Dammarane Triterpenes of *Commiphora confusa* Resin

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Fractionation of a steam distilled residue of *Commiphora confusa* resin has yielded four novel dammarane triterpenes characterised as (20*S*)-3β-acetoxy-12β,16β-trihydroxydammar-24-ene, (20*S*)-12β,16β-trihydroxydammar-24-ene-3β-*O*-β-glucopyranoside, (20*S*)-3β-acetoxy-12β,16β,25-tetrahydroxydammar-23-ene, and (20*S*)-3β,12β,16β,25-pentahydroxydammar-23-ene. The known compounds β-amyrin, 3β-amyrinacetate, 2-methoxyfuranodienone, 2-acetoxyfuranodienone, (20*R*)-3β-acetoxy-16β-dihydroxydammar-24-ene, (20*R*)-3β,16β-trihydroxydammar-24-ene, 3β-acetoxy-16β-hydroxydammar-24-ene, 3β-hydroxydammar-24-ene, 3β-acetoxydammar-24-ene, and β-sistosterol were also isolated from the same extract. The structures of the compounds were determined using spectroscopic, physical, and chemical methods.

Key words dammarane triterpene; resin; *Commiphora confusa*; Burseraceae

The genus *Commiphora* (Burseraceae) is widely distributed in the tropics and subtropics particularly in Africa, Asia, and Australia.[1,2] Over 46 species are found in Kenya and a good number of them are characterised by oleoresins used in medicine, food, and the perfumery industry.[3] *Commiphora confusa* is one such species growing in the wild and produces myrrh, an article of commerce.[4] The plant is also used in herbal medicine as a remedy for microbial infections.[5] Previously, no phytochemical work has been reported on this plant. In this paper we report the isolation of four novel dammarane triterpenes (**1**—**4**) along with several known compounds; β-amyrin and its acetate derivative,[6] 2-methoxyfuranodienone and 2-acetoxyfuranodienone,[7,8] (20*R*)-3β-acetoxy-16β-dihydroxydammar-24-ene (**5**), 3β-hydroxydammar-24-ene (**6**), 3β-acetoxydammar-24-ene (**7**), 3β-acetoxy-16β-hydroxydammar-24-ene (**8**), (20*R*)-3β,16β-trihydroxydammar-24-ene (**9**),[9,10] and β-sistosterol.[11] Their structures were established on the basis of extensive spectroscopic and chemical studies.

Results and Discussion

Compound **1** displayed the characteristics of a dammarane triterpene skeleton in the mass spectrum[9] as evidenced by fragment peaks at *m/z* 435 $[a]^+$, 373 $[b]^+$, 250 $[c]^+$, 189 $[d-CH_3COOH]^+$ (100%), and 109 [side chain$-H_2O]^+$ (Fig. 1). Its ^{13}C-NMR spectrum exhibited 32 carbon atoms and their multiplicity assignments using distortionless enhancement by polarization transfor (DEPT) experiments revealed the presence of 9 methyls, 8 methylenes, 8 methines, and 7 quartenary carbon atoms. The ^{1}H-NMR spectrum corroborated the findings by showing nine signals for the methyls, two of which were deshielded to δ 1.60 and 1.71. By homonuclear decoupling experiments, the deshielded methyls were shown to couple allylically with a vinyl proton (δ 5.16), thus suggesting the presence of a terminal $-CH_2-CH=C(CH_3)_2$ group in the C-17 side chain.[12]

This was confirmed by ^{1}H-detected heteronuclear multiple quantum coherence (HMQC) experiments whereby the two carbons at δ 128.30 (C-24) and 138.40 (C-25) represented a double bond and the former peak was linked to the proton at δ 5.16. Further insight into the structure of **1** was provided by close examination of both the mass spectrum and the ^{1}H-NMR data. The fragments at *m/z* 500 $[M-H_2O]^+$, 482 $[M-2H_2O]^+$, and 458 $[M-CH_3COOH]^+$ together with the ^{1}H-NMR peaks at δ 4.60 (dd, J=11.1, 4.5 Hz), δ 4.30 (ddd, J=7.8, 7.5, 5.2 Hz), and 3.85 (ddd, J=8.0, 5.0, 3.7 Hz) indicated the presence of an acetoxy group and two equatorial secondary hydroxyls, respectively, in the compound. Spin decoupling experiments showed that the acetoxy group was linked in a $CH_2-CH_{ax}(OAc_{eq})-C(CH_3)_2$ system requiring it to be at C-3 as β and because of acetylation there was appreciable

