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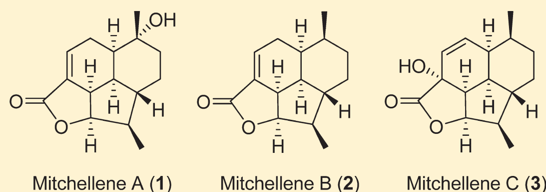
Mitchellenes A−E, Cyclic Sesquiterpenes from the Australian Plant *Eremophila mitchellii*

Emma C. Barnes, Anthony R. Carroll, and Rohan A. Davis*

Eskitis Institute, Griffith University, Brisbane, QLD 4111, Australia

Supporting Information

ABSTRACT: Chemical investigations of the Australian plant *Eremophila mitchellii* resulted in the isolation of the novel tetracyclic sesquiterpene lactones mitchellenes A−C (**1**−**3**), the new sesquiterpene acids mitchellenes D and E (**4** and **5**), and the previously reported natural products 14-hydroxy-6,12-muuroloadien-15-oic acid (**6**), casticin, and centaureidin. The chemical structures of all compounds were determined by extensive 1D/2D NMR and MS data analysis. Mitchellenes A−C are the first tetracyclic sesquiterpene lactones to be reported; a biosynthetic pathway is proposed for these unique secondary metabolites.



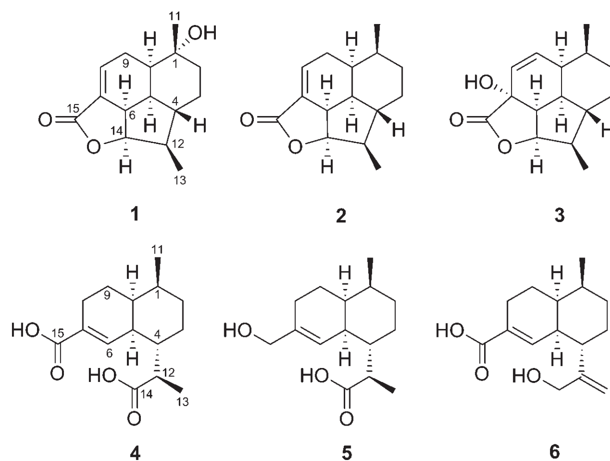
The *Eremophila* genus (family Myoporaceae) contains around 200 species, all of which are native to Australia.[1,2] This genus is the source of over 100 characterized natural products, of which the majority belong to the terpenoid structural class.[2−5] These plants have a wide distribution across mainland Australia, and, as such, a number of *Eremophila* species were used by Australian Aboriginal people for ceremonial purposes and for treating ailments such as abrasions, colds, headache, and scabies.[2,4−6] More recently, extracts or pure natural products from this genus have been shown to possess antibacterial activity,[6−10] xanthine oxidase inhibition,[11] cyclooxygenase inhibition,[12] and cardioactivity.[13,14]

The species *Eremophila mitchellii* Benth. is native to inland northern New South Wales and Queensland and grows as an aromatic shrub or small tree.[1,5] The natural products isolated from this species include eremophilone[15−17] and its derivatives.[4,18−21] Oil extracts from this plant have been extensively studied due to their termiticidal and insecticidal activities,[21−24] and one study has found the wood oil to possess antimicrobial activity.[25]

In this study, *E. mitchellii* was investigated as a potential source of large quantities (>200 mg) of the previously isolated compound, 14-hydroxy-6,12-muuroloadien-15-oic acid (**6**).[26] This sesquiterpene carboxylic acid was identified after searching the literature as possessing an attractive muurolane[27] scaffold that could be elaborated chemically to generate unique drug-like or molecular probe screening libraries. During the large-scale isolation of **6**, a number of novel and new natural products were also identified, and herein we report the structure elucidation of these compounds.

