New Eudesmane Derivatives from *Melampodium camphoratum* from the Suriname Rainforest[1]

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Bioassay-guided fractionation of an EtOAc extract of the leaves of *Melampodium camphoratum* using an assay for inhibitors of the degradation of hemin resulted in the isolation of six new eudesmane sesquiterpenes (**1**−**6**) and the known 6-*epi*-*β*-verbesinol coumarate (**7**). The structures of compounds **1**−**6** were established as 6α-(4′-*O*-methyl-7′*E*-coumaryloxy)eudesm-4(14)-ene (**1**), 6α-({4′-*O*-stearyl}-7′*E*-coumaryloxy)eudesm-4(14)-ene (**2**), 6α-({4′-*O*-palmityl}-7′*E*-coumaryloxy)eudesm-4(14)-ene (**3**), 6α-({4′-*O*-[9″*Z*-hexadecenoyl]}-7′*E*-coumaryloxy)eudesm-4(14)-ene (**4**), 6α-(7′*Z*-coumaryloxy)eudesm-4(14)-ene (**5**), and 6α-({4′-acetoxy}-7′*Z*-coumaryloxy)eudesm-4(14)-ene (**6**). Compounds **1**−**4** showed weak activity in the hemin degradation assay, while compounds **5**−**7** were inactive.

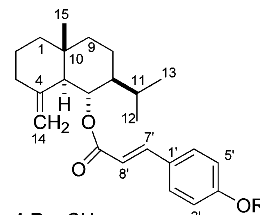
Malaria is a parasitic disease that is a major scourge of the tropical regions of the world, causing more than 300 million acute illnesses and one million deaths annually.[2] Plants have been an important source of antimalarial compounds, with quinine and artemisinin being clinically used drugs, but with many other plant natural products showing activity against the parasite.[3] In an extension of our research on the isolation of bioactive compounds from plants of the Suriname and Madagascar rainforests, as a part of the mission of an International Cooperative Biodiversity Group (ICBG),[4−7] we have recently started working on the isolation and identification of potential antimalarial components from these plants. The bioassay used in this work was a new assay based on the inhibition of the degradation of hemin by glutathione.

The malarial parasite *Plasmodium falciparum* hydrolyzes the hemoglobin content of the erythrocytes of the host to obtain its essential amino acids, and hemin [Fe(III)-protoporphyrin IX] is formed as a byproduct; this is toxic to the parasite. The hemin is detoxified by polymerization or by reaction with glutathione, and several current antimalarial drugs such as chloroquine, quinine, and amodiaquine are thought to act by inhibiting the degradation of hemin by one or both mechanisms, leaving the hemin free to kill the malarial parasite.[8] A simple microtiter plate based assay has been developed to determine the extent of hemin degradation in the presence of drug and glutathione.[9] Not all antiplasmodial compounds are active in the hemin degradation assay, and not all compounds that inhibit the degradation of hemin have antiplasmodial activity, but the correlation is reasonably good, with the assay successfully detecting 16 of 19 antiplasmodial compounds from a collection of 42 natural products and giving only six false positives.[9]

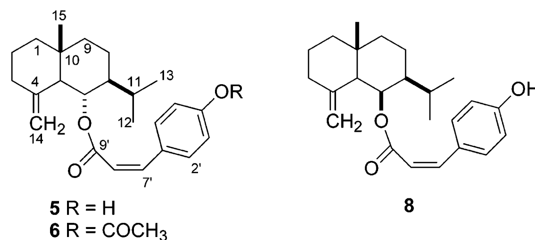
A sample of *Melampodium camphoratum* (L.f.) Baker (Asteraceae) from Suriname tested positive in an initial antimalarial assay conducted in Panama[10] and also in the hemin degradation assay, and it was thus selected for investigation. *M. camphoratum* is a small herb that grows in Brazil, Suriname, and elsewhere; its local name in Suriname is “talangpasibita”. The genus *Melampodium* (Asteraceae) has found extensive use in folk medicine for treatment of malaria, stomach pains, and influenza.[11] Monoterpene glycosides, sesquiterpenes, sesquiterpene lactones, diterpenes, diterpene lactones, flavonoid glycosides, and coumarins have been reported previously from this genus.[12] The only previous studies on the chemistry of *M. camphoratum* have reported the isolation of flavonoids and a sesquiterpene lactone[13] and the variation of its essential oils.[14]