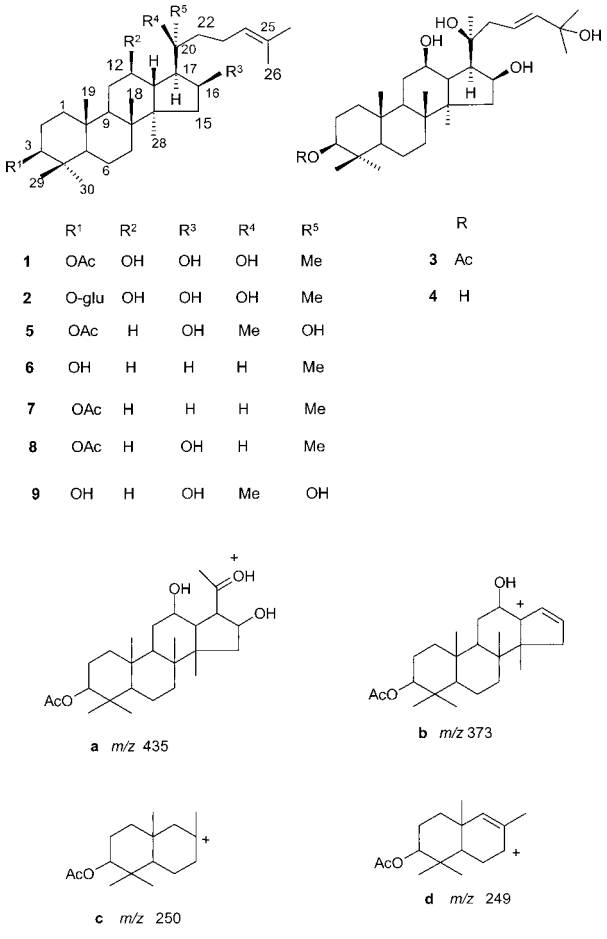

Fig. 1. Significant Fragments of Compound **1** during 70 eV Ionization

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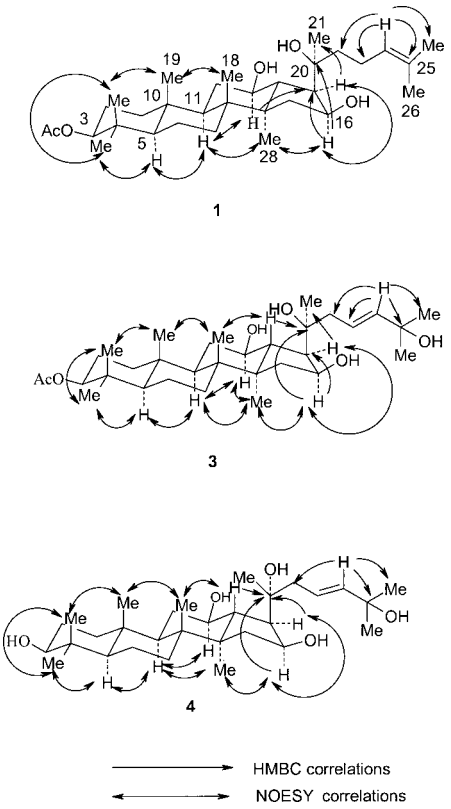