RESULTS AND DISCUSSION

The air-dried and ground leaves of *E. mitchellii* were extracted exhaustively with sequential washes of *n*-hexane, CH_2Cl_2, and CH_3OH. The CH_2Cl_2 and CH_3OH extracts were combined and fractionated using a diol-bonded silica flash column and a *n*-hexane/EtOAc gradient. Further purification was achieved by either diol-bonded silica HPLC (*i*-PrOH/*n*-hexane) or C_{18}-bonded silica HPLC (CH_3OH/H_2O). This yielded three novel tetracyclic sesquiterpene lactones, namely, mitchellenes A−C (**1**−**3**), and two new sesquiterpene carboxylic acids, mitchellenes D (**4**) and E (**5**). The known compounds 14-hydroxy-6,12-muuroloadien-15-oic acid (**6**), casticin,[28,29] and centaureidin[30,31] were also isolated.



Mitchellene A (**1**) was isolated as an optically active, brown gum. The ^{1}H NMR spectrum of **1** showed six aliphatic signals between δ_H 0.76 and 4.86, one olefinic resonance at δ_H 6.57, three methylene moieties (δ_H 2.37/1.83, 1.39/1.37, and 1.40/1.21), and two methyl resonances (δ_H 0.82 and 1.08) (Table 1). The ^{13}C NMR spectrum of **1** (Table 2) contained signals that indicated the presence of an oxygenated carbon (δ_C 70.9), an olefin (δ_C 136.8 and 127.8), and a carbonyl moiety (δ_C 170.7) within the molecule.

Investigation of the ^{1}H−^{1}H COSY, HSQC, and HMBC spectra permitted the planar structure of **1** to be constructed (Figure 1). HMBC correlations from H-3, H-5, and H-11 to C-1 allowed this carbon to be assigned as δ_C 70.9. The downfield

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Table 1. ^{1}H NMR Data for Mitchellenes A−C (**1**−**3**)[a]

position	**1**[b]	**2**[b]	**3**[b]
1		1.72 (m)	1.63 (m)
2α	1.39 (m)	1.43 (dddd, 13.2, 4.2, 3.6, 3.0)	1.47 (m)
2β	1.35 (ddd, 13.8, 12.0, 4.2)	1.07 (dddd, 13.2, 13.2, 12.6, 3.6)	0.80 (m)
3α	1.21(dddd, 12.0, 11.4, 11.4, 4.2)	0.91 (dddd, 13.2, 12.0, 11.4, 3.6)	0.89 (m)
3β	1.40 (m)	1.65 (m)	1.68 (m)
4	0.76 (dddd, 11.6, 11.4, 11.4, 3.0)	0.79 (dddd, 12.6, 12.3, 12.0, 3.0)	0.81 (m)
5	2.21 (ddd, 11.6, 4.8, 4.2)	1.68 (m)	1.71 (m)
6	3.07 (m)	3.14 (m)	2.83 (dd, 7.8, 7.8)
OH- 7			6.02 (brs)
8	6.57 (ddd, 4.8, 3.0, 2.0)	6.60 (ddd, 4.2, 3.0, 2.4)	5.72 (brdd, 10.2, 3.0, 3.0)
9α	2.37 (dddd, 21.0, 8.4, 3.6, 3.0)	2.24 (dddd, 21.0, 8.4, 4.2, 2.4)	5.94 (brd, 10.2)
9β	1.83 (dddd, 21.0, 9.0, 5.4, 4.8)	1.94 (dddd, 21.0, 8.4, 4.2, 4.2)	
10	1.91 (ddd, 9.0, 8.4, 4.2)	2.11 (m)	2.45 (m)
11	1.08 (s)	0.88 (d, 7.2)	1.00 (d, 7.2)
12	1.63 (ddq, 11.4, 7.2, 7.2)	1.63 (m)	1.46 (m)
13	0.82 (d, 7.2)	0.83 (d, 7.2)	0.90 (d, 7.2)
14	4.86 (dd, 7.2, 7.2)	4.85 (dd, 7.2, 7.2)	4.90 (dd, 7.8, 6.0)

[a] Spectra were recorded in DMSO-d_6 at 30 °C. [b] ^{1}H (mult., J in Hz).

