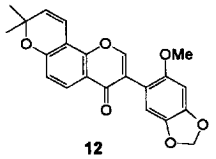

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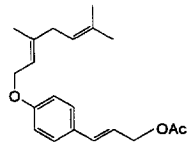

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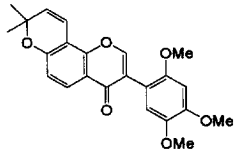

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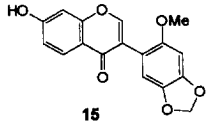

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Cotton effect at *ca* 340 nm. According to Lami *et al.* [8] and an earlier study by Unai *et al.*, a negative Cotton Effect at 340 nm is indicative of absolute stereochemistry of 6a*S*,12a*R* at the B/C ring junction. Consequently **1** can be assigned as 6a*R*,12a*S* (6a-β, 12a-α).

Reduction of **1** with $NaBH_4$ gave the 12,12a-diol corresponding to **1**, from which subsequent treatment with HCl yielded millettone (**3**) in which the stereochemistry at the B/C junction has become *cis*, with H-6a and H-12a both having β orientation. The change of stereochemistry at the B/C ring junction under these reaction conditions has been reported for a synthetic 12-hydroxyrotenoid with a *trans*-B/C junction [11]. Compound **1** is the 12a-epimer of the known compound millettosin (**2**) [12], and hence the name (+)-12a-epimillettosin is proposed.

From the ^1H NMR (Table 1) and ^{13}C NMR (Table 2) spectra it was evident that the second new compound, which analysed by HREI mass spectroscopy for $C_{18}H_{12}O_8$, was a 12a-hydroxyrotenoid with two methylenedioxy substituents. The presence of *ortho*-coupled aromatic protons ($J = 8.4$ Hz) in the ^1H NMR spectrum, appearing at δ 7.88 (H-11) and 6.73 (H-10), places one of the methylenedioxy groups at C-8/C-9. This was also supported by ^{13}C NMR resonances which were consistent with C-7a (145.2 ppm), C-8 (135.2 ppm) and C-9 (154.8 ppm) oxygenation.

The ^1H NMR spectrum further revealed the presence of two isolated aromatic protons, allowing the placement of the other methylenedioxy group at C-2/C-3. As in **1**, H-1 is strongly deshielded, indicating a *trans*-B/C ring junction. The coupling constant between H-6a and one of the C-6 protons was again indicative of a 1,2-*trans* diaxial relationship ($J \approx 11.2$ Hz). Absolute stereochemistry was again assigned as 6a*R*,12a*S* from a positive Cotton effect at 340 nm (Fig. 1). Thus structure **4** could be assigned, to which we have given the trivial name (+)-usararotenoid-A.

The HREI mass spectrum of a third new rotenoid revealed a $[M]^+$ peak at m/z 358.0463, corresponding to the empirical formula $C_{18}H_{14}O_8$. The UV spectrum showed a single benzenoid absorbance band at 285 nm and the IR spectrum did not indicate a carbonyl functional group, suggesting this compound to be a 12-dihydrorotenoid derivative [13]. This was supported by a singlet at δ 5.31 for H-12 (72.0 ppm for C-12) which is consistent with the occurrence of an oxymethine at C-12. The rest of the spectral data for this compound was in agreement with that anticipated for the 12-dihydroderivative of **4** having the same stereochemistry at the B/C junction. The NOESY spectrum showed a strong NOE interaction between H-6a and H-12, so requiring a 1,3-diaxial relationship, and hence the stereochemistry of H-12 must be β. This compound is named 12α-hydroxy-12-dihydro-(+)-usararotenoid-A (**5**).