Fig. 2. Pertinent Cross Peaks Observed in the HMBC and NOESY Spectra for Compounds **1**, **3** and **4**

ble shielding of Me_{eq}-4 compared to an equivalent in **3**.[9] The double of doublets at δ 4.30 and 3.85 represented protons in a CH_2–$CH_{ax}(OH_{eq})$–CH system and by spin decoupling experiments in combination with heteronuclear multiple bond connectivity (HMBC) and nuclear Overhauser effect spectroscopy (NOESY) correlations (Fig. 2) were located at C-12 and C-16 both as β. Supportive of these data was the ^{13}C-NMR oxygenated carbon atoms at δ 74.20 (C-3), 75.50 (C-16), 74.30 (C-12), and an ester carbonyl resonance at δ 170.40. Another major feature of the ^{1}H-NMR spectrum was a relatively deshielded methyl singlet at δ 1.27 suggesting that C-20 carried a tertiary hydroxyl group in addition to a methyl. The stereochemistry at C-20 as *S* follows from the cd spectrum in MeOH which afforded a positive Cotton effect at 292 nm ($\Delta\varepsilon$ 2.7) similar to that reported for gugulsterol (294 nm with ($\Delta\varepsilon$ 2.4))[13] and (20*S*)-3β,16β-trihydroxydammar-24-ene (293 nm, $\Delta\varepsilon$ 2.8).[14] On this basis, the structure of compound **1** was concluded to be (20*S*)-3β-acetoxy-12β,16β-trihydroxydammar-24-ene.

Compound **2** which had its mass spectrum obtained at 70 eV, failed to show the molecular ion peak but instead gave an apparent ion at *m*/*z* 476 corresponding to $[M-\text{glucose}]^+$. From preliminary observation, the ^{1}H- and ^{13}C-NMR data were quite similar to those of **1** and the main features of a dammarane glycoside were evident.[15] This was supported by acid hydrolysis, which afforded glucose confirmed by co-spotting with an authentic sample. The ^{13}C-NMR spectrum showed a total of 36 carbons; their multiplicity assignments using DEPT established the presence of 8 methyls, 9 methylenes, 13 methines, and 6 quartenary carbons. By comparison of the respective ^{1}H- and ^{13}C-NMR spectral data, **2** differed from **1** only in the glucose moiety. Like in compound **1**, the ^{1}H-NMR spectrum of **2** showed a double doublet at δ 3.60 (J=11, 4.3 Hz) typical of an axial oxymethine (H-3) proton, and the double doublets at δ 3.75 (J=8.1, 7.4, 5 Hz, H-12) and 4.30 (J=8.5, 7.7, 5.2 Hz, H-16) indicated protons in a CH_2–$CH_{ax}(OH_{eq})$–CH system, thus suggesting two equatorial secondary hydroxyl groups on the tetracyclic ring.[16] Also, the presence of a deshielded methyl singlet at δ 1.30 showed that C-20 carried a hydroxyl group as well as the C-21 methyl. The stereochemistry at C-20 was similarly suggested to be *S* from cd spectrum data which provided a positive croton effect at 290 nm ($\Delta\varepsilon$ 2.4) similar to those reported for compound **1**. On this basis, compound **2** was established to be (20*S*)-12β,16β-trihydroxydammar-24-ene-3β-*O*-glucopyranoside.

Compound **3** afforded a molecular ion peak at *m*/*z* 534 corresponding to a formula of $C_{32}H_{54}O_6$. Its ^{1}H- and ^{13}C-NMR data were quite similar to those of compound **1** with a notable difference being the presence of a double bond (δ 5.67, 5.61) and a hydroxy isopropyl moiety (δ 1.34) in the side chain as evidenced by ^{13}C-NMR and DEPT experiments. The latter revealed the presence of 9 methyls, 7 methylenes, 9 methines, and 7 quartenary carbons, out of which

