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REARRANGED LIMONOIDS FROM *KHAYA SENEGALENSIS*

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Key Word Index—*Khaya senegalensis*; Meliaceae; limonoids; coumarins; sterols; biochemical systematics.

Abstract—The stems of *Khaya senegalensis* yielded three limonoids which appear to be novel. These compounds were identified on the basis of spectroscopic analysis as methyl 1α,6,8α,14β,30β-pentahydroxy-3-oxo-[3.3.1^{10,2}.1^{1,4}]-tricyclomeliac-7-oate; methyl 1α,2β,3α,6,8α,14β-hexahydroxy-[4.2.1^{10,30}.1^{1,4}]-tricyclomeliac-7-oate and methyl 1α-acetoxy-2β,3α,6,8α,14β-pentahydroxy-[4.2.1^{10,30}.1^{1,4}]-tricyclomeliac-7-oate. The two latter ones represent a novel group of methyl tricyclomeliac-7-oates. Scopoletin, scoparon, sitosterol, stigmasterol, Campesterol and 3β-*O*-β-D-glucopyranosylsitosterol were also isolated.

INTRODUCTION

The genus *Khaya* A. Juss. is the main source of African mahogany, and is closely related to the South American genus *Swietenia*, the original source of mahogany. *Khaya senegalensis* (Desr.) A. Juss occurs in the sub-Saharan savannah area from Senegal to Uganda [1]. However, this was introduced in Brazil and showed excellent growth. As part of a continuing study of the chemosystematic of the Meliaceae we have now undertaken a further investigation of stems of exotic *K. senegalensis*. Here, we reported the isolation of three new limonoids, two of which represent a novel group of methyl tricyclomeliac-7-oates.

RESULTS AND DISCUSSION

A dichloromethane-soluble fraction of the methanol extract of the stems of *K. senegalensis* was purified by repeated column chromatography on silica gel and preparative TLC to give 3β-*O*-β-D-glucopyranosylsitosterol and three new limonoids **1**, **2** and **3**.

The major compound was identified as **1** on the basis of the following data. The ^{1}H NMR spectrum (Table 1) indicated the presence of three tertiary methyl groups (δ 0.99, 1.11 and 1.33), one methoxyl singlet (δ 3.66), three downfield shifted signals attributable to a β-substituted furan ring (δ 7.59, 7.56 and 6.47), three signals characteristic of protons attached to a carbon adjacent to an oxygen atom (δ 5.40 *s*; 4.37 *d*, J = 10.3 Hz; 4.22 *dd*, J = 8.0 and 5.0 Hz) and two isolated AB-type methylenes, suggesting the presence of a methyl 1,29-cyclomeliacate skeleton [2–4]. From the ^{1}H–^{13}C long-range correlation spectrum (Table 2) the observed correlations between the methyl proton at δ 0.99 and the ^{13}C signals at δ 206.5 (^3J) and 49.3 (^2J) led to their assignments as C-28, C-3 and C-4, respectively. The methyl proton at δ 1.33 showed long-range correlation with the ^{13}C signal at δ 83.6 (^3J). The oxymethine proton at δ 4.37 was coupled to the ^{1}H signal at δ 2.88 and both signals also showed cross peaks with the ^{13}C signal at δ 83.6 (^3J and ^2J, respectively), so placing the secondary hydroxyl substituent at C-30 and a tertiary hydroxyl substituent at C-1. The signals at δ 1.33 and 2.88 were then assigned to C-19 and H-2, respectively. The oxymethine proton at δ 4.22 showed one-bond correlation with the ^{13}C signal at δ 70.0 and was coupled to the ^{1}H signal at δ 3.25, which showed long-range correlation with the C-4 signal (δ 49.3) and the ^{13}C signal at δ 70.0, thus indicating the hydroxyl group to be located at C-6 and permitting the assignment of the signal at δ 3.25 to H-5. A hydroxyl group must also be connected at C-8 due to the observed correlation between the H-2 signal (δ 2.88) and the ^{13}C signal at δ 86.8. These correlations resulted in the construction of a 1,6,8,30-tetrahydroxy-3-oxo-tricyclo-[3.3.1^{10,2}.1^{1,4}]-decane system.

