Palmadorins A−C, Diterpene Glycerides from the Antarctic Nudibranch *Austrodoris kerguelenensis*[||]

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Received October 1, 2009

The nudibranch *Austrodoris kerguelenensis* is distributed widely around the Antarctic coast and continental shelves. Earlier collections from McMurdo Sound and the Weddell Sea shelf have afforded a suite of diterpene glyceride esters, a compound class implicated as a chemical defense in nudibranchs. The present chemical investigation of *A. kerguelenensis* collected near Palmer Station on the Western Antarctic Peninsula has revealed additional examples, palmadorins A−C (**1**−**3**), as the first three members of a new series of clerodane diterpenes. In this paper we describe their isolation, structure elucidation, and stereochemical analysis using a combination of one- and two-dimensional NMR spectroscopy and wet chemical methods.

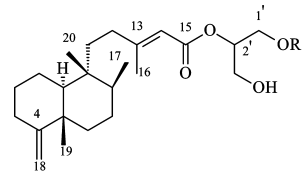
The absence of body armor (shell) on nudibranchs, or sea slugs, taken with their limited mobility and high visibility makes them seemingly vulnerable to predation. Yet they are seldom attacked by predators because they typically employ toxic chemicals as their defense.[1,2] Some nudibranchs derive their defensive metabolites from their diet, such as sponges or soft corals. Such sequestered chemicals can be stored in specialized spherical dorsal glands called mantle dermal formations, from which they can be easily discharged.[3] On the other hand, some nudibranchs are capable of *de novo* biosynthesis of their chemical defenses.[1,2]

Austrodoris kerguelenensis (Nudibranchia: Doridiidae) is one of the most common nudibranch species found in Antarctic shallow and deep waters.[4] These white to yellow mollusks, varying in size from 2 to 15 cm, have recently been the subject of taxonomic analysis, leading at least one authority to classify them with the *Doris* genus.[5] However, many researchers continue to use the more traditional genus *Austrodoris*,[6] the classification used herein for consistency with the history of the mollusks in the chemical literature, as described below.

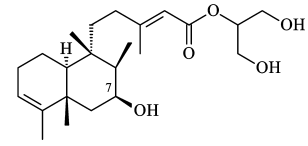
McMurdo Sound and Weddell Sea collections of *A. kerguelenensis* have been extensively studied, leading to the description of several suites of diterpene glycerides including *ent*-labdane, halimane, clerodane, and isocopalane skeletons, along with several nor-sesquiterpenes.[7−13] Nudibranch diterpene glycerides from other dorid nudibranchs, including *Archidoris montereyensis*, *Archidoris odhnery*, and *Doris verrucosa*, have a rich history of chemical, pharmacological, and ecological study.[1] In particular, dorid chemistry has been documented as examples of *de novo* biosynthesis,[14,15] and *A. kerguelenensis* appears unexceptional in this regard.[16] The biological role of diterpene esters in nudibranchs, which are present in the mantle tissue, is linked to providing protection; they are toxic toward freshwater fish[17] and deter feeding in marine fish.[16,18] Diterpene diacylglycerides of *A. kerguelenensis* extracted from mantle tissue deterred feeding of the common Antarctic predatory seastar *Odontaster validus*.[16] Some diterpene 1,2-diacylglcerols display potent activation of protein kinase C and are active in regenerative tests with the freshwater hydrozoan *Hydra vulgaris*.[17] In this paper, we report a new series of clerodane-type diterpenoids (**1**−**3**) from *A. kerguelenensis* collected near the U.S. Antarctic Program's Palmer Station on the Western Antarctic Peninsula.

Results and Discussion

In the austral summers of 2000 and 2001, *A. kerguelenensis* were routinely collected from shallow-water habitats, to −40 m, among the Anvers Island archipelago, near Palmer Station, Antarctica. They were frozen immediately and subsequently freeze-dried and extracted with $CHCl_3$. The $CHCl_3$ extract of the nudibranchs was fractionated by flash chromatography on silica gel. Further purification by HPLC on silica gel, then C_{18} silica gel, yielded palmadorin A (**1**) (24 mg, 0.96% dry wt), palmadorin B (**2**) (7 mg, 0.28% dry wt), and palmadorin C (**3**) (7 mg 0.28% dry wt).



Palmadorin A (**1**, R = H)
Palmadorin B (**2**, R = Ac)



Palmadorin C (**3**)

HRFABMS analysis of palmadorin A (**1**) provided a molecular formula of $C_{23}H_{38}O_4$ ($[M + H]^+$ *m/z* 379.2842). The ^{13}C NMR spectrum of **1** (Table 1) displayed only 22 carbon signals, including four olefinic carbons and one quaternary carbon at δ_C 166.7, the latter of which was assigned as an ester-equivalent carbonyl. In addition to 15 high-field resonances, the sp^3-carbon-bearing oxygen region had two resonances, at δ_C 74.4 and 62.7. The signal at δ_C 62.7 exhibited unusually high intensity, indicative of two coincident signals, completing the carbon distribution supported by the mass spectrometric data.