Results and Discussion

Initial liquid−liquid partition of the EtOAc extract of *M. camphoratum* indicated that the activity was concentrated in the hexane- and $CHCl_3$-soluble portions of the hexane/aqueous MeOH and $CHCl_3$/aqueous MeOH partitions. Purification of the hexane- and $CHCl_3$-soluble portions by chromatography over MCI gel followed by reversed-phase HPLC furnished the six new eudesmane sesquiterpenes **1**−**6** and the known 6-*epi*-*β*-verbesinol coumarate (**7**).[15]



1 R = CH_3
2 R = $CO(CH_2)_{16}CH_3$
3 R = $CO(CH_2)_{14}CH_3$
4 R = $CO(CH_2)_7CH{=}CH(CH_2)_5CH_3$
7 R = H



5 R = H
6 R = $COCH_3$

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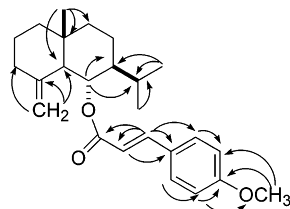

Figure 1. Selected HMBC correlations for **1**.

Compound **1** was isolated as a colorless optically active viscous liquid whose molecular formula was established as $C_{25}H_{34}O_3$ from HRFABMS, ^{13}C NMR, and APT (attached proton test) spectral data. The IR absorption bands at 1701 cm^{-1} suggested the presence of a carbonyl functional group in **1**. The ^{1}H NMR spectrum showed the presence of an oxymethine group as a triplet at δ 5.17 ($J = 10.3$ Hz); one methyl singlet at δ 0.79; two methyl doublets at δ 0.89 ($J = 6.8$ Hz) and 0.90 ($J = 6.8$ Hz); an exocyclic methylene group as singlets at δ 4.56 and 4.75; two olefinic protons as doublets at δ 6.25 ($J = 15.8$ Hz) and 7.59 (2H, $J = 15.8$ Hz); four aromatic protons as doublets at δ 6.87 (2H, $J = 8.9$ Hz) and 7.45 ($J = 8.5$ Hz); one methoxy singlet at δ 3.82; and five methylenes and three methines between δ 1.24 and 2.26. The ^{13}C NMR values for all 25 carbons were assigned on the basis of APT, HMQC, and HMBC spectral data, which indicated the presence of three sp^2 quaternary carbons, two sp^2 methyls, one sp^2 methylene, six sp^2 methines, one sp^3 quaternary carbon, two sp^3 methyls, five sp^3 methylenes, four sp^3 methines, and a carbonyl group in its structure. A search in the literature indicated that the ^{1}H NMR spectral data of **1** were similar to those of the sesquiterpene 6-*epi*-β-verbesinol coumarate (**7**),[15] except for the presence of an additional *O*-methyl group. In the absence of any other oxygenated carbons, the methoxy group in **1** was located at the C-4′ position. Further, methylation of compound **7** with CH_2N_2 yielded **1**, confirming the structure and stereochemistry of the latter compound. This structure was fully consistent with the observed COSY (H-2/H-1, H-3; H-6/H-5, H-7; H-8/H-7, H-9; H-11/H-7, H-12, H-13; H-2′/H-3′; H-4′/H-5′; H-7′/H-8′) and HMBC correlations (Figure 1). The stereochemistry was consistent with the ROESY spectrum of **1**, in which the oxymethine proton at C-6 showed a correlation to the methyl group at C-10 and did not show correlations to the C-5 or C-7 methine protons. The *E* stereochemistry of the double bond between C-7′ and C-8′ was supported by the $J_{7'-8'}$ coupling constant of 15.8 Hz. On the basis of the above spectral data, the structure of **1** was assigned as 6α-(4′-*O*-methyl-7′*E*-coumaryloxy)eudesm-4(14)-ene.