Table 2. ^{13}C NMR Data for Mitchellenes A−E (**1**−**5**) and 14-Hydroxy-6,12-muuroloadien-15-oic Acid (**6**)[a]

position	**1**[b]	**2**[b]	**3**[b]	**4**[b]	**5**[b]	**6**[b]
1	70.9, C	34.5, CH	34.4, CH	34.2, CH	34.8, CH	34.2, CH
2	34.0, CH_2	28.0, CH_2	30.7, CH_2	28.6, CH_2	28.9, CH_2	29.0, CH_2
3	23.9, CH_2	28.8, CH_2	27.7, CH_2	26.7, CH_2	26.4, CH_2	33.8, CH_2
4	41.4, CH	41.4, CH	39.0, CH	40.2, CH	38.0, CH	42.4, CH
5	39.0, CH	45.2, CH	47.0, CH	38.7, CH	40.4, CH	41.2, CH
6	41.7, CH	42.0, CH	47.0, CH	140.1, CH	122.6, CH	142.0, CH
7	127.8, C	127.9, C	69.6, C	131.4, C	138.8, C	129.7, C
8	136.8, CH	136.7, CH	127.4, CH	25.2, CH_2	26.2, CH_2	25.1, CH_2
9	25.4, CH_2	22.6, CH_2	129.9, CH	16.2, CH_2	16.4, CH_2	16.1, CH_2
10	38.9, CH	32.2, CH	36.8, CH	38.4, CH	39.0, CH	38.6, CH
11	27.9, CH_3	18.5, CH_3	18.3, CH_3	19.2, CH_3	19.3, CH_3	19.3, CH_3
12	43.2, CH	43.3, CH	42.5, CH	39.0, CH	38.6, CH	152.4, C
13	11.6, CH_3	11.5, CH_3	11.2, CH_3	9.6, CH_3	9.2, CH_3	107.4, CH_2
14	83.2, CH	82.9, CH	83.9, CH	176.8, C	177.1, C	63.2, CH_2
15	170.7, C	170.6, C	177.6, C	168.5, C	64.9, CH_2	168.4, C

[a] Spectra were recorded in DMSO-d_6 at 30 °C. [b] ^{13}C, mult.

chemical shift of C-1 indicated a hydroxy group was attached to this position.[32] The methyl signal at δ_H 1.08 (H-11) showed HMBC correlations to C-2 (δ_C 34.0) and C-10 (δ_C 38.9), establishing that it was also attached to C-1. A second methyl group resonating at δ_H 0.82 exhibited HMBC correlations with C-4 (δ_C 41.4) and C-14 (δ_C 83.2), allowing it to be positioned at C-12 (δ_C 43.2). The chemical shift of C-14 (δ_C 83.2) indicated that it was next to an oxygen atom,[32] and both H-8 (δ_H 6.57) and H-14 (δ_H 4.86) showed HMBC correlations to a carbon signal at δ_C 170.7 (C-15). This allowed a γ-lactone ring to be constructed, giving the planar tetracyclic ring system shown in Figure 1. The conjugated γ-lactone moiety was supported by the absorption at 1744 cm^{-1} in the IR spectrum of **1**.[32]

The relative configuration of **1** was assigned after analysis of the ROESY spectrum (Figure 1) and ^{1}H−^{1}H coupling constant data. ROESY correlations between H-10/H-5, H-5/H-6, H-6/H-14, and H-14/H-12 established that these protons all had *cis* orientations around the tetracyclic ring system. The ^{1}H−^{1}H coupling constants between H-5 and H-10 ($J_{5\alpha,10\alpha}$ = 4.2 Hz) and between H-6 and H-14 ($J_{6\alpha,14\alpha}$ = 7.2 Hz) further supported the *cis* orientation of these bridgehead protons.[26,33] In a similar

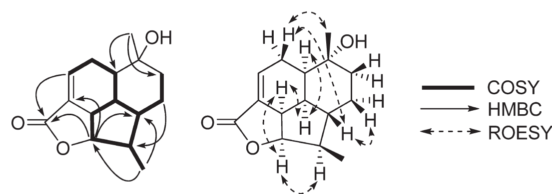


Figure 1. Key COSY, HMBC, and ROESY correlations for **1**.