The large geminal coupling constant of the C-15 methylene protons was consistent with their situation α to a carbonyl group with C-14 fully substituted. This was supported by the long-range ^{1}H–^{13}C COSY, which

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Table 1. ^{1}H NMR chemical shifts for **1**, **1a**, **2** and **3**

H	1	1a	2	3
2	2.88 *d* (10.3)	3.36 *d* (10.4)	4.28 *dd* (7.2, 9.2)	4.45 *dd* (6.7, 9.5)
3			3.21 *dd** (7.2, 9.0)	3.42 *br d* (6.7)
5	3.25 *d* (8.0)	3.21 *d* (3.6)	2.88 *d* (6.8)	2.93 *d* (7.0)
6	4.22 *dd** (8.0, 5.0)	5.13 *d* (3.6)	4.03 *dd** (6.8, 3.6)	4.15 *dd** (7.0, 5.2)
9	2.28 *br d* (10.0)	2.31 *br d* (9.6)	2.03 *br d* (9.0)	2.16 *br d* (7.0)
11β	1.90 *m*	1.37 *m*	1.74 *m*	
11α	1.77–1.83 *m*	1.95 *m*	1.60 *m*	
12β	1.77–1.83 *m*	1.66 *m*	1.78 *m*	
12α	0.85 *br d* (9.0)	0.95 *br d* (10.8)	0.62 *br d* (12.0)	0.81 *m*
15β	2.69 (AB, 18.6)	2.66 (AB, 18.8)	2.63 (AB, 18.5)	2.69 (AB, 18.7)
15α	3.10	3.13	2.86	3.04
17	5.40 *s*	5.51 *s*	5.61 *s*	5.50 *s*
18	1.11 *s*	1.08 *s*	0.96 *s*	1.05 *s*
19	1.33 *s*	1.44 *s*	1.07 *s*	1.21 *s*
21	7.59 *m*	7.40 *m*	7.58 *m*	7.33 *m*
22	6.47 *m*	6.36 *m*	6.45 *m*	6.33 *m*
23	7.56 *m*	7.30 *m*	7.62 *m*	7.37 *m*
28	0.99 *s*	1.10 *s*	0.95 *s*	1.03 *s*
29a	1.78 (AB, 12.6)	2.09 (AB, 12.8)	1.16 (AB, 12.0)	1.74 (AB, 12.2)
29b	2.11	2.86	1.68	2.23
30	4.37 *d* (10.3)	4.42 *d* (10.4)	2.48 *d* (9.2)	3.15 *d* (9.5)
OMe	3.66 *s*	3.71 *s*	3.57 *s*	3.69 *s*
OH	5.44 *d** (5.0)		5.41 *d** (3.6)	5.24 *br s*
OH	4.97 *br s*		4.95 *br s*	4.88 *br s*
OH	4.60 *br s*		4.50 *br s*	
OH			3.70 *d** (9.0)	3.62 *d** (5.2)
OCOMe		2.17 *s*		1.97 *s*
OCOMe		2.10 *s*		

Resonances for **1**, **1a** and **2** were confirmed by ^{1}H/^{1}H and ^{13}C/^{1}H shift-correlated 2D spectra. Coupling constants (Hz, in parentheses).

*Exchangeable with D_2O.

showed the relationship of the 2H-15 signals at δ 2.69 and 3.10 to the C-16 signal at δ 169.9 (^2J) as well as of the C-18 signal at δ 1.11 to the oxygen-bearing C-14 signal at δ 83.0. Moreover, the existence of correlation between the ^{1}H signal at δ 5.40, assigned to H-17, and the ^{13}C signal at δ 141.5 (C-21) determined the position of the furan ring at C-17. The ^{1}H–^{1}H and one-bond ^{1}H–^{13}C COSY experiments suggested the presence of an isolated structural unit, –CH–CH_2–CH_2– in ring C (H-9, δ 2.28; 2H-11, δ 1.90 and 1.77–1.83; 2H-12, δ 1.77–1.83 and 0.85), since it was the only location left in the nucleus.