The molecular formula of **2** was deduced as $C_{42}H_{66}O_4$ from its HRFABMS and ^{13}C NMR spectral data. The ^{1}H NMR spectrum of **2** was very similar to that of **1**, except for the replacement of the methoxy signal at δ 3.82 with signals at δ 0.86 (t, 3H, $J = 7.0$ Hz), 1.29 (br s, 28 H), 1.68 (m, 2H), and 2.54 (t, 2H, $J = 7.3$ Hz). Alkaline hydrolysis of **2** furnished 6-*epi*-β-verbesinol coumarate (**7**)[15] and stearic acid ([M$^{+\bullet}$], *m/z* 284), indicating the presence of a stearyl side chain at the C-4′ position. The presence of the stearyl side chain at the C-4′ position was further supported by the IR spectrum of **2**, which showed the presence of two carbonyl groups. One carbonyl absorption was at 1764 cm^{-1}, and the second, as in **1**, was at 1708^{-1}. The ^{13}C NMR spectrum also showed the presence of two carbonyl carbons by signals at δ 167.0 and 172.0. The ^{13}C NMR values for all the carbons were assigned on the basis of HMQC and HMBC spectral data and by comparison with **1** and **7**. The stereochemistry at the four chiral centers (C-5, C-6, C-7, and C-10) was assigned as for **1** and **7** on the basis of their almost identical coupling constants and ^{13}C NMR values. The structure of **2** was thus assigned as 6α-({4′-*O*-stearyl}-7′*E*-coumaryloxy)eudesm-4(14)-ene.

Compound **3** was isolated as a colorless liquid and was determined to have the molecular formula $C_{40}H_{62}O_4$ by HRFABMS. The IR and ^{1}H and ^{13}C NMR spectral data of **3** were almost superimposable on those of **2**, suggesting that it was also a 6-*epi*-β-verbesinol coumarate moiety with a fatty acid side chain at the C-4′ position. Alkaline hydrolysis of **3** furnished **7** and palmitic acid ([M$^{+\bullet}$], *m/z* 256), indicating the presence of a palmityl side chain at the C-4′ position instead of the stearyl side chain in **2**. The structure of compound **3** was thus assigned as 6α-({4′-*O*-palmityl}-7′*E*-coumaryloxy)eudesm-4(14)-ene.

HRFABMS and ^{13}C NMR spectral data deduced the molecular formula of compound **4** as $C_{40}H_{60}O_4$. The IR spectrum showed the presence of two carbonyl functional groups similar to those in **2** and **3**. The ^{1}H NMR spectral data of **4** were similar to those of **3**, except for the side chain at the C-4′ position. The ^{1}H NMR of the side chain showed the presence of a triplet at δ 0.87 ($J = 7.1$ Hz), a broad singlet for 16 protons, three multiplets centered at δ 1.64 (2H), 2.01 (4H), 5.33 (2H), and a triplet at δ 2.34 ($J = 7.3$ Hz), suggesting the presence of a double bond in the ester side chain of **4**. Alkaline hydrolysis of **4** furnished **7** and a compound identified tentatively as palmitoleic acid from its mass spectrum ([M$^{+\bullet}$], *m/z* 254). The structure of **4** was confirmed by partial synthesis from **7**. Esterification of **7** with palmitoleic acid in the presence of EDCI and DMAP furnished **4**, confirming its structure and stereochemistry. The ^{13}C NMR values for all the carbons were assigned on the basis of COSY, HMQC, and HMBC spectral data and in comparison with **2** and **3**. Thus, the structure of **4** was assigned as 6α-({4′-*O*-[9″*Z*-hexadecenoyl]}-7′*E*-coumaryloxy)eudesm-4(14)-ene.

Compound **5** was isolated as a colorless oil, with a molecular formula of $C_{24}H_{32}O_3$ (HRFABMS), identical to that of **7**. The IR spectrum showed the presence of a hydroxy group (3390 cm^{-1}). A comparison of the ^{1}H NMR spectral data of **5** with those of 6β-(7′*Z*-coumaroyloxy)-eudesm-4(14)-ene (**8**)[16] and **7** indicated that the spectrum of the sesquiterpene part of compound **5** was superimposable on that of **7** and the spectrum of the coumaryl part of **5** was superimposable on that of **8**. This finding indicated that **5** was the *Z* isomer of **7**. This was supported by the ROESY spectrum of **5**, which showed a strong correlation between the two protons at C-7′ and C-8′. On the basis of the above spectral evidence, the structure of **5** was assigned as 6α-(7′*Z*-coumaryloxy)eudesm-4(14)-ene.