Table 3. ^{1}H NMR Data for Mitchellenes D and E (4, 5) and 14-Hydroxy-6,12-muuroloadien-15-oic acid (6)[a]

position	4[b]	5[b]	6[b]
1	1.63 (m)	1.61 (m)	1.72 (m)
2α	1.38 (m)	1.37 (m)	1.37 (m)
2β	1.09 (m)	1.09 (brddd, 13.2, 13.2, 11.4)	1.11 (dddd, 13.2, 12.6, 12.6, 3.0)
3α	1.16 (brddd, 13.2, 13.2, 11.4)	1.10 (m)	1.32 (dddd, 12.6, 12.6, 11.4, 3.0)
3β	1.32 (brd, 13.2)	1.28 (m)	1.62 (m)
4	1.84 (brdd, 12.0, 11.4)	1.81 (m)	1.82 (ddd, 11.4, 11.4, 3.0)
5	2.05 (m)	1.82 (m)	2.28 (ddd, 11.4, 5.4, 4.8)
6	6.88 (brd, 4.2)	5.67 (brd, 3.6)	6.77 (brd, 4.8)
8α	2.03 (m)	1.88 (m)	1.98 (m)
8β	2.37 (brdd, 18.6, 4.8)	2.02 (brdd, 18.0, 5.4)	2.35 (brdd, 18.0, 5.4)
9α	1.58 (m)	1.53 (brdd, 12.6, 6.6)	1.57 (brdd, 13.2, 6.6)
9β	1.40 (m)	1.40 (m)	1.35 (m)
10	1.61 (m)	1.62 (m)	1.63 (m)
11	0.88 (d, 6.0)	0.87 (d, 6.6)	0.91 (d, 7.2)
12	2.53 (dq, 7.2, 3.6)	2.62 (dq, 7.2, 3.0)	
13	0.97 (d, 7.2)	0.93 (d, 7.2)	5.05 (s), 4.90 (s)
14			3.80 (d, 15.0), 3.86 (d, 15.0)
15		3.77 (s)	

[a] Spectra were recorded in DMSO-d_6 at 30 °C. [b] ^{1}H (mult., J in Hz).

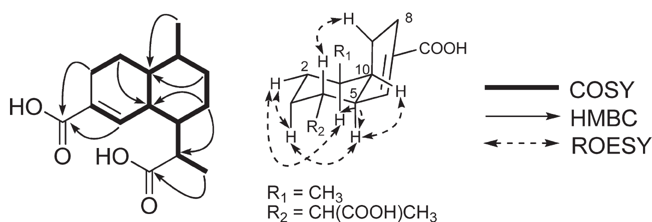


Figure 2. Key COSY, HMBC, and ROESY correlations for **4**.

manner, the ^{1}H−^{1}H coupling constant data between H-4 and H-5 ($J_{4\beta,5\alpha}$ = 11.6 Hz) and H-3α and H-4 ($J_{3\alpha,4\beta}$ = 11.4) supported a *trans* relationship of these protons.[26,33] With the relative configuration determined, structure **1** was assigned to mitchellene A.

The molecular formula, $C_{15}H_{20}O_2$, was assigned to mitchellene B (**2**) on the basis of HRESIMS and NMR data (Table 1). The NMR data of **2** were similar to those of **1**, with the only major differences being that **2** showed one extra proton signal at δ_H 1.72 and an oxygenated quaternary carbon was missing at δ_C 70.9, which was replaced by an upfield signal at δ_C 34.5. The (+)-LRESIMS of **2** showed an ion at m/z 233 $[M + H]^+$, demonstrating a molecular weight difference between **2** and **1** of 16 Da. HMBC correlations from H-3, H-9, and H-10 to the carbon signal at δ_C 34.5, in addition to a HSQC correlation from this carbon to δ_H 1.72, were used to position these signals at C-1 and H-1, respectively. These data indicated that OH-1 in **1** was replaced with a hydrogen atom in **2**. The ROESY spectrum and ^{1}H−^{1}H coupling data for **2** were essentially identical to those of **1**; hence structure **2** was assigned to mitchellene B.