The stereochemistry suggested for **1** was based on the biosynthesis of limonoids [5]. However, for C-30 the stereochemistry has been assigned by chemical conversions from known limonoids [2, 6]. In a comparable compound, xylocarpin (**4**), the stereochemistry of the oxide was assigned by a study of the alkaline hydrolysis and subsequent methylation [2]. Two products were obtained, one of which was **5**, in which $J_{2,30}$ was 10 Hz (δ 4.43 *d*, H- 30). Inspection of a model for product **5** showed that after opening of the lactone ring, it is possible for the alkoxyl ion produced at C-17 to open an 8α,30α-oxide ring by attack at C-30, forming a six-membered ether ring and an 8α-hydroxyl-group. A similar attack is stereochemically impossible in a β-oxide. Thus, these observations allowed Okorie *et al.* to suggest that, in xylocarpin, H-30 is β and the oxide ring α, in which $J_{2,30}$ is 3 Hz [2].

Based on the above evidence, in compound **1** the coupling constant between H-30 and H-2 (δ 4.37 *d*, J = 10.3 Hz) is characteristic of 30β-oxygenated methyl meliacate derivatives, as in **5**. A model shows that, in **1** and **5**, ring B is nearer to a boat than to a

Table 2. ^{1}H–^{13}C LRCOSY for **1** and HMBC for **1a** and **2**

	C		
H	**1**	**1a**	**2**
2	1, 8	1, 2, 3, 8, 10	
5	4, 6	3, 4, 5, 9, 10, 19	
6		4, 5, 7, 10, Ac	
9		5, 8, 9, 10, 11, 12, 19	
11β		10	
12β		18	
15α	16	14, 15, 16	16
15β	16	13, 14, 16	16
17	21	13, 20, 21, 22, 18	18, 20, 21, 22
18	13, 14	12, 13, 14, 17, 18	12, 13, 14
19	1, 10	1, 5, 9, 10, 19	1, 9, 10
21		20, 22, 23	
22	21, 23	20, 22, 23	22, 23
23	21	20, 22	20, 22, 23
28	3, 4	3, 4, 28, 29/5	4
29a	10	1, 2, 3, 4, 28	
29b	1, 2, 4	1, 3, 10, 29/5	1
30	1	1, 3, 30	1
OMe	7	7, OMe	7, OMe
MeCO (δ 2.17)		Ac, MeCO (δ 21.0)	
MeCO (δ 2.10)		Ac, MeCO (δ 21.9)	

chair conformation, in which H-2 and H-30α are eclipsed. For **1** this was supported by NOESY experiments (Table 3), which showed correlations of the signal of H-30α (δ 4.37) with the signals of H-2 (δ 2.88) and H-15β (δ2.69). In addition, the signal of H-15α (δ 3.10) showed cross peaks with the signal of H_3-18 (δ 1.11), suggesting a spatial proximity of H-15α to H_3-18, which requires the hydroxyl group at C-14 to be *anti* (β) to H_3-18 (α). The correlations from H-17 (δ 5.40), H-5 (δ 3.25), H_3-18 (δ 1.11) and H-9 (δ 2.28) to the signals at δ 1.77–1.83 permitted the assignments of H-12β (H-17β and H-5$\beta \rightarrow$H-12β) and H-11α (H-9α and Meα-18$\rightarrow$H-11α) at δ 1.77–1.83. Moreover, the existence of a correlation from H_3-19 (δ 1.33) to the signal at δ 1.90, showed that this signal can be attributed to H-11β and thus the signal at δ 0.85 corresponds to H-12α. All these correlations require rings C and D nearer to chair conformations. These conclusions were supported by several NOEDIF experiments (Table 4). Moreover, the nOes of the hydroxyl proton at δ 5.19, coming from H_3-18 and H-15α, allowed the assignment of this signal to 8-OH, also showing that the hydroxyl group is thus in the α-configuration. In the same way, the nOes of the hydroxyl proton at δ 4.89, coming from H_3-19 and H-29b, show that this signal can be attributed to the 1α-OH.

The assignment of the stereochemistry of C-30 received further support from acetylation of **1**, which yielded **1a**. The ^{1}H NMR spectrum of **1a** (Table 1) showed the downfield shift of the signal for H-6 (δ 5.13), while the signal for H-30 was still visible as a

Table 3. NOESY 2D NMR for **1**, **1a** and **2**

	H		
H	**1**	**1a**	**2**
2	30	30	30
3			5
5	12β	12β	
6	5, 19, 28, 6-OH	5, 19, 28	19, 28
9	11α, 19	11α, 19	19
11α	18	11β, 12α, 18	
11β	19		
12β	12α	12α	
15α	15β, 18	15β, 18	15β, 30
15β	15α, 30	15α, 30	15α, 30
17	12β, 21	12β, 21, 22	21, 22
21	17	17, 18	17, 18
22	18, 23	17, 18, 23	17, 18
23	22	22	22
29b	19, 28, 29a	28, 29a, Ac	