Compound **6** was isolated as a colorless liquid; its molecular formula was established as $C_{26}H_{34}O_4$ by HRFABMS and ^{13}C NMR spectra. Its ^{1}H NMR spectrum was very similar to that of **5**, except for the presence of a signal for an additional acetate group as a singlet at δ 2.29. The presence of additional carbon signals at δ 169.2 and 21.1 in the ^{13}C NMR of **6** suggested the replacement of the hydroxy group at the C-4′ position with an acetoxy group. This was supported by the absence of the IR absorption band corresponding to that of the hydroxy group and the presence of an additional carbonyl absorption at 1767 cm^{-1}. Further, acetylation of **5** furnished compound **6**, confirming its structure and stereochemistry. Thus, **6** was assigned as 6α-({4′-acetoxy}-7′*Z*-coumaryloxy)eudesm-4(14)-ene.

The isolated compounds were tested in a hemin degradation bioassay, and the four compounds **1**−**4** were all weakly active, with DC_{50} values of 48, 110, 75, and 52 μg/mL,

Table 1. ^{1}H NMR Data for Compounds **1**−**6** ($CDCl_3$)[a]

position	**1**	**2**	**3**	**4**	**5**	**6**
1	1.96 m	1.95 m	1.95 m	1.95 m	1.95 m	1.95 m
2	1.65 m	1.66 m	1.66 m	1.66 m	1.68 m	1.68 m
3	1.50 m, 1.24 m	1.50 m, 1.26 m	1.52 m, 1.26 m	1.52 m, 1.30 m	1.52 m, 1.32 m	1.50 m, 1.30 m
5	2.10 d 10.7	2.10 d 10.7	2.10 d 10.6	2.10 d 10.6	2.04 d 10.8	2.09 d 10.6
6	5.17 t 10.3	5.17 t 10.4	5.17 t 10.1	5.17 t 10.3	5.12 t 10.3	5.17 t 10.3
7	1.42 m	1.40 m	1.38 m	1.42 m	1.38 m	1.42 m
8	1.54 m, 1.30 m	1.56 m, 1.23 m	1.56 m, 1.24 m	1.54 m, 1.24 m	1.56 m, 1.26	1.58 m, 1.26 m
9	2.26 m	2.27 m	2.28 m	2.28 m	2.29 m	2.26 m
11	1.60 m	1.62 m	1.64 m	1.60 m	1.62 m	1.60 m
12	0.89 d 6.8	0.89 d 7.0	0.89 d 6.7	0.89 d 6.9	0.87 d 6.7	0.89 d 6.7
13	0.90 d 6.8	0.89 d 7.0	0.89 d 6.7	0.89 d 6.9	0.88 d 6.7	0.89 d 6.7
14	4.75 s, 4.56 s	4.74 s, 4.54 s	4.74 s, 4.54 s	4.74 s, 4.55 s	4.77 s, 4.59 s	4.74 s, 4.54 s
15	0.79 s	0.78 s	0.78 s	0.78 s	0.76 s	0.78
2′,6′	6.87 d 8.9	7.07 d 8.5	7.07 d 8.9	6.81 d 8.5	6.77 d 8.5	7.05 8.7
3′,5′	7.45 d 8.5	7.50 d 8.5	7.50 d 8.6	7.40 d 8.5	7.60 d 8.6	7.72 d 8.7
7′	7.59 d 15.8	7.60 d 15.8	7.60 d 15.8	7.57 d 15.8	6.78 d 12.8	6.85 d 13.3
8′	6.25 d 15.8	6.33 d 15.8	6.33 d 15.8	6.24 d 15.8	5.75 d 12.8	5.83 d 13.3
2″		2.54 t 7.3	2.54 t 7.3	2.34 t 7.3		
3″		1.68 m	1.68 m	1.64 m		
4″−7″		1.29 br s	1.28 br s	1.30 br s		
8″		1.29 br s	1.28 br s	2.01 m		
9″,10″		1.29 br s	1.28 br s	5.33 m		
11″		1.29 br s	1.28 br s	2.01 m		
12″−15″		1.29 br s	1.28 br s	1.30 br s		
16″		1.29 br s	0.87 t 7.0	0.87 t 7.1		
17″		1.29 br s				
18″		0.86 t 7.0				
OCOCH$_3$						2.29 s
OCH$_3$	3.82 s					

[a] Assignments made on the basis of COSY, HMQC, and HMBC spectral data and in comparison with the literature data.[15,16]

respectively. Compounds **5**−**7** were inactive in this assay, with DC$_{50}$ values > 200 μg/mL. The compounds were also tested for antiplasmodial activity, but disappointingly none of them showed significant activity. It thus appears probable that the hemin degradation assay did not track well with the presumed antiplasmodial activity in this case and that further work will be needed to isolate the compound or compounds giving rise to the latter activity.