Reduction of the 12-keto group in **4** with $NaBH_4$ gave a diol, which was identical to **5**. Furthermore on

Table 1. ^{1}H NMR chemical shift data for rotenoids (400 MHz)

H	1	3	4	5	6	7
1	7.67 *s*	6.76 *s*	8.22 *s*	8.63 *s*	6.70 *s*	8.29 *s*
4	6.37 *s*	6.42 *s*	6.69 *s*	6.67 *s*	6.44 *s*	6.70 *s*
6α	4.44 *dd*	4.17 *d*	4.82 *dd*	4.82 *dd*	4.18 *dd*	4.94 *dd*
6β	4.36 *dd*	4.61 *dd*	4.54 *dd*	4.50 *dd*	4.65 *dd*	4.63 *dd*
6a	4.59 *dd*	4.90 *dd*	4.99 *dd*	4.61 *dd*	4.98 *dd*	4.94 *dd*
10	6.54 *d*	6.64 *d*	6.73 *d*	6.70 *d*	6.58 *d*	6.79 *d*
11	7.74 *d*	7.74 *d*	7.88 *d*	7.57 *d*	7.60 *d*	8.05 *d*
12				5.31 *d*		
12a		3.80 *d*			3.86 *d*	
2′-Me	1.45 *s*	1.40 *s*				
	1.48 *s*	1.48 *s*				
3′	5.62 *d*	5.56 *d*				
4′	6.62 *d*	6.64 *d*				
CH_2O	5.91 *s*	5.82 *d*	5.93 *d*	5.92 *d*	5.83 *d*	5.93 *s*
		5.87 *d*	5.95 *d*	5.97 *d*	5.88 *d*	5.95 *s*
			6.03 *d*	5.90 *d*	6.01 *d*	
			6.18 *d*	6.06 *d*	6.08 *d*	
OMe						3.79 *s*
						3.92 *s*

Compounds **1**, **3** and **6** were run in $CDCl_3$, **4**, **5** and **7** run in C_5D_5N. In compounds **1**, **4**, **5** and **7** $J_{6\alpha,6a} \approx 11.2$ Hz, $J_{6\alpha,6\beta} \approx 9.9$ Hz, $J_{6\beta,6a} \approx 4.6$ Hz; in compounds **3** and **6** $J_{6\alpha,6a} \approx 0$ Hz, $J_{6\alpha,6\beta} \approx 12.0$ Hz, $J_{6\beta,6a} \approx 3.6$ Hz, $J_{6a,12a} \approx 4.1$ Hz. In all compounds $J_{10,11} \approx 8.4$ Hz.

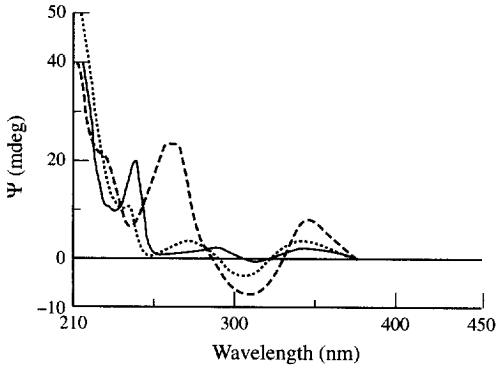
Table 2. ^{13}C NMR chemical shift data for rotenoids (100 MHz)

C	1	3	4	5	6	7
1	109.6	107.2	111.2	109.2	107.0	111.2
2	142.5	142.5	142.9	143.0	144.8	142.9
3	149.5	148.0	149.8	149.4	148.2	149.8
4	98.6	98.9	99.2	98.9	99.2	99.2
4a	150.8	148.6	151.4	150.4	148.8	151.5
6	61.9	66.5	62.7	63.4	66.3	62.9
6a	76.8	72.4	78.2	75.0	72.8	78.2
7a	155.8	157.0	145.2	139.7	142.5	155.2
8	109.1	109.3	135.2	134.6	134.6	137.6
9	159.8	160.3	154.8	148.9	154.9	159.5
10	112.3	111.7	104.4	102.9	104.0	107.4
11	129.9	128.9	124.7	122.7	123.7	125.1
11a	110.8	112.9	118.5	124.3	115.7	116.9
12	189.9	189.3	189.1	72.0	188.9	189.9
12a	66.6	44.9	67.8	65.2	45.6	67.3
12b	113.7	106.0	113.5	118.2	105.4	113.7
2′	78.0	79.9				
2′-Me	28.6	28.7				
	28.4	28.4				
3′	129.4	128.8				
4′	115.6	115.9				
O-CH_2	101.8	101.4	102.4	102.3	101.4	102.4
			103.8	102.5	103.0	
OMe						56.7
						61.2