Table 1. ^{13}C-NMR of Compounds **1**—**4**

Carbon	**1**	**2**	**3**	**4**
1	38.40	38.70	39.10	38.50
2	26.30	25.50	27.40	26.20
3	74.20	73.80	74.50	77.20
4	42.40	42.50	42.80	43.00
5	53.30	55.10	54.70	55.30
6	20.10	19.50	18.90	19.60
7	35.10	34.80	34.50	35.30
8	40.40	41.30	42.10	41.20
9	51.50	52.30	49.80	50.40
10	37.40	36.50	37.30	38.10
11	22.00	21.70	22.30	21.90
12	74.30	76.70	76.30	76.40
13	49.30	49.40	49.70	50.30
14	46.50	47.30	46.80	47.10
15	44.50	44.60	43.90	43.50
16	75.50	74.65	73.20	74.70
17	41.40	40.90	41.25	40.69
18	18.10	17.80	17.70	18.30
19	16.30	15.50	15.75	15.60
20	76.15	75.75	77.80	76.30
21	26.10	27.30	25.75	25.90
22	44.60	43.70	45.40	43.80
23	21.30	22.70	128.15	129.35
24	128.30	125.70	140.10	138.40
25	138.40	132.40	81.20	82.60
26	25.70	25.80	25.00	25.30
27	18.10	17.90	25.10	25.50
28	26.60	27.30	26.40	26.70
29	19.40	21.30	20.70	20.30
30	16.60	15.80	16.20	15.90
OAc	170.40		170.30	171.20
OAC-$\underline{CH_3}$	24.70		23.00	21.30
glucose				
1′		103.70		
2′		75.50		
3′		78.40		
4′		71.50		
5′		77.80		
6′		62.60		

one methyl and one quartenary carbon are from the acetoxy group, a fact supported by an ester carbonyl peak at δ 170.30 and its corresponding methyl at δ 23.0. On this basis **3** was suggested to have an additional hydroxyl group in the side chain. The acetoxy methine proton at δ 4.50 was assigned to C-3 by analogy with compound **1** and the large magnitude of its coupling constant reflected the equatorial configuration of the acetoxy group. This was further supported by the ^{13}C-NMR spectrum, which exhibited the expected shielding and deshielding effect of the 3β acetoxy group on adjacent carbon atoms in the ring.[17] On the other hand, the downfield protons at δ 4.40 and 3.70 showed connectivity with three other protons, thus limiting the position of the corresponding hydroxyl groups to C-12 and C-16. The H-12 showed cross peaks with both protons at C-11 and C-13 while H-16 similarly gave cross peaks with protons on C-15 and C-17 and in both cases the coupling constants were similar to those of **1** and agreed with what has been reported.[9]

Further support for structure **3** was obtained from ^{13}C-NMR spectra, which showed two peaks at δ 77.80 (C-20) and 81.20 (C-25) for tertiary carbons bearing hydroxyl groups as confirmed by the presence of the deshielded C-21 methyl singlet (δ 1.26) and a hydroxy isopropyl moiety (δ 1.34) in the ^{1}H-NMR spectrum, a fact further corroborated by cross peaks in the HMBC spectrum (Fig. 2). The stereochemistry of C-20 sustaining the methyl group in the axial position was established from the cd spectrum which gave a positive Cotton effect at 288 nm ($\Delta\varepsilon$ 2.6). On this basis, compound **3** was structurally elucidated to be (20*S*)-3β-acetoxy-12β,16β,25-tetrahydroxydammar-23-ene.

The last new compound **4** was isolated after repeated flash chromatography on a 2% oxalic acid de-activated silica gel. It showed eight well resolved tertiary methyl groups at δ 1.32 ($CH_3\times2$), 1.26, 1.06, 1.05, 1.02, 0.96, and 0.88 in the ^{1}H-NMR spectrum. The ^{13}C-NMR spectrum exhibited 30 carbon resonances. The DEPT experiment suggested the presence of 8 methyls, 7 methylenes, 9 methines, and 6 quartenary carbon atoms. The HMQC, HMBC, and NOESY experiments were employed in assigning all the ^{1}H- and ^{13}C-NMR values and by comparison with the respective spectral data from compound **3**, it differed from the latter only in the acetate peak.

According to ^{1}H- and ^{13}C-NMR data, the side chain moiety of the compound consisted of two OH groups on substituted quartenary carbons (C-20, δ 76.30 and C-25, δ 82.60) and two germinal methyl groups (C-26 and C-27, δ_H 1.32 with respective δ_C 25.50 and 25.30). Furthermore, the H-24 signal (δ 5.70) exhibited long range coupling with carbon signals of C-22 (δ 43.80), C-23 (δ 129.35), C-26 (δ 25.30), and C-27 (δ 25.50).