Experimental Section

General Experimental Procedures. Optical rotations were recorded on a Perkin-Elmer 241 polarimeter. IR and UV spectra were measured on MIDAC M-series FTIR and Shimadzu UV-1201 spectrophotometers, respectively. NMR spectra were obtained on a JEOL Eclipse 500 spectrometer. The chemical shifts are given in δ (ppm) with TMS (tetramethylsilane) as internal reference, and coupling constants are reported in Hz. Mass spectra were obtained on a JEOL HX-110 instrument. MCI gel (CHP20P) was used for column chromatography. Reversed-phase preparative TLC was performed on Baker Si-C$_{18}$F plates. HPLC was performed on a Shimadzu LC-10AT instrument with an ODS A323 column (250 × 10 mm).

Antimalarial Assays. Activity against *P. falciparum* was determined by using the microfluorimetric assay previously reported;[17] results are expressed as IC$_{50}$ values (μg/mL). A modified 96-well hemin-glutathione degradation assay was used for the in vitro antimalarial assay.[9] Data are presented as DC$_{50}$ values, corresponding to the dose required to reduce the decomposition of hemin by 50% as compared with a control experiment without drug. In this determination, the control drug quinine has a DC$_{50}$ value of 1.3 μg/mL.

Plant Material and Extract Preparation. The leaves, stems, and twigs of *Melampodium camphoratum* (L.f.) Baker (Asteraceae) were collected in early 2000 on cultivated land in the district of Sipaliwini, Suriname, and were assigned collector number 988DeErAK02903. The voucher specimen is deposited in the National Herbarium of Suriname, Paramaribo. The plant samples of *M. camphoratum* were dried and extracted with EtOAc to yield crude extract E980306.

Isolation of Bioactive Compounds. The crude EtOAc extract (0.30 g) was suspended in aqueous MeOH (MeOH−H_2O, 9:1, 50 mL) and extracted with hexane (3 × 50 mL). The aqueous layer was then diluted to 70% MeOH (v/v) with H_2O and extracted with $CHCl_3$ (3 × 50 mL). The aqueous layer was concentrated, and the residue obtained was suspended in H_2O (25 mL) and extracted with *n*-BuOH (2 × 25 mL). The *n*-hexane and $CHCl_3$ extracts were selected for further fractionation based on their activity and ^{1}H NMR patterns. The residue from the *n*-hexane extract (0.15 g) was chromatographed over MCI gel using MeOH−H_2O (1:1 to 100:0) to yield seven fractions (A−G), of which fractions D and E were found to be active. Fraction D upon reversed-phase HPLC with the mobile phase CH_3CN−H_2O (90:10) yielded the known sesquiterpene **7** (4.0 mg) and a new sesquiterpene **6** (2.4 mg). Fraction E on reversed-phase HPLC with the mobile phase CH_3CN−H_2O (98:2) yielded two new sesquiterpenes, **2** (4.3 mg) and **3** (7.6 mg).

The residue from the $CHCl_3$ extract (0.12 g) was chromatographed over MCI gel using MeOH−H_2O (1:1 to 100:0) to yield nine fractions (A−I), of which fractions E−G were found to be active. Fraction E on reversed-phase HPLC with the mobile phase CH_3CN−H_2O (85:15) yielded the known compound **7** (1.8 mg) in addition to the new sesquiterpene **1** (2.1 mg). Fraction F on reversed-phase HPLC with the mobile phase CH_3CN−H_2O (90:10) yielded the known sesquiterpene **7** (14.6 mg) and the new sesquiterpene **5** (5.2 mg). Fraction G on reversed-phase HPLC with the mobile phase CH_3CN−H_2O (70:30) furnished the new sesquiterpene **4** (2.8 mg) and the known sesquiterpene **7** (2.1 mg). The known compound **7** was identified as 6-*epi*-β-verbesinol coumarate on the basis of the comparison of its ^{1}H and ^{13}C NMR and HRFABMS spectroscopic data with those reported in the literature.[15]

6α-(4′-*O*-Methyl-7′*E*-coumaryloxy)eudesm-4(14)-ene (1): colorless liquid; $[\alpha]^{25}_{D}$ −54° (*c* 0.015, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 218 (3.85), 280 (4.12) nm; IR ν_{max} 2930, 2866, 1701, 1603, 1511, 1252, 1170, 826, 755 cm^{-1}; ^{1}H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 383.2577 [M + H]$^+$ (calcd for $C_{25}H_{35}O_3$, 383.2586).