Compounds **1**, **3** and **6** were run in $CDCl_3$, **4**, **5** and **7** in C_5D_5N.
C assignments were established using HC-COBI and HMBC experiments.

treatment with HCl this reduction product, as well as the natural compound (**5**), afforded compound **6** in which the stereochemistry of the B/C ring junction has become *cis*. This is only the fourth 12-dihydrorotenoid to be reported from nature [5].

The fourth new rotenoid, named (+)-usa-



CD spectrum for rotenoids
----Compound 1 ; —— 4 ; ····· 7 .Run in methanol.

Fig. 1. CD spectra of compounds **1**, **4** and **7**.

rarotenoid-B (**7**) analysed for $C_{19}H_{16}O_8$ by HREI mass spectroscopy. The NMR spectra (Tables 1 and 2) showed it to be a further 12a-hydroxyrotenoid substituted by two methoxyl and a methylenedioxy group. Comparison of chemical shift data with **4** indicated an identical oxygenation pattern, so the only difference was that one of the methylenedioxy groups has been replaced by two methoxyl groups in **7**. The positions of the methoxyl groups were fixed at C-8 and C-9 from a NOESY spectrum, which showed interaction between the methoxyl at C-9 and H-10. This assignment was supported by the EI mass spectrum which showed a fragment ion at *m/z* 192 (**7a**), clearly indicating the placement of the methylenedioxy group in ring-A. The coupling constant between H-6a and one of the C-6 protons, and the chemical shift value for H-1 (Table 1) and the positive Cotton Effect at 340 nm were all consistent with the presence of the same 6a*R*,12a*S* *trans* configuration as seen in **1** and **4**.

A yellow oil, analysing for $C_{25}H_{28}O_8$ by HREI mass spectroscopy, exhibited a UV spectrum typical of a chalcone. The occurrence of this skeleton was supported by the ^{1}H NMR spectrum which showed two *trans*-oriented olefinic protons at δ 7.42 and 7.82 (J = 15.4 Hz) corresponding to H-α and H-β, respectively. The presence of ABX and AA′BB′ spin-systems in the ^{1}H NMR was consistent with oxygenation at C-4, C-2′ and C-4′. In addition both the ^{1}H and ^{13}C NMR spectra revealed the presence of a geranyloxy group. Hydrolysis of this compound afforded isoliquiritigenin (**9**), a compound which was also identified as a constituent of this plant. In a NOE diff. experiment, irradiation of the methylene protons at C-1″ of the geranyl side-chain resulted in enhancement of the signals for H-3′ and H-5′, so placing the geranyloxy group at C-4′. Thus this compound is identified as 4′-geranyloxyisoliquiritigenin (**8**), a compound which has previously been isolated from *Millettia ferruginea* subsp. *darassana* [14].

Compound **10** was isolated as a colourless oil. The HREI mass spectrum showed a $[M]^+$ peak at *m/z* 410.2124 corresponding to the molecular formula $C_{22}H_{34}O_7$. The presence of an α-hydroxydihydrochalcone skeleton was suggested by the UV spectrum (λ_{max} 270, 304 nm), an ABX spin system (δ 2.87, 3.13 for H2-β, δ 5.22 for H-α) and carbon resonances at 202.9 ppm (carbonyl), 72.9 ppm (C-α), 42.2 ppm (C-β), typical of this type of compound [15]. In the ^{1}H NMR spectrum the presence of a chelated hydroxyl group (δ 12.12) and an aromatic ABD spin system is consistent with a C-2′, C-4′ oxygenated A-ring (cf. **8**). The B-ring protons displayed an AA′BB′ spin system which required oxygenation at C-4. The presence of a geranyloxy group was also evident from the NMR spectra. The latter was assigned to C-4′ on the basis of an HMBC study which showed a ^3J correlation peak between H-1″ and C-4′. Therefore, the structure of this compound must be 4′-geranyloxy-α,4,2′-trihydroxydihydrochalcone (**10**), which is novel. Such α-hydroxydihydrochalcones are very rare in nature [15], and compound **10** is the first example of a geranylated α-hydroxydihydrochalcone.