Table 4. NOEDIF for **1**

Irradiated proton	Observed NOE
Me-18	15α, 21, 22, 8-OH
Me-19	6, 9, 29b, 1-OH (δ 4.89), 6-OH (δ 5.90)
Me-28	6, 29a, 29b, OMe
5	12β, 6
6	19, 28, 6-OH
9	19, 8-OH, 1-OH
15α	15β, 18, 8-OH
17	21, 22
29b	19, 29a, 1-OH
30	2, 15β
8-OH (δ 5.19)	2, 9

doublet at δ 4.42, indicating that it was resistant to acetic anhydride–pyridine–4-dimethylaminopyridine, for steric reasons, on the β-side of the molecule. However, the ^{1}H and ^{13}C NMR spectra (Tables 1 and 5) indicated the presence of two acetoxyl (δ_C 169.4, 169.3 and δ_H 2.10, 2.17). HMBC experiments with **1a** (Table 2) clearly showed correlations from H_2-15 and H_3-18 to the C-14 signal at δ 83.5, from H-9 and H-2 to the C-8 signal at δ 87.6 and from H-2, 2H-29, H-30 and H_3-19 to the ^{13}C signal at δ 91.2, thus indicating the second acetoxyl group to be located at C-1. This is consistent with the stereochemistry for C-1, the least hindered tertiary hydroxyl group of the molecule.

Approach of reagents appears to be accessible only from the least hindered side of the molecule, since Adesogan *et al.* also showed that acetylation of the 3β,30α-dihydroxymexicanolide derivative (**6**) (δ5.35 *d*, J = 3.6 Hz, H-30β), catalysed by 4-dimethylaminopyridine, gave a 30-monoacetate, leaving the 3β-hydroxyl-group unacylated [6].

As a further confirmation of the stereochemistry of **1**, a NOESY experiment was obtained for **1a** (Table 3). The new natural product is, therefore, methyl 1α, 6, 8α, 14β, 30β-pentahydroxy-3-oxo-[3.3.1^{10,2}.1^{1,4}]-tricyclomeliac-7-oate (**1**). The structural assignment was also supported by comparison of the ^{13}C NMR spectrum with those of chukrasin A and mexicanolide [7].

Compound **2** showed spectral characteristics close to those of **1**. The principal change observed in the ^{13}C NMR spectrum (Table 5) of **2** was the replacement of the resonance for a ketonic carbonyl by a signal for an oxymethine (δ 78.2). The ^{1}H–^{1}H COSY spectrum, in addition to correlation between H-6 and H-5, revealed two more oxymethine protons (δ 4.28 and 3.21) which

Table 5. ^{13}C NMR chemical shift for **1**, **1a**, **2** and **3**

C	**1**	**1a**	**2**	**3**
1	83.6	91.2	83.5	91.1
2	62.7	59.6	72.3	72.0
3	206.5	204.0	78.2	78.1
4	49.3	52.1	42.7	44.1
5	41.9	40.7	40.8	39.1
6	70.0	72.7	71.4	71.5
7	173.6	170.1	175.1	175.1
8	86.8	87.6	86.6	86.8
9	54.6	57.3	55.3	55.9
10	59.3	62.0	59.2	60.9
11	16.2	16.5	16.4	16.4
12	26.3	25.6	25.9	26.0
13	36.9	37.5	37.1	37.5
14	83.0	83.5	80.7	81.5
15	33.1	32.4	32.3	32.0
16	169.9	169.7	170.5	170.3
17	80.2	79.9	80.1	80.5
18	14.6	14.2	14.5	14.4
19	18.3	20.5	18.6	18.1
20	120.7	120.4	121.3	120.6
21	141.5	141.1	143.2	142.6
22	110.4	110.1	110.5	110.1
23	143.3	143.0	141.2	140.9
28	15.3	15.6	19.7	19.3
29	44.3	40.8	45.3	41.1
30	74.5	74.0	63.1	58.7
OMe	51.6	52.4	51.4	52.2
OCOMe		169.4		170.4
OCOMe		169.3		
OCOMe		21.9		22.0
OCOMe		21.0		