Methylation of 7. To a solution of compound **7** (2.0 mg) in diethyl ether (2 mL) was added a freshly prepared solution of CH_2N_2 (5 mL), and the mixture was left in the refrigerator

Table 2. ^{13}C NMR Data for Compounds **1**−**6** ($CDCl_3$)[a]

carbon	1	2	3	4	5	6
1	40.3	40.3	40.3	40.3	40.3	40.3
2	24.2	24.4	24.3	24.2	24.3	24.3
3	42.3	42.3	42.3	42.2	42.2	42.2
4	146.4	146.4	146.4	146.4	146.5	146.4
5	56.0	56.0	56.0	56.0	55.9	56.0
6	71.2	71.5	71.5	71.4	71.1	71.6
7	49.7	49.6	49.6	49.7	49.5	49.6
8	18.4	18.4	18.4	18.4	18.4	18.4
9	38.2	38.1	38.2	38.1	38.2	38.2
10	38.3	38.2	38.3	38.2	38.3	38.3
11	26.2	26.2	26.2	26.2	26.2	26.2
12	17.7	17.7	17.7	17.7	17.6	17.6
13	21.5	21.5	21.5	21.5	21.4	21.4
14	106.9	106.8	106.8	106.8	106.7	118.8
15	16.2	16.2	16.2	16.2	16.2	16.2
1′	127.4	132.3	132.3	127.4	127.6	132.4
2′,6′	129.8	129.2	129.2	130.0	132.4	129.3
3′,5′	114.3	122.1	122.1	115.8	115.0	122.1
4′	161.3	152.1	152.1	157.7	156.4	152.0
7′	144.3	146.4	146.4	144.5	143.3	143.5
8′	116.2	118.8	118.8	116.0	114.9	115.9
9′	167.5	167.1	167.1	167.8	166.6	167.1
1″		172.1	172.1	179.4		
2″		34.5	34.5	34.0		
3″		25.0	25.0	24.7		
4″−6″		29.3−29.8	29.3−29.8	29.2−29.8		
7″		29.3−29.8	29.3−29.8	31.9		
8″		29.3−29.8	29.3−29.8	27.2		
9″		29.3−29.8	29.3−29.8	130.1		
10″		29.3−29.8	29.3−29.8	130.1		
11″,12″		29.3−29.8	29.3−29.8	27.2		
13″		29.3−29.8	29.3−29.8	31.9		
14″		29.3−29.8	32.0	29.2−29.8		
15″		29.3−29.8	22.8	22.7		
16″		32.0	14.2	14.2		
17″		22.8				
18″		14.2				
$OCOCH_3$						169.2
$OCOCH_3$						21.2
OCH_3	55.4					

[a] Assignments made on the basis of COSY, HMQC, and HMBC spectral data and comparison with the literature values.[15,16]

for 16 h. The product was dried under vacuum and the residue was purified over reversed-phase preparative HPLC with the mobile phase MeOH−H_2O (85:15) to furnish a product (1.2 mg) that was identified as **1** by co-TLC and its ^1H NMR spectrum.

6α-({4′-*O*-Stearyl}-7′*E*-coumaryloxy)eudesm-4(14)-ene (2): colorless liquid; $[\alpha]^{25}_D$ −55.4° (*c* 0.035, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 217.8 (4.21), 280 (4.08) nm; IR ν_{max} 2918, 2849, 1764, 1708, 1637, 1506, 1209, 1176, 758 cm^{-1}; ^1H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 635.5034 [M + H]$^+$ (calcd for $C_{42}H_{67}O_4$, 635.5039).