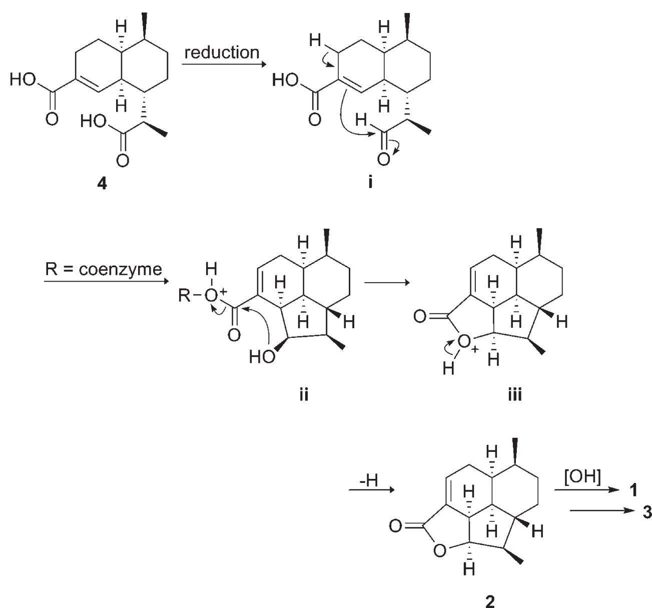
The minor natural product **3** was isolated as a stable opaque gum. Comparison of the NMR and MS data of **3** and **1** indicated that these two molecules were isomers. The ^{1}H NMR spectrum of **3** lacked the methylene signals at H-9 seen in **1**; however, additional signals for two olefinic protons at δ_H 5.94 and 5.72, a hydroxy group resonance at δ_H 6.02, and an aliphatic proton at δ_H 1.63 were observed (Table 1). Carbon C-1 in **3** resonated at δ_C 34.4 compared to δ_C 70.9 in **1**. A correlation in the HSQC spectrum permitted the proton at δ_H 1.63 to be placed at C-1. HMBC correlations were seen between H-6/H-8/H-9/H-14 and the oxygenated carbon at C-7 (δ_C 69.6). HMBC correlations from the hydroxy proton to C-6, C-7, C-8, and C-15 allowed this group to be placed at C-7. HMBC correlations from H-8 (δ_H 5.72) to C-6, C-10, and C-15 and from C-9 (δ_H 5.94) to C-1, C-5, and C-7 indicated that the endocyclic double bond seen in **1** had migrated to C-8/C-9 in **3**. In a similar manner to **1** and **2**, the relative configuration of **3** was assigned on the basis of ROESY and ^{1}H−^{1}H coupling constant data and found to be the same as that of mitchellenes A and B. With the relative configuration determined, structure **3** was assigned to mitchellene C.

Compound **4** was isolated as a light brown gum and assigned the molecular formula $C_{15}H_{22}O_4$ on the basis of HRESIMS and NMR data (Table 3). The ^{1}H NMR spectrum of **4** showed five aliphatic signals between δ_H 1.61 and 2.53, one olefinic signal at δ_H 6.88, four methylene signals (δ_H 2.37/2.03, 1.58/1.40, 1.38/1.09, and 1.32/1.16), and two methyl signals (δ_H 0.97 and 0.88). The ^{13}C NMR spectrum of **4** (Table 2) suggested that the molecule contained an olefin (δ_C 140.1 and 131.4) and two carbonyl moieties (δ_C 168.5 and 176.8).[32]

Analysis of the ^{1}H−^{1}H COSY, HSQC, and HMBC spectra allowed for the planar bicyclic structure of sesquiterpene **4** to be constructed (Figure 2). The proton at H-4 (δ_H 1.84) showed a HMBC correlation to the methyl group at H-13 (δ_H 0.97). It was also found that H-4 (δ_H 1.84), H-12 (δ_H 2.53), and H-13 (δ_H 0.97) all showed HMBC correlations to a carbon at δ_C 176.8, allowing a carboxyl group to be positioned at C-14. HMBC correlations from both H-6 and H-8 to C-15 (δ_C 168.5) enabled a second carboxylic acid side chain to be positioned at C-7 (δ_C 131.4).

The relative configuration of **4** was obtained following investigation of the ROESY spectrum and from the ^{1}H−^{1}H coupling constants. This assignment initially proved difficult due to

Scheme 1. Proposed Biogenetic Pathway for Mitchellenes A–C (1–3)



overlapping or broadened signals in the ^{1}H NMR spectrum; however, comparison with data obtained on **1**–**3** and **6** identified that **4** also possessed a muurolane skeleton.[27] The relative configuration of the methyl group at C-13 was assigned as having a β-orientation on biosynthetic grounds, as it was postulated that **4** is a biosynthetic intermediate for mitchellenes A–C. Hence structure **4** was assigned to mitchellene D.