Compound **11** analysed for $C_{21}H_{16}O_6$ by HREI mass spectroscopy and showed the spectroscopic features of an isoflavone with 2,2-dimethylpyrano, methylenedioxy and hydroxyl functional groups. In the ^{1}H NMR spectrum the presence of AB doublets at δ 8.25 and 6.97 (J = 8.7 Hz, corresponding to H-5 and H-6, respectively) was consistent with a 7/8-substituted benzopyran moiety. Two *meta*-coupled protons at δ 7.39 and 7.11 were attributable to H-2′ and H-6′ of a 3′,4′,5′-trisubstituted aryl ring and the chemical shift of the three substituted carbons (143.0, 135.0, 150.0 ppm) indicated all were oxygenated allowing the placement of the hydroxyl at C-3′ and the methylenedioxy at C-4′/C-5′. By default the 2,2-dimethylpyran system must therefore be placed at C-7/C-8 with the oxygen at C-7 (157.9 ppm). Structure **11** could thus be assigned. The trivial name norisojamaicin is proposed, relating it to the known compound jamaicin (**12**) which was also isolated.

Compound **13** was isolated as colourless oil. The HREI mass spectrum showed an ion at *m/z* 328.2088, indicating an empirical formula of $C_{21}H_{28}O_3$. In the ^{1}H NMR spectrum the presence of four aromatic protons in an AA′BB′ spin system (δ 6.87, 7.31, J = 6.8, 2.0 Hz) and two *trans*-olefinic protons (δ 6.15, J = 15.8 Hz and 6.60, J = 15.8, 6.6 Hz) adjacent to an oxymethylene (δ 4.70, J = 6.6 Hz) was consistent with a 4-hydroxycinnamyl alcohol. The presence of acetoxy and geranyloxy substituents were also obvious from the ^{1}H and ^{13}C NMR spectra. A NOESY study confirmed placement of the geranyloxy substituent at C-4 by showing a strong interaction between H-1′ of the geranyl chain and H-3 and H-5 and on this basis the isolate must be 4-*O*-geranylcinnamyl acetate (**13**).

The remaining compounds were identified as jamaicin (**12**) [16], barbigerone (**14**) [17] and maximaisoflavone G (**15**) [18] by comparison of their spectroscopic data with that reported in the literature.

EXPERIMENTAL

Plant material. The stem bark of *Millettia usaramensis* subsp. *usaramensis* was collected in Jadini Forest, Coast province, Kenya in August 1994. The plant was identified at the Herbarium, Botany Department, University of Nairobi, where a voucher specimen is deposited.

Isolation of compounds from the stem bark of M. usaramensis *subsp.* usaramensis. Dried, ground stem bark (1 kg) was extracted with EtOAc by cold percolation. Evapn of the solvent afforded 45 g of crude extract. A 30 g portion of this was subjected to CC on silica gel (300 g) eluting with mixts of hexane and EtOAc, with increasing polarity. The fr. eluted with 3% EtOAc gave **13** (23 mg); 5% EtOAc gave lupeol (1.5 g); 10% EtOAc gave **1** (421 mg), 12% EtOAc gave a mixt. of two compounds which were sepd by CC on Sephadex LH-20 (eluting with $CHCl_3$–MeOH, 1:1) to afford **12** (246 mg) and **8** (387 mg); 15% EtOAc gave **4** (356 mg) and then **14** (68 mg); 20% EtOAc gave **7** (53 mg) and then **9** (17 mg); 25% EtOAc gave **5** (109 mg) followed by **10** (75 mg) and 30% EtOAc gave **11** (56 mg) and then **15** (44 mg).