The stereochemistry of the chiral carbon C-20 sustaining OH group in an equatorial orientation was defined on the basis of the positive Cotton effect of the cd spectrum (289 nm, ($\Delta\varepsilon$ 2.5)) which was similar to those of compounds **1** and **3**. This was further supported by the similarity in the ^{1}H-NMR data of these compounds. Thus, compound **4** was determined to be (20*S*)-3β,12β,16β,25-pentahydroxydammar-23-ene.

Experimental

General Experimental Procedures Melting points (uncorrected) were determined on a Buchi 535 melting point apparatus. $[\alpha]_D$ values were measured on a JASCO Dip-360 instrument. ^{1}H (360 MHz) and ^{13}C (90 MHz) were determined on a Brucher AM-360 spectrometer. Chemical shifts are reported in δ ppm, with trimethylsilane (TMS) as an internal standard. Eims were measured on a VAT 8200A Varian MAT Bremen instrument. IR data were run as KBr pellets on an infrared FTIR 600 spectrophotometer while UV data were obtained on an IMR-25 spectrophotometer.

Resin Samples The samples were obtained from well identified trees around SALTLIK in the Isiolo district of Kenya in July 1997. Botanical samples including leaves, branches and pods were collected and a voucher specimen (NWFP/KEFRI/97/CC) was deposited at the Kenya Forestry Research Institute Herbarium. Samples were stored in airtight polythene bags and kept in a refrigerator until studied.

Extraction and Isolation The steam distilled resin residue was extracted in the cold with acetone (1 l×3) and the combined extract evaporated under reduced pressure to give a dark gummy material (105 g). A portion of the extract (80 g) was subjected to column chromatography over silica gel using a gradient of EtOAc in *n*-hexane and concluded with MeOH. Fractions of 200 ml each were collected. Based on TLC profiles the collected fractions were combined into five groups (A—E). Group A (fractions 20—102, 17.5 g) was further submitted to chromatography on a silica gel column eluted with a mixture of *n*-hexane–CH_2Cl_2 of increasing polarity of the latter to afford β-amyrin (200 mg) and its acetate derivative (150 mg), 2-methoxyfuranodienone (75 mg), and 2-acetoxyfuranodienone (120 mg). Group B (fractions 103—133, 11.0 g) was purified by repeated low pressure column chromatography using an *n*-hexane–EtOAc mixture of varying concentrations to afford β-sistosterol (85 mg), 3β-hydroxydammar-24-ene (54 mg), and 3β-acetoxydammar-24-ene (36 mg). Group C (fractions 134—150, 9.45 g) upon flash chromatography with an *n*-hexane–EtOAc mixture in the ratios 3 : 1, 1 : 1, and 2 : 1 afforded compound **1** (57 mg), (20*R*)-3β-acetoxy-16β-dihydroxydammar-24-ene (45 mg), and 3β-acetoxy-16β-hydroxydammar-24-ene (65 mg). Group D (fractions 151—175, 14.70 g) upon flash chromatography on de-activated silica gel with solvent systems; I (*n*-hexane–EtOAc, 3 : 1, 1 : 1 and 2 : 1) and II (CH_2Cl_2–MeOH, 9.5 : 0.5) afforded further **1** (21 mg), **3** (76 mg), **4** (36 mg) and (20*R*)-3β,16β-trihydroxydammar-24-ene (45 mg). Group E (fractions 176—220, 11.45 g) was similarly purified as in group D using CH_2Cl_2–MeOH (9 : 1) to afford **2** (60 mg).

Identification of Known Compounds The structures of known compounds were identified by comparison of their physical and spectroscopic data with those of known compounds.