Assignments based on ^{1}H–^{13}C COSY/^{1}H–^{13}C LRCOSY for **1** and HMQC/HMBC for **1a** and **2**.

were coupled to each other and only one (δ 4.28) to a methine proton (δ 2.48). These observations could not be explained by the presence of hydroxyl groups at C-3 and C-30, indicating that we were not dealing with a normal 1,3,30-trihydroxy-1,29-cyclomeliacate skeleton, tricyclo-[3.3.1^{10,2}.1^{1,4}]-decane system. A pinacol pinacolone rearrangement of a 2,30-dihydroxy-3-oxo-1,29-cyclomeliacate precursor and subsequent reduction may have occurred, resulting in a tricyclo-[4.2.1^{10,30}.1^{1,4}]-decane as in **2**, whose spectroscopic properties accord with the above data. The signals at δ 4.28, 3.21 and 2.48 were then assigned to H-2, H-3 and H-30, respectively. The NOESY experiments (Table 3) showed correlations between H-30 and 2H-15 (δ 2.86, H-15α, a weak, but definite correlation; 2.63, H-15β). This implies that the bridge-head proton and 2H-15 are close to each other in space and demonstrate that the tricyclo is [4.2.1^{10,30}.1^{1,4}]-decane and not [3.3.1^{10,2}.1^{1,4}]-decane, since in **1** these protons are not close.

The NOESY experiments also showed correlations of H-5 with H-3 and of H-30 with H-2, which require a *trans*-relationship for H-3 and H-2, with H-3 on the β-side and H-2 on the α-side of the molecule. This is consistent with the coupling constants between H-3 and H-2 ($J = 7.2$), and between H-2 and H-30 ($J = 9.2$), in which the rearranged six-ring is nearer to a boat conformation where H-2 and H-30 must be eclipsed. Moreover, the observed correlations of H-21 and H-17 and H_3-18, and H-23 with H-22, were very similar to those of **1** and **1a**, except that H-23 (δ 7.62) is more deshielded than H-21 (δ 7.58). In the same way, HMQC experiments showed that C-21 (δ 143.2) is more deshielded that C-23 (δ 141.2). The structure of the new natural product was thus established as methyl 1α,2β,3α,6,8α,14β-hexahydroxy-[4.2.1^{10,30}.1^{1,4}]-tricyclomeliac-7-oate (**2**). The third compound exhibited similar NMR spectra to **2** (Tables 1 and 5) except for the presence of a tertiary acetoxyl (δ_C 170.4, 22.0 and δ_H 1.97). Significant upfield shifts for C-29 and C-30 in the ^{13}C NMR spectrum, when compared with **2** and **1a**, determined the position of the acetoxyl at C-1. Thus, the structure of the new limonoid was characterized as methyl 1α-acetoxy 2β,3α,6,8α,14β-pentahydroxy-[4.2.1^{10,30}.1^{1,4}]-tricyclomeliac-7-oate (**3**).

The dichloromethane extract from the stem of *K. senegalensis* gave a mixture of sterols and two coumarins, which were identified by comparison with published data as scopoletin and scoparon [8]. The mixture of sterols was acetylated and analysed by GC-mass spectrometry, which established that the sterols were sitosterol, stigmasterol and campesterol.

As reported in previous papers [5, 9] species of *Khaya* explore limonoid chemistry along only one route, which leads to the mexicanolide group, bicyclo-[3.3.1^{10,2}]-nonane system. This appears to be the first record of limonoids with a tricyclo-[3.3.1^{10,2}.1^{1,4}]-decane system from *Khaya*, while **2** and **3** represent further enlargement of the biosynthetic mexicanolide-pathways.

EXPERIMENTAL

NMR: on a Bruker ARX 400, on a Bruker ACF 200 and on a Varian Gemini 300, with TMS as int. standard; GC-MS: low resolution on a HP-2576 instrument; MS: 70 eV, direct probe insert and elevated temp.

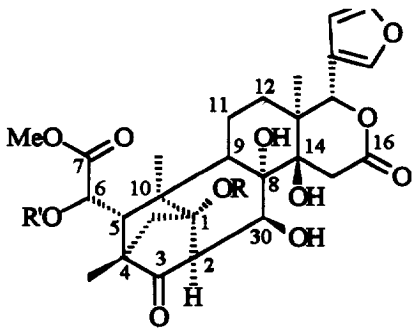
Plant material. Khaya senegalensis was collected in Viçosa, MG, Brazil, and a voucher is deposited in the Herbarium of the Departámento de Engenharia Florestal, Universidade Federal de Viçosa, Viçosa, MG.