(+)-12a-*Epimillettosin* (**1**). Needles from MeOH, mp 256–258°, $[\alpha]_D$ +198° (*c* 0.01, MeOH). Found: $[M]^+$ 394.0991; $C_{22}H_{18}O_7$ requires: 394.1047; UV λ_{max} nm (log ε): 235 (4.54), 240 (2.38), 276 (4.38), 312 (3.99). IR ν_{max}^{nujol} cm^{-1}: 3400 (OH), 1675 (C=O), 1590, 1575; ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 394 $[M]^+$ (44), 361 (6), 331 (10), 203 (32, $[C_{13}H_{11}O_3]^+$), 192 (100, $[C_{10}H_8O_4]^+$), 191 (68, $[C_{10}H_7O_4]^+$).

Conversion of **1** *into* **3**. To a round bottom flask containing **1** (50 mg) in MeOH, $NaBH_4$ (15 mg) was added and the mixt. stirred for 30 min. The reaction mixt. was concd, diluted with H_2O and extracted into EtOAc. Purification of the product by CC on silica gel (eluent, CH_2Cl_2) afforded a diol, 12-dihydromillettosin (45 mg) as an amorphous powder, EIMS *m/z* (rel. int.): 396 $[M]^+$ (11), 381 (20, $[M\text{-}Me]^+$), 378 (25, $[M\text{-}H_2O]^+$), 349 (100). The diol was then treated with methanolic HCl (1 drop in 5 ml) at room temp. for 48 hr to give millettone (**3**) (39 mg) as needles from MeOH, mp 178–180° (lit. [12] 180–181°). Found: $[M]^+$ 378.1065 $C_{22}H_{18}O_6$ requires 378.1098. UV, IR identical to lit [12]. ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 378 $[M]^+$ (89), 363 (9, $[M\text{-}Me]^+$), 349 (51), 203 (24, $[C_{12}H_{11}O_3]^+$), 192 (24), 187 (54, $[C_{11}H_7O_3]^+$), 176 (100, $[C_{10}H_8O_3]^+$).

(+)-*Usararotenoid-A* (**4**). Needles from MeOH, mp 208–210°, $[\alpha]_D$ +327° (*c* 0.01, MeOH). Found: $[M]^+$ 356.0463; $C_{18}H_{12}O_8$ requires: 356.0528. UV λ_{max} nm (log ε): 245 (4.38), 295 (3.98). IR ν_{max}^{nujol} cm^{-1}: 3560 (OH), 1685 (C—O), 1630, 1620. ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 356 $[M]^+$ (42), 210 (3), 192 (100, $[C_{10}H_8O_4]^+$), 191 (73).

Conversion of **4** *into* **5**. Compound **4** (50 mg) was reduced with $NaBH_4$, as described above, to give **5** (46 mg). The product was found to be identical (co-TLC, UV, MS) with the compound isolated from this plant.

(+)-12α-*Hydroxy*-12-*dihydrousararotenoid A* (**5**). Needles from MeOH, mp 202–204°, $[\alpha]_D$ +335° (*c* 0.01, MeOH). Found: $[M]^+$ 358.0757, $C_{18}H_{14}O_8$ requires 358.0684. UV λ_{max} nm (log ε): 240 (4.36), 285 (3.60). IR ν_{max}^{nujol} cm^{-1}: 3500 (OH), 3420 (OH), 1630, 1620. ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 358 $[M]^+$ (58), 192 (100, $[C_{10}H_8O_4]^+$), 175 (28), 165 (65).