(20*S*)-3β-Acetoxy-12β,16β-trihydroxydammar-24-ene (**1**): Colourless needles, mp 188—190 °C, $[\alpha]_D^{25}$ +46.7° (c=1.0, CH_2Cl_2). CD (c=0.05, MeOH): 240 ($\Delta\varepsilon$ 3.8) and 292 ($\Delta\varepsilon$ 2.7) nm. IR ν_{max} (KBr) cm^{-1}: 3400, 1735, 1630, 1075, and 1040. ^{1}H-NMR δ_H: 5.16 (1H, br t, J=6.7, 1.4 Hz, H-24), 4.60 (1H, dd, J=11.1, 4.5 Hz, H-3), 4.30 (1H, ddd, J=7.8, 7.5, 5.2 Hz, H-16), 3.85 (1H, ddd, J=8.0, 5.0, 3.7 Hz, H-12), 2.80 (1H, m, H-13), 2.50 (1H, ddd, J=14.8, 9.4, 7.3 Hz, H-2$_{ax}$), 2.45 (1H, ddd, J=14.8, 8.0, 5.5 Hz, H-2$_{eq}$), 2.25 (1H, m, H-11), 2.05 (3H, s, CH_3CO–), 1.94 (1H, ddd, J=13, 7.5, 4.4 Hz, H-1$_{eq}$), 1.85 (1H, dd, J=12, 7.3 Hz, H-17), 1.71 (3H, s, CH_3-27), 1.70 (1H, dd, J=13.0, 7.0 Hz, H-15$_{eq}$), 1.60 (1H, s, CH_3-26), 1.53 (1H, dd, J=13, 6 Hz, H-15$_{ax}$), 1.27 (3H, s, CH_3-21), 1.05, 0.98, 0.95, 0.88, 0.85 (15H, s, $CH_3\times5$). ^{13}C-NMR data: see Table 1. Hreims $[M]^+$ 518.2776 (Calcd for $C_{32}H_{54}O_6$, 518.2786). Electron ionization (EI)-MS (eV): m/z (%) M^++1 519 (1), M^+ 518 (3), 500 $[M-H_2O]^+$ (5), 482 $[M-2H_2O]^+$ (11), 458 $[M-CH_3COOH]^+$ (7), 467 (4), 440 (6), 435 $[a]^+$ (15), 418 $[a-OH]^+$ (5), 373 $[b]^+$ (10), 357 (40), 313 $[b-CH_3COOH]^+$ (14), 250 $[c]^+$ (65), 249 $[d]^+$ (10), 248 (2), 207 (75), 189 $[c-CH_3COOH-H]^+$ (100), 141 (33), 109 [side chain$-H_2O]^+$ (45), 55 (30).

(20*S*)-12β,16β-Trihydroxydammar-24-ene-3β-*O*-glucopyranoside (**2**): Colourless crystals, mp >250 °C, $[\alpha]_D^{25}$ +40° (c=0.5, CH_2Cl_2). CD (c=0.05, MeOH): 230 ($\Delta\varepsilon$ 3.7) and 290 ($\Delta\varepsilon$ 2.4). IR ν_{max} (KBr) cm^{-1}: 3450, 1470, 1250, 1150, and 1050. ^{1}H-NMR δ_H: 5.13 (1H, br t, J=7.0, 1.8 Hz, H-24), 4.30 (1H, ddd, J=8.5, 7.7, 5.2 Hz, H-16), 3.75 (1H, ddd, J=8.1, 7.4, 5.0 Hz, H-12), 3.60 (1H, dd, J=11, 4.3 Hz, H-3), 2.74 (1H, m, H-13), 2.46 (1H, ddd, J=15.6, 9.4, 4.4 Hz, H-2$_{ax}$), 2.42 (1H, ddd, J=15.6, 8.5 Hz, H-2$_{eq}$), 1.90 (1H, m , H-11), 1.88 (1H, ddd, J=13.3, 7.5, 4.4 Hz, H-1$_{eq}$), 1.80 (1H, dd, J=12.1, 7.3 Hz, H-17), 1.65 (3H, s, CH_3-27), 1.60 (1H, dd, J=12.8, 5.5 Hz, H-15$_{eq}$), 1.55 (3H, s, CH_3-26), 1.45 (1H, dd, J=12.8, 5.5 Hz, H-15$_{ax}$), 1.35 (1H, ddd, J=13.3, 7.6, 4.6 Hz, H-1$_{ax}$), 1.30 (3H, s, CH_3-21), 1.18, 1.06, 1.01, 0.90, 0.85 (15H, s, $CH_3\times5$); glucose: 4.75 (1H, d, J=7.6 Hz, H-1′), 4.10 (1H, dd, J=12.1, 3.0 Hz, H-6′$_B$), 3.85 (1H, dd, J=12.1, 5.2 Hz, H-6′$_A$), 3.65 (1H, t, J=9 Hz, H-3′), 3.50 (1H, t, J=8.4 Hz, H-4′), 3.43 (1H, dd, J=9.5, 7.5 Hz, H-2′), 3.35 (1H, ddd, J=11, 5.6, 2.8 Hz, H-5′). ^{13}C-NMR data: see Table 1. EI-MS (70 eV): m/z (%) M^+ 476 $[M-glucose]^+$ (1), 458 $[M-glucose-H_2O]^+$ (2), 440 $[M-glucose-2H_2O]^+$ (20), 425 [M−