Isolation of compounds. Ground stems (600 g) was extracted with hexane, then CH_2Cl_2 and finally with MeOH. The conc. CH_2Cl_2 extract was submitted to vacuum chromatography over silica gel using a hexane–CH_2Cl_2–EtOAc–MeOH gradient. Frs 1–3 gave a mixt. of sterols, fr. 5 gave scoparon (17 mg) and fr. 6 gave scopoletin (20 mg). The coumarins were purified by prep. TLC (silica gel, EtOAc–hexane, 3:2). The mixt. of sterols was acetylated with Ac_2O–pyridine prior to analysis by low resolution GC-MS, which established that the sterols were sitosterol, stigmasterol and campesterol. The conc. MeOH extract was partitioned into CH_2Cl_2-, EtOAc- and *n*-BuOH-soluble frs. The conc. CH_2Cl_2 extract was subjected to CC over silica gel. Elution with a hexane–CH_2Cl_2–EtOAc–MeOH gradient afforded a mixt. of limonoids and 3β-*O*-β-D-glucopyranosylsitosterol (40 mg). The mixt. of limonoids was repeatedly chromatographed on silica gel (hexane–EtOAc–MeOH gradient; hexane–CH_2Cl_2–MeOH, 2:18:1) yielding, after final purification by TLC (hexane–CH_2Cl_2–MeOH, 4:16:1), compounds **1** (70 mg), **2** (17 mg) and **3** (5 mg).

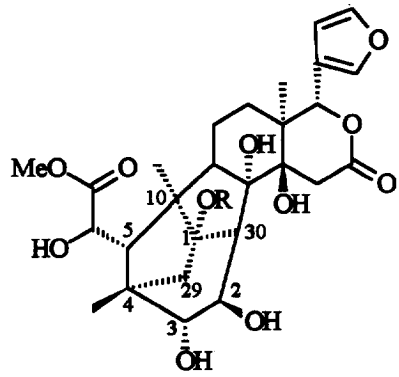
Methyl 1α,6,8α,14β,30β-pentahydroxy-3-oxo-[3.3.1^{10,2}.1^{1,4}]-tricyclomeliac -7-oate (**1**). Amorphous solid, mp 302–305, $[\alpha]_D$ +22.0 (DMSO; *c* 0.064). IR ν_{max}^{KBr} cm^{-1}: 3467, 2954, 1724, 1456, 1281, 1239, 1164, 1087, 1036, 875. ^{1}H NMR (200 MHz, Me_2CO-d_6/trace of DMSO-d_6): see Table 1; ^{13}C NMR (50.32 MHz, DMSO-d_6): see Table 5; ^{1}H–^{13}C COSY long-range correlation (200/50.32 MHz DMSO-d_6): see Table 2; NOESY–TPPI (400 MHz, DMSO-d_6): see Table 3; NOEDIF (300 MHz, DMSO-d_6): see Table 4. MS *m/z* (rel. int.): 516 [M − H_2O]$^+$ (76); 378 (100): associated with retro-Diels–Alder cleavage of ring D and subsequent loss of H_2O; 198 (53); 165 (53); 125 (52); 97 (80); 95 (93): associated with retro-Diels–Alder cleavage of ring D and subsequent loss of H·; 91 (44); 69 (51).

Acetylation of compound **1**. Compound **1** (10 mg) in Ac_2O (0.5 ml), pyridine (0.3 ml) and 4-dimethylaminopyridine (2 mg) was stirred for 10 hr at room temp. and then the mixt. was poured into H_2O. The reaction product (**1a**) was extracted with CH_2Cl_2: amorphous solid (7 mg). ^{1}H NMR (400 MHz, $CDCl_3$): see Table 1; ^{13}C NMR (100 MHz, $CDCl_3$): see Table 5; HMBC (400/100 MHz, $CDCl_3$): see Table 2; NOESY-TPPI (400 MHz, $CDCl_3$): see Table 3.