Conversion of **5** *into* **6**. Compound **5** (50 mg) was treated with methanolic HCl as described above. Purification of the product by CC over silica gel (eluting with CH_2Cl_2) afforded **6** (37 mg). Amorphous. Found $[M]^+$ 340.0556; $C_{18}H_{12}O_7$ requires 340.0579. ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 340 $[M]^+$ (36), 311 (28), 176 (100), 175 (70).

(+)-*Usararotenoid-B* (**7**). Needles from MeOH, mp 176–178°, $[\alpha]_D$ +340° (*c* 0.01, MeOH). Found: $[M]^+$ 372.0821; $C_{19}H_{16}O_8$ requires 372.0840. UV λ_{max} nm (log ε): 246 (4.38), 296 (3.98). IR ν_{max}^{nujol} cm^{-1}: 3450 (OH), 1700 (C=O), 1600, 11 590. ^1H NMR (see Table 1). ^{13}C NMR (see Table 2). EIMS *m/z* (rel. int.): 372 $[M]^+$ (50), 354 (3, $[M\text{-}H_2O]^+$), 279 (3), 242 (2), 210 (11), 192 (100, $[C_{10}H_8O_4]^+$), 191 (85).

4′-O-*Geranylisoliquiritigenin* (**8**). Yellow oil. Found: $[M]^+$ 392.1867, $C_{25}H_{28}O_4$ requires 392.1980. UV, ^1H and ^{13}C NMR identical to lit [13]. EIMS *m/z* (rel. int.): 392 $[M]^+$ (30), 257 (30), 256 (100, $[M\text{-}C_{10}H_{16}]^+$).

Hydrolysis of **8** *to give* **9**. In a round bottom flask containing **8** (20 mg) methanolic H_2SO_4 (5%) was added and the mixt. was refluxed for 1 hr. The solvent was concd, then diluted with H_2O and extracted into EtOAc. Purification of the product by CC over silica gel (eluent, 2% MeOH in CH_2Cl_2) afforded **9** (13 mg), identical (co-TLC, UV, MS) with isoliquiritigenin (**9**) isolated from this plant.

Isoliquiritigenin (**9**). Yellow oil. Found: $[M]^+$ 256.0749; $C_{15}H_{23}O_4$ requires 256.0732. EIMS *m/z* (rel. int.): 256 $[M]^+$ (100), 255 (55), 239 (14), 192 (26), 163 (37), 150 (20), 137 (90), 120 (50).

α,4,2′-*Trihydroxy*-4′-O-*geranyldihydrochalcone* (**10**). Colourless oil. Found $[M]^+$ 410.2124; $C_{25}H_{30}O_5$ requires 410.2085. UV λ_{max} nm (log ε): 270 (4.30), 304 (3.88). ^1H NMR ($CDCl_3$, 400 MHz): δ 6.99 (2H, *d*, *J* = 8.5 Hz, H-2, H-6), 6.69 (2H, *d*, *J* = 8.5 Hz, H-3, H-5), 6.49 (1H, *d*, *J* = 2.4 Hz, H-3′), 6.51 (1H, *dd*, *J* = 2.4, 8.8 Hz, H-5′), 7.59 (1H, *d*, *J* = 8.8 Hz, H-6′), 12.12 (1H, *s*, 2′-OH), 5.22 (1H, *dd*, *J* = 4.0, 6.8 Hz, H-α), 2.87 (1H, *dd*, *J* = 6.8, 14.2 Hz, H-β), 3.13 (1H, *dd*, *J* = 4.0, 14.2 Hz, H-β), 4.11 (2H, *d*, *J* = 6.6 Hz, H-1″), 4.48 (1H, *t*, *J* = 6.6 Hz, H-2″), 2.12 (4H, *m*, H-4″, H-5″), 5.10 (1H, *m*, H-6″), 1.62 (*s*, Me), 1.69 (*s*, Me) 1.76 (*s*, Me). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 128.0 (C-1), 130.8 (C-2, C-6), 115.6 (C-3, C-5), 155.0 (C-4), 110.6 (C-1′), 165.9 (C-2′), 102.0 (C-3′), 166.4 (C-4′), 109.2 (C-5′), 131.2 (C-6′), 202.9 (C=O), 72.9 (C-α), 42.2 (C-β), 65.6 (C-1″), 118.4 (C-2″), 142.7 (C-3″), 39.7 (C-4″), 26.4 (C-5″), 123.8 (C-6″), 132.1 (C-7″), 25.8 (C-Me), 17.9 (C-Me), 16.9 (C-Me). EIMS

m/z (rel. int.): 410 $[M]^+$ (3), 392 (10, $[M-H_2O]^+$), 304 (10), 256 (60), 137 (100).