glucose$-2H_2O-CH_3]^+$ (5), 373 (3), 360 (11), 315 (20), 208 (8), 164 (33), 112 (2), 109 (100), 81(55).

(20*S*)-3β-Acetoxy-12β,16β,25-tetrahydroxydammar-23-ene (**3**): Colourless crystals, mp 244—246 °C, $[\alpha]_D^{25}$ +76° (c=1.0, CH_2Cl_2). CD (c=0.05, MeOH): 236 ($\Delta\varepsilon$ 4.1) and 288 ($\Delta\varepsilon$ 2.6) nm. IR ν_{max} (KBr) cm^{-1}: significant peaks at 3500, 1470, and 1050. ^{1}H-NMR δ_H: 5.67 (1H, m, H-24), 5.61 (1H, m, H-23), 4.50 (1H, dd, J=9.0, 4.3 Hz, H-3), 4.40 (1H, ddd, J=8.0, 7.6, 5.1 Hz, H-16), 3.70 (1H, ddd, J=8.6, 7.7, 5.3 Hz, H-12), 2.89 (1H, dd, J=10.5, 3.5 Hz, H-13), 2.44 (1H, ddd, J=15.6, 9.5, 4.6 Hz, H-2_{ax}), 2.40 (1H, ddd, J=16, 8.0, 4.5 Hz, H-2_{eq}), 1.96 (1H, dd, J=11, 7.3 Hz, H-11), 1.92 (1H, ddd, J=13.3, 7.5, 4.5 Hz, H-1_{eq}), 1.85 (1H, dd, J=12, 5.3 Hz, H-17), 1.70 (1H, dd, J=12.8, 7.2 Hz, H-15_{eq}), 1.50 (1H, dd, J=12.6, 5.2 Hz, H-15_{ax}), 1.34 (6H, s, $(CH_3)_2$-25), 1.30 (1H, m, H-1_{ax}), 1.26 (3H, s, CH_3-21), 1.06, 1.02, 0.98, 0.94, 0.86 (15H, s, $CH_3\times5$. ^{13}C-NMR data: see Table 1. Hreims $[M]^+$ 534.0982 (Calcd for $C_{32}H_{52}O_6$, 534.1968). EI-MS (70 eV): m/z (%) M^++2 536 (1), M^+ 534 (2), 516 $[M-H_2O]^+$ (3), 498 $[M-2H_2O]^+$ (18), 469 $[M-2H_2O-CHO]^+$ (12), 458 $[M-H_2O-CH_3COOH]^+$ (3), 250 (8), 249 (2), 196 (100), 109 (75), 81 (40).