The NMR data of mitchellene E (**5**) (Tables 1 and 2) were very similar to those of **4**, with the only differences being that **5** contained an additional oxygenated methylene signal (δ_H 3.77, δ_C 64.9) and was missing one carboxylic acid ^{13}C NMR resonance. HMBC correlations from δ_H 3.77 to C-8 (δ_C 26.2) and C-6 (δ_C 122.6) suggested this $-OCH_2-$ moiety was attached to C-7. These data, along with a difference of 14 Da in the (+)-LRESIMS of **4** compared to **5**, indicated that the carboxylic acid at C-15 in **4** was replaced with a dihydro derivative in **5**.

After analysis of the ROESY spectrum and ^{1}H–^{1}H coupling constants, the relative configuration of **5** was found to be the same as that of **4** and **6**. As in **4**, the relative configuration of the methyl group at C-13 was assigned as having a β-orientation on the basis of biosynthetic reasoning. Structure **5** was thus assigned to mitchellene E. Compound **5** was found to be a new natural product; however, a diastereoisomer of this molecule has been produced in a degradation study undertaken on the related natural product arteannuin H by Sy et al.[34]

The previously isolated natural product, 14-hydroxy-6,12-muuroloadien-15-oic acid, was assigned as **6** after 1D/2D NMR and MS data analysis and by comparison with literature values.[26] Compound **6** has only been partially characterized by spectroscopic methods. Herein we report the ^{1}H NMR (Table 3), ^{13}C NMR (Table 2), IR, UV, and HRESIMS data for **6**.

The known compounds casticin and centaureidin were identified after 1D/2D NMR and MS data analysis and by comparison with literature values.[28–31]

The biosynthesis of bicyclic sesquiterpenes such as **4**–**6** has been well studied, and several detailed investigations have been reported in the literature. These natural products are cyclized from the common sesquiterpene precursor farnesyl diphosphate.[35–38] Mitchellenes A–C (**1**–**3**) are the first tetracyclic sesquiterpene lactones to be reported. The only other natural products with a similar tetracyclic lactone skeleton are the liverwort diterpenes pallavicinolides A–C, which were proposed to be generated biosynthetically via a labdane-based diterpenoid pathway.[39,40]

Scheme 1 shows a proposed biosynthetic pathway for **1**–**3** starting from natural product **4**. Similar pathways for the generation of **1**–**3** would also be possible from compounds **5** and **6**. Reduction of the C-12 carboxylic acid in **4** to form the aldehyde **i**, followed by a proton abstraction at C-8 and intramolecular cyclization, generates the first five-membered ring as shown in **ii**. Attack by the hydroxy group at the conjugated carboxylic acid, aided by an unspecified coenzyme, allows for the formation of the lactone ring. Loss of hydrogen then gives **2**. Addition of a hydroxy group at either C-1 or C-7 facilitates the formation of **1** and **3**, respectively.

■ EXPERIMENTAL SECTION

General Experimental Procedures. Optical rotations were recorded on a JASCO P-1020 polarimeter. IR and UV spectra were recorded on a Bruker Tensor 27 spectrophotometer and a JASCO V-650 UV/vis spectrophotometer, respectively. NMR spectra were recorded at 30 °C on either a Varian 500 or 600 MHz Unity INOVA spectrometer. The latter spectrometer was equipped with a triple-resonance cold probe. The ^{1}H and ^{13}C chemical shifts were referenced to the solvent peaks for DMSO-d_6 at δ_H 2.49 and δ_C 39.5. LRESIMS were recorded on a Mariner time-of-flight spectrometer equipped with a Gilson 215 eight probe injector. HRESIMS were recorded on a Bruker Apex III 4.7 T Fourier transform ion cyclotron resonance mass spectrometer. An Edwards Instrument Company Bioline orbital shaker was used for plant extractions. For the HPLC work, a Waters 600 pump equipped with a Waters 966 PDA detector and Gilson 715 liquid handler were used. Alltech Davisil diol-bonded silica, 30–40 μm, 60 Å, or Alltech Davisil C_{18}-bonded silica, 35–75 μm, 150 Å, was used for preadsorption work. Either a YMC-pack diol 5 μm 120 Å (20 mm × 150 mm) column or a ThermoElectron C_{18} Betasil 5 μm 143 Å (21.2 mm × 150 mm) column was used for semipreparative HPLC separations. All solvents used for chromatography, $[\alpha]_D$, UV, IR, and MS were Lab-Scan HPLC grade, and the H_2O was Millipore Milli-Q PF filtered.