Norisojamaicin (**11**). Amorphous. Found: $[M]^+$ 364.0968 $C_{21}H_{16}O_6$ requires 364.0942. UV λ_{max} nm ($\log\varepsilon$): 265 (4.30), 302 (4.16). IR ν_{max}^{nujol} cm^{-1}: 3300, 1640, 1620, 1540. ^1H NMR (C_5D_5N, 400 MHz) δ 8.24 (1H, *s*, H-2), 8.25 (1H, *d*, $J = 8.7$ Hz, H-5), 6.97 (1H, *d*, $J = 8.7$ Hz, H-6), 7.11 (1H, *d*, $J = 1.5$ Hz, H-2′ or H-6′), 7.39 (1H, *d*, $J = 1.5$ Hz, H-6′ or H-2′), 5.76 (1H, *d*, $J = 10.0$ Hz, H-3″), 6.80 (1H, *d*, $J = 10.0$ Hz, H-4″), 1.47 (6H, *s*, 2″-Me 2), 6.04 (2H, *s*, O-CH_2-O). ^{13}C NMR (C_5D_5N, 100 MHz): δ 153.4 (C-2), 127.5 (C-3), 175.9 (C-4), 119.4 (C-4a), 127.4 (C-5), 115.7 (C-6), 157.9 (C-7), 110.1 (C-8), 152.9 (8a), 125.4 (C-1′), 102.6 (C-2′ or C-6′), 143.0 (C-3′), 135.9 (C-4′), 150.0 (C-5′), 114.2 (C-6′ or C-2′), 78.5 (C-2″), 131.2 (C-3″), 115.6 (C-4″), 28.5 (Me2-2″), 102.0 (O-CH_2O). EIMS *m/z* (rel. int.): 364 (81), $[M]^+$, 349 (100, $[C_{20}H_{14}O_6]^+$), 187 (87).

Jamaicin (**12**). Needles from MeOH, mp 190–192° (Lit. [16] 189–192°). Found: $[M]^+$ 378.1186; $C_{22}H_{18}O_6$ requires 378.1098. UV, ^1H and ^{13}C NMR identical to lit [16]. EIMS *m/z* (rel. int.): 378 $[M]^+$, (100), 363 (100, $[C_{21}H_{15}O_6]^+$), 347 (100, $[C_{21}H_{15}O_5]^+$).

4-O-*Geranylcinnamyl acetate* (**13**). Oil. Found: $[M]^+$ 328.2059; $C_{21}H_{28}O_3$ requires 328.2088. ^1H NMR ($CDCl_3$, 400 MHz): δ 7.31 (2H, *d*, $J = 8.8$ Hz, H-2, H-6), 6.87 (2H, *d*, $J = 8.8$ Hz, H-3, H-5), 6.60 (1H, *d*, $J = 15.8$ Hz, H-α), 6.15 (1H, *dt*, $J = 6.6$, 15.8 Hz, H-β), 4.70 (2H, *d*, $J = 6.6$ Hz, H-γ), 4.54 (2H, *d*, $J = 6.5$ Hz, H-1′), 5.48 (1H, *t*, $J = 6.5$ Hz, H-2′), 2.15 (4H, *m*, H-4′, H-5′), 5.10 (1H, *m*, H-6′), 1.61, 1.68, 1.74 (3 × *s*, 3 × Me), 2.10 (3H, *s*, OAc). ^{13}C NMR ($CDCl_3$, 100 MHz) δ 129.0 (C-1), 128.0 (C-2, C-6), 115.2 (C-3, C-5), 159.2 (C-4), 121.0 (C-α), 134.3 (C-β), 65.1 (C-$\hat{\gamma}$), 65.6 (C-1′), 119.6 (C-2′), 141.5 (C-3′), 39.8 (C-4′), 26.5 (C-5′), 124.0 (C-6′), 132.0 (C-7′), 25.9 (C-8), 17.9 (C-9), 16.9 (C-10′), 171.1, 21.1 (OAc).