(20*S*)-3β,12β,16β,25-Pentahdroxydammar-23-ene (**4**): Colourless needle crystals, mp >250 °C, $[\alpha]_D^{25}$ +47.6° (c=0.5, CH_2Cl_2). CD (c=0.05, MeOH): 228 ($\Delta\varepsilon$ 4.3) and 289 ($\Delta\varepsilon$ 2.5) nm. IR ν_{max} (KBr) cm^{-1}: 3500, 1610, 1250, 1115, and 1080. ^{1}H-NMR δ_H: 5.70 (1H, m, H-24), 5.65 (m, H-23), 4.50 (1H, ddd, J=7.8, 7.6, 5.5 Hz, H-16), 3.80 (1H, ddd, J=9, 8.0, 5.4 Hz, H-12), 3.70 (1H, dd, J=10.4, 4.4 Hz, H-3), 2.85 (1H, m, H-13), 2.50 (1H, ddd, J=14.8, 10, 7.7 Hz, H-2_{ax}), 2.46 (1H, ddd, J=14.8, 9.4, 7.3 Hz, H-2_{eq}), 2.14 (1H, dd, J=14.3, 7.7 Hz, H-11), 2.0 (1H, ddd, J=12.5, 7, 4.5 Hz, H-1_{eq}), 1.75 (1H, dd, J=12, 7 Hz, H-15_{eq}), 1.46 (1H, dd, J=12.7, 5.5 Hz, H-15_{ax}), 1.32 (1H, m, H-1_{ax}), 1.30 (6H, s, $(CH_3)_2$-25), 1.26 (3H, s, CH_3-21), 0.80 (1H, dd, J=11.5, 3.0 Hz, H-5), 1.06, 1.05, 1.02, 0.96, 0.88 (15H, s, $CH_3\times5$). ^{13}C-NMR data: see Table 1. Hreims $[M]^+$ 492.2667 (Calcd for $C_{30}H_{52}O_5$, 492.2673). EI-MS (70 eV): m/z (%) M^+ 492 (1), 474 $[M-H_2O]^+$ (26), 456 $[M-2H_2O]^+$ (6), 441 $[M-2H_2O-CH_3]^+$ (2), 393 (21), 360 (11), 315 (35), 208 (100), 207 (13), 190 (65), 91 (95).

Acid Hydrolysis Compound **2** (20 mg) in a mixture of 8% HCl (2 ml) and MeOH (20 ml) was heated under reflux for 2 h. The reaction mixture was dried under pressure, dissolved in H_2O (3 ml) and neutralised with NaOH. The neutralised product was then subjected to TLC analysis with an EtOAc–MeOH–H_2O–HOAc (6 : 2 : 1 : 1) solvent system. The chromatograms were sprayed with aniline hydrogen phthalate followed by heating at 100 °C. Glucose was identified after comparison with authentic sample (Rf 5.7).

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References and Notes

1) Dale I. R., Greenway P. G., "Kenya Trees and Shrubs," Buchanan's Kenya Ltd., Nairobi, 1961, pp. 86—95.
2) Beentje H., "Kenya Trees, Shrubs and Lianas," National Museum of Kenya, Nairobi, 1994, pp. 45—51.
3) Trease G. E., Evans W. C., "Pharmacognosy," 11th ed., Balliere Tindall, London, 1988, pp. 463—477.
4) Provan G. J., Waterman P. G., *Phytochemistry*, **49**, 829—834 (1988).
5) Kokwaro J. O., "Medicinal Plants of East Africa," East African Literature Bureau, Nairobi, 1976, pp. 76—85.
6) Ahmad V. U., Ali A., Baqai F. T., Zafar F. N., *Phytochemistry*, **24**, 1035—1037 (1985).
7) Brieskorn C., Noble P., *Tetrahedron Lett.*, **21**, 1511—1514 (1983).
8) Brieskorn C., Noble P., *Phytochemistry*, **22**, 1207—1211 (1983).
9) Fattorusso E., Santacrose C., Xaasan F. C., *Phytochemistry*, **24**, 1035—1037 (1985).
10) Provan G. J., Waterman P. G., *Phytochemistry*, **25**, 917—922 (1986).
11) Manguro Arot L. O., Midiwo J. O., Kraus W., *Planta Medica*, **33**, 156—157 (1997).
12) Verotta L., Orsini F., Tato M., El-Sebakhy N. D., Toima S. M., *Phytochemistry*, **49**, 845—2587 (1984).
13) Anand N., Nityanand S., "Natural Products and Drug Development," Munksgaard, Copenhagen, 1984, pp. 79—84.
14) Khalid S. A., "Chemistry and Chemical Taxonomy of Rutales," Academic Press, London, 1983, pp. 2281—2287.
15) Hasan C. M., Ahmed M., Mofizud-Din A., Waterman P. G., *Phytochemistry*, **23**, 2583—2587 (1984).
16) Ganzera M., Ellemerer-Mueller E. P., Stuppner H., *Phytochemistry*, **49**, 835—838 (1998).
17) Grande M., Torres P., Piera F., Bellido I., *Phytochemistry*, **31**, 1826—828 (1992).