Plant Material. The leaves of *Eremophila mitchellii* were collected from Currawinya National Park, QLD, Australia, in March 1996. Collection and identification were undertaken by P. Forster and G. Guymer from the Queensland Herbarium. A voucher specimen (AQ603041) has been deposited at the Queensland Herbarium, Brisbane, Australia.

Extraction and Isolation. The air-dried and ground leaves of *E. mitchellii* (27.5 g) were extracted exhaustively with *n*-hexane (250 mL), CH_2Cl_2 (2 × 250 mL), and CH_3OH (2 × 250 mL). All CH_2Cl_2/CH_3OH extractives were combined to yield a dark brown gum (4.94 g). This crude extract was divided into ~500 mg portions and purified by a diol-bonded silica flash column (35 × 130 mm) using a 20% stepwise gradient from *n*-hexane to EtOAc followed by CH_3OH. The 40% EtOAc/60% *n*-hexane fraction was further purified by diol-bonded silica semipreparative HPLC. Isocratic conditions of *n*-hexane were held for the first 10 min, followed by a linear gradient to 20% *i*-PrOH/80% *n*-hexane in 40 min, then isocratic conditions of 20% *i*-PrOH/80% *n*-hexane for 10 min, all at a flow rate of 9 mL/min. Sixty fractions (1 min each) were collected. Fractions 25 and 26 contained impure **2** (56.5 mg) and were further purified by semipreparative diol-bonded HPLC. Isocratic conditions of *n*-hexane were held for the first

5 min, followed by a linear gradient to 15% *i*-PrOH/75% *n*-hexane in 40 min, then a linear gradient to 20% *i*-PrOH/80% *n*-hexane in 5 min, followed by isocratic conditions of 20% *i*-PrOH/80% *n*-hexane for 10 min, all at a flow rate of 9 mL/min. Sixty fractions (1 min each) were collected. Compound **2** eluted in fractions 41 and 42 (28.3 mg, 0.103% dry wt).

The 60% EtOAc/40% *n*-hexane fraction from the diol flash column was further purified by C_{18}-bonded silica semipreparative HPLC at a flow rate of 9 mL/min and isocratic conditions of 10% CH_3OH/90% H_2O for 10 min, followed by a linear gradient to CH_3OH in 40 min, then isocratic conditions of CH_3OH for 10 min. Sixty fractions (1 min each) were collected. Fraction 45 contained semipure **3** (5.4 mg) and was further purified by semipreparative diol-bonded silica HPLC, using isocratic conditions of *n*-hexane for 10 min, followed by a linear gradient to 20% i-PrOH/80% *n*-hexane in 40 min, then isocratic conditions of 20% *i*-PrOH/80% *n*-hexane for 10 min all at a flow rate of 9 mL/min. Sixty fractions (1 min each) were collected. Fraction 29 contained pure **3** (2.2 mg, 0.008% dry wt).

The 80% EtOAc/20% *n*-hexane and EtOAc fractions from the initial diol flash column were combined and further purified using the same diol-bonded silica column and conditions as for the 40% EtOAc/60% *n*-hexane fraction, above. This yielded **4** in fraction 31 (9.6 mg, 0.035% dry wt), **6** in fractions 32 and 33 (201.7 mg, 0.734% dry wt), **5** in fraction 36 (8.4 mg, 0.031% dry wt), **1** in fractions 37 and 38 (10.5 mg, 0.038% dry wt), casticin in fractions 47−49 (9 mg, 0.033% dry wt), and centaureidin in fractions 53 and 54 (9.7 mg, 0.035% dry wt).