Barbigerone (**14**). Needles from MeOH, mp 153–156° (Lit. [17] 155–157°). Found: $[M]^+$ 394.1405; $C_{23}H_{22}O_6$ requires 394.1410. UV, ^1H and ^{13}C NMR identical to lit [15]. EIMS *m/z* (rel. int.): 394 $[M]^+$, (100), 379 (83, $[C_{22}H_{19}O_6]^+$), 363 (27), 349 (15), 293 (26), 187 (22).

Maximaisoflavone G (**15**). Amorphous. Found: $[M]^+$ 3123.0356; $C_{17}H_{13}O_6$ requires 313.0408. UV, ^1H and ^{13}C NMR identical to lit [18]. EIMS *m/z* (rel. int.): 312 (100).

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REFERENCES

1. Yenesew, A., Midiwo, J. O. and Waterman, P. G., *Phytochemistry*, 1996, **41**, 951.
2. Yenesew, A., Midiwo, J. O. and Waterman, P. G., *Journal of Natural Products* (submitted for publication).
3. Gillet, J. B., Polhill, R. M. and Verdcourt, B., *Flora of Tropical East African—Leguminosae*. Whitefriars Press, London, U.K., 1971, p. 123.
4. Kokwaro, J. O., *Medicinal Plants of East Africa*. East African Literature Bureau, Nairobi, Kenya, 1976.
5. Dewick, P. M., *The Flavonolds: Advances in research since* 1986, ed. J. B. Harborne. Chapman and Hall, London, 1994, p. 117.
6. Messana, I., Ferrari, F. and Santlana, E. G., *Phytochemistry*, 1986, **25**, 2688.
7. Oberholzer, M. E., Rall, G. J. H. and Roux, D. G., *Tetrahedron Letters*, 1974, 2211.
8. Lami, N., Kadota, S., Tezuka, Y. and Kikuchi, T., *Chemistry and Pharmacy Bulletin (Tokyo)*, 1990, **38**, 1558.
9. Fukami, H. and Nakajima, M., in *Naturally Occurring Insecticides*, ed. M. Jacobson and D. G. Crosby. Marcel Dekker, New York, 1971, p. 71.
10. Unai, T., Yamamoto, I., Cheng, H. and Casida, J. E. *Agricultural and Biological Chemistry*, 1973, **37**, 387.
11. Crombie, L. and Godin, P. J., *Journal of the Chemical Society*, 1961, 2861.
12. Ollis, W. D., Rhodes, C. A. and Sutherland, I. O., *Tetrahedron*, 1967, **23**, 4741.
13. Abe, F., Donnelly, D. M. X., Moretti, C. and Polonsky, J., *Phytochemistry*, 1985, **24**, 1071.
14. Dagne, E., Bekele, A., Noguchi, H., Shibuya, M. and Sankawa, U., *Phytochemistry*, 1990, **29**, 2671.
15. Fuchs, A., De Vries, F. W., Landheer, C. A. and Van Veldhuizen, A., *Phytochemistry*, 1984, **23**, 2199.
16. Dagne, E., Bekele, A. and Waterman, P. G., *Phytochemistry*, 1989, **28**, 1897.
17. Dagne, E. and Bekele, A., *Phytochemistry*, 1990, **29**, 2679.
18. Rao, E. V., Murthy, M. S. R. and Ward, R. S., *Phytochemistry*, 1984, **23**, 1493.