Alkaline Hydrolysis of 2. To a solution of compound **2** (2 mg) in MeOH (1 mL) was added 5% methanolic KOH (2 mL), and the reaction mixture was refluxed for 1 h. The mixture was concentrated, and water (5 mL) was added. The aqueous layer was extracted with CH_2Cl_2 (3 × 10 mL), and the combined organic layer was concentrated to yield a brown solid, which on purification by preparative TLC (hexane−EtOAc, 75:25) furnished a colorless solid (0.5 mg) identified as 6-*epi*-*β*-verbesinol coumarate (**7**) by spectral (^1H NMR and HRFABMS) data.[15] The aqueous layer was acidified with 1 N HCl and extracted with EtOAc (3 × 10 mL) to yield a brown viscous liquid, which on preparative TLC (hexane−EtOAc, 50:50) furnished stearic acid (0.4 mg, [$M^{+\bullet}$], *m/z* 284).

6α-({4′-*O*-Palmityl}-7′*E*-coumaryloxy)eudesm-4(14)-ene (3): colorless liquid; $[\alpha]^{25}_D$ −15.5° (*c* 0.065, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 218 (4.32), 273 (3.93) nm; IR ν_{max} 2918, 2849, 1762, 1705, 1637, 1506, 1463, 1210, 1165, 756 cm^{-1}; ^1H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 607.4730 [M + H]$^+$ (calcd for $C_{40}H_{63}O_4$, 607. 4726).

Alkaline Hydrolysis of 3. Hydrolysis of **3** (2.5 mg) as reported above furnished **7** and palmitic acid ([$M^{+\bullet}$], *m/z* 256).

6α-({4′-*O*-[9″*Z*-Hexadecenoyl]}-7′*E*-coumaryloxy)-eudesm-4(14)-ene (4): colorless liquid; $[\alpha]^{25}_D$ −18.6° (*c* 0.015, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 205 (4.08), 282 (3.85) nm; IR ν_{max} 2928, 2853, 1708, 1605, 1515, 1190, 1167, 832 cm^{-1}; ^1H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 605.4570 [M + H]$^+$ (calcd for $C_{40}H_{61}O_4$, 605.4570).

Alkaline Hydrolysis of 4. Hydrolysis of **4** (1.4 mg) as reported above furnished **7** and palmitoleic acid ([$M^{+\bullet}$], *m/z* 254).

Esterification of 7. Compound **7** (5 mg) in CH_2Cl_2 (1 mL) and EDCI (6 mg) were added to a solution of palmitoleic acid (5 mg) and DMAP (2 mg) in CH_2Cl_2 (1 mL), and the resulting mixture was stirred at room temperature for 5 h. EtOAc (10 mL) was added to quench the reaction, and the mixture was concentrated to yield a pale brown viscous liquid. The brown viscous liquid on purification by reversed-phase preparative TLC (MeOH−H_2O, 80:20) furnished a colorless liquid (1.2 mg), which was identified as **4** by co-TLC and ^1H NMR spectral data.

6α-(7′*Z*-Coumaryloxy)eudesm-4(14)-ene (5): colorless liquid; $[\alpha]^{25}_D$ −30.8° (*c* 0.018, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 216 (4.12), 279 (4.42) nm; IR ν_{max} 3390, 2930, 2871, 1648, 1604, 1512, 1442, 1156, 855, 747 cm^{-1}; ^1H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 369.2428 [M + H]$^+$ (calcd for $C_{24}H_{33}O_3$, 369.2430).

6α-({4′-Acetoxy}-7′*Z*-coumaryloxy)eudesm-4(14)-ene (6): colorless liquid; $[\alpha]^{25}_D$ −56.3° (*c* 0.019, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 218 (3.92), 282 (4.14) nm; IR ν_{max} 2930, 2866, 1767, 1704, 1636, 1506, 1201, 1176, 1009, 912, 833, 756 cm^{-1}; ^1H and ^{13}C NMR, see Tables 1 and 2; HRFABMS *m/z* 411. 2519 [M + H]$^+$ (calcd for $C_{26}H_{35}O_4$, 411.2535).

Acetylation of 5. Compound **5** (1.2 mg) was dissolved in pyridine (0.4 mL) and acetic anhydride (0.4 mL), and the mixture was stirred for 20 h at room temperature. The product was dried under vacuum, and the residue obtained was purified by reversed-phase HPLC with the mobile phase MeOH−H_2O (80:20) to furnish **6** (0.7 mg), identified by co-TLC and ^1H NMR spectral data.

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Supporting Information Available: ^1H and ^{13}C NMR spectra of compounds **1**−**6**. This material is available free of charge via the Internet at http://pubs.acs.org.

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