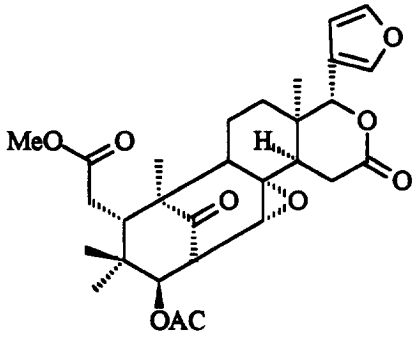
Methyl 1α,2β,3α,6,8α,14β-hexahydroxy-[4.2.1^{10,30}.1^{1,4}]-tricyclomeliac-7-oate (**2**). Amorphous solid, mp 298–303, $[\alpha]_D$ +3.0 (DMSO, 0.024). IR



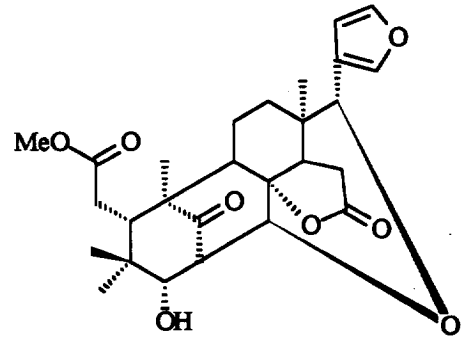
1: R = R' = H
1a: R = R' = Ac



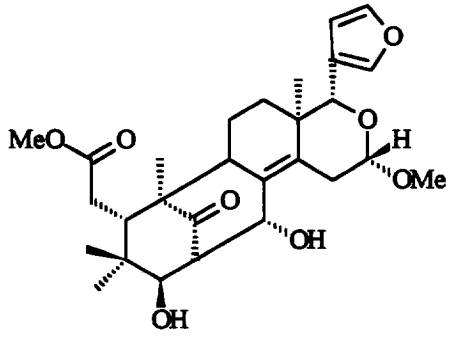
2: R = H
3: R = Ac



4



5



6

ν_{max}^{Film} cm^{-1}: 3400, 2926, 1723, 1450, 1247, 1033. ^1H NMR (400 MHz, DMSO-d_6): see Table 1; ^{13}C NMR (100 MHz, DMSO-d_6): see Table 5; HMBC (400/100 MHz, DMSO-d_6): see Table 2; NOESY-TPPI (400 MHz, DMSO-d_6): see Table 3. MS *m/z* (rel. int.): 518 $[M - H_2O]^{\cdot +}$ (73); 380 (100); 95 (80).

Methyl 1α-*acetoxy*-2β,3α,6,8α,14β-*pentahydroxy*-[4.2.1^{10,30}.1^{1,4}]-*tricyclomeliac*-7-*oate* (**3**). Amorphous solid. IR ν_{max}^{Film} cm^{-1}: 3417, 2927, 1727, 1584, 1459, 1252, 1035. ^1H NMR (400 MHz, $CDCl_3$): see Table 1; ^{13}C NMR (100 MHz, $CDCl_3$/trace of DMSO-d_6): see Table 5.

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REFERENCES

1. Adesida, G. A., Adesogan, E. K., Okorie, D. A., Taylor, D. A. H. and Styles, B. T. (1971) *Phytochemistry* **10**, 1845.
2. Okorie, D. A. and Taylor, D. A. H. (1970) *J. Chem. Soc. (C)* 211.
3. Taylor, D. A. H. (1983) *Phytochemistry* **22**, 1297.
4. Taylor, D. A. H. (1974) *J. Chem. Soc., Perkin 1* 437.
5. Silva, M. F. das G. F. da and Gottlieb, O. R. (1987) *Biochem. Syst. Ecol.* **15**, 85.
6. Adesogan, E. K., MacLachlan, L. K. and Taylor, D. A. H. (1987) *S. Afr. J. Chem.* **40**, 25.
7. Banerji, B. and Nigam, S. K. (1984) *Fitoterapia* **15**, 3.
8. Chatterjee, A., Banerji, B., Ganguly, S. N. and Sircar, S. M. (1974) *Phytochemistry* **13**, 2012.
9. Agostinho, S. M. M., Silva, M. F. das G. F. da, Fernandes, J. B., Vieira, P. C., Pinheiro, A. L. and Vilela, E. F. (1994) *Biochem. Syst. Ecol.* **22**, 323.