Mitchellene A (1): brown gum; $[\alpha]^{25}_{D}$ −45 (*c* 0.060, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 205 (4.20), 214 (4.22), 289 (3.10) nm; IR ν_{max} (KBr) 3420, 1744, 1457, 1375, 1278, 1202, 1059, 983, 956 cm^{-1}; ^{1}H and ^{13}C NMR data (DMSO-d_6), see Tables 1 and 2; (+)-LRESIMS *m/z* 271 (100) $[M + Na]^+$, 249 (100) $[M + H]^+$; (+)-HRESIMS *m/z* 271.1300 ($C_{15}H_{20}O_3Na$ $[M + Na]^+$ requires 271.1305).

Mitchellene B (2): opaque gum; $[\alpha]^{25}_{D}$ −58 (*c* 0.080, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 204 (4.54), 216 (4.61) nm; IR ν_{max} (KBr) 2920, 1761, 1667, 1454, 1379, 1347, 1287, 1197, 1166, 1141, 1124, 1062, 1012, 981, 962, 952 cm^{-1}; ^{1}H and ^{13}C NMR data (DMSO-d_6), see Tables 1 and 2; (+)-LRESIMS *m/z* 255 (100) $[M + Na]^+$, 233 (100) $[M + H]^+$; (+)-HRESIMS *m/z* 255.1355 ($C_{15}H_{20}O_2Na$ $[M + Na]^+$ requires 255.1356).

Mitchellene C (3): opaque gum; $[\alpha]^{25}_{D}$ −58 (*c* 0.107, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 205 (4.36), 232 (3.74) nm; IR ν_{max} (KBr) 3382, 1764, 1594, 1455, 1378, 1357, 1289, 1199, 1094, 1033, 989 cm^{-1}; ^{1}H and ^{13}C NMR data (DMSO-d_6), see Tables 1 and 2; (+)-LRESIMS *m/z* 271 (100) $[M + Na]^+$; (+)-HRESIMS *m/z* 271.1311 ($C_{15}H_{20}O_3Na$ $[M + Na]^+$ requires 271.1305).

Mitchellene D (4): light brown gum; $[\alpha]^{26}_{D}$ −34 (*c* 0.120, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 203 (3.92), 219 (4.04) nm; IR ν_{max} (KBr) 3390, 1694, 1644, 1454, 1416, 1380, 1269, 1224, 1180 cm^{-1}; ^{1}H and ^{13}C NMR data (DMSO-d_6), see Tables 2 and 3; (+)-LRESIMS *m/z* 289 (100) $[M + Na]^+$; (+)-HRESIMS *m/z* 289.1402 ($C_{15}H_{22}O_4Na$ $[M + Na]^+$ requires 289.1410).

Mitchellene E (5): light brown gum; $[\alpha]^{26}_{D}$ −30 (*c* 0.093, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 203 (4.11), 225 (3.70) nm; IR ν_{max} (KBr) 3409, 1722, 1709, 1693, 1679, 1513, 1380, 1238, 1224, 994 cm^{-1}; ^{1}H and ^{13}C NMR data (DMSO-d_6), see Tables 2 and 3; (+)-LRESIMS *m/z* 275 (100) $[M + Na]^+$; (+)-HRESIMS *m/z* 275.1629 ($C_{15}H_{24}O_3Na$ $[M + Na]^+$ requires 275.1618).

14-Hydroxy-6,12-muuroloadien-15-oic acid (6): light green gum; $[\alpha]^{26}_{D}$ −154 (*c* 0.120, $CHCl_3$); UV (CH_3OH) λ_{max} (log ε) 203 (4.68), 220 (4.75) nm; IR ν_{max} (KBr) 3340, 3074, 2628, 1685, 1644, 1531, 1453, 1432, 1268, 1219, 1086, 1051, 903 cm^{-1}; ^{1}H NMR and ^{13}C NMR data (DMSO-d_6), see Tables 2 and 3; (+)-LRESIMS *m/z* 273 (100) $[M + Na]^+$; (+)-HRESIMS *m/z* 273.1464 ($C_{15}H_{22}O_3Na$ $[M + Na]^+$ requires 273.1461).

■ ASSOCIATED CONTENT

Supporting Information. 1D and 2D NMR spectra of mitchellenes A−E (**1**−**5**) and 14-hydroxy-6,12-muuroloadien-15-oic acid (**6**). This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Author

*Tel: +61-7-3735-6043. Fax: +61-7-3735-6001. E-mail: r.davis@griffith.edu.au.

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