The ^1H NMR spectrum (Table 1) of palmadorin A (**1**) showed a distinct sharp doublet at δ_H 4.48 (H_2-18), integrating for two protons, which exhibited a gHSQC correlation with the carbon signal at δ_C 102.5 (C-18), suggesting the presence of an exomethylene. H_2-18 displayed strong HMBC correlations (Figure 1) to the quaternary carbons C-4 (δ_C 160.4) and C-5 (δ_C 40.0) as well as methylene C-3 (δ_C 33.0), defining the lower portion of the A ring, with the exocyclic double bond at C-4. The HMBC correla-

[||] Dedicated to the late Dr. John W. Daly of NIDDK, NIH, Bethesda, Maryland, and to the late Dr. Richard E. Moore of the University of Hawaii at Manoa for their pioneering work on bioactive natural products.

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10.1021/np900617m
Published on Web 02/01/2010

Table 1. ^{1}H and ^{13}C NMR Data of Palmadorins A (**1**), B (**2**), and C (**3**) in $CDCl_3$[a]

	palmadorin A (**1**)			palmadorin B (**2**)			palmadorin C (**3**)		
position	δ_C	δ_H (J (Hz))	gHMBC	δ_C	δ_H (J (Hz))	gHMBC	δ_C	δ_H (J (Hz))	gHMBC
1	21.6	1.43 (1H, m) 1.48 (1H, m)		21.8	1.42 (m) 1.46 (m)		18.2	1.54 (1H, m) 1.58 (1H, m)	
2	28.6	1.23 (1H, m) 1.87 (1H, m)	1, 3	29.0	1.21 (m) 1.86 (m)	3	27.0	1.98 (1H, m) 2.08 (1H, m)	
3	33.0	2.09 (1H, m) 2.27 (1H, m)	1, 4, 5, 18, 2, 4, 18	34.4	2.09 (m) 2.27 (m)	2, 5, 18, 2, 18	120.0	5.13 (1H, m)	
4	160.4			160.7			145.2		
5	40.0			40.4			37.8		
6	37.2	1.50 (1H, m) 1.58 (1H, m)	5, 8, 5, 7, 10, 19	37.7	1.47 (m) 1.57 (m)	8, 19, 5, 7, 8, 10, 19	43.1	1.39 (1H, m) 2.10 (1H, m)	4, 5, 10, 19, 5, 7, 8, 9, 10, 19
7	27.4	1.44 (1H, m) 1.47 (1H, m)	5, 8, 5, 8	27.8	1.44 (m) 1.47 (m)		73.9	4.02 (1H, m)	
8	36.7	1.39 (1H, m)		37.0	1.39 (m)		39.5	1.51 (1H, m)	17
9	39.3			39.7			38.5		
10	48.7	1.03 (1H, m)	2, 5, 19, 20	49.1	1.03 (m)	5, 6, 7, 8	46.9	1.37 (1H, m)	1, 4, 5, 6, 9, 19, 20
11	36.1	1.33 (1H, m) 1.44 (1H, m)	8, 9, 10, 12, 13, 20, 8, 9, 10, 12, 13, 20	36.4	1.33 (m) 1.44 (m)	9, 10, 12, 8, 9, 10, 12, 20	37.6	1.39 (1H, m) 1.51 (1H, m)	5, 10, 19, 8
12	34.6	1.85 (1H, m) 1.96 (1H, m)	11, 13, 14, 16, 11, 13, 14, 16	34.9	1.85 (m) 1.96 (m)	11, 16, 11, 16	35.1	1.93 (1H, m) 1.98 (1H, m)	16
13	163.7			163.9			163.4		
14	114.3	5.68 (br s)	12, 15, 16	114.7	5.68 (br s)	12, 15, 16	114.8		12, 15, 16
15	166.7			166.8			167.0	5.70 (1H, s)	
16	19.5	2.14 (3H, s)	12, 13, 14	19.8	2.14 (3H, s)	12, 13, 14	19.7	2.16 (3H, s)	12, 13, 14
17	16.0	0.79 (3H, d, 6)	7, 8, 9	16.4	0.78 (3H, d, 6.5)	7, 8, 9	12.7	1.01 (3H, d, 7.5)	7, 8
18	102.5	4.48 (2H, d, 1.5)	3, 4, 5	102.9	4.48 (2H, d, 1)	3, 4, 5	18.3	1.60 (3H, s)	3, 4, 5
19	20.8	1.02 (3H, s)	4, 5, 6, 10	22.3	1.02 (3H, s)	4, 5, 6, 10	22.0	1.27 (3H, s)	4, 5, 6, 10
20	18.0	0.71 (3H, s)	8, 9, 10, 11	18.5	0.72 (3H, s)	8, 9, 10, 11	20.3	0.99 (1H, s)	8, 9, 10, 11
1′	62.7	3.83 (2H, d, 4.5)	2′, 3′	62.9	4.26 (2H, m)	2′, 3′ $\underline{C}O_2CH_3$	63.1	3.82 (2H, d, 4.5)	
2′	74.4	4.91 (1H, m)	1′, 3′, 15	72.0	5.06 (1H, m)	15, 1′	74.8	4.91 (1H, m)	
3′	62.7	3.83 (2H, d, 4.5)	1′, 2′	62.2	3.72 (2H, d, 4)	1′, 2′,$\underline{C}O_2CH_3$	63.1	3.82 (2H, d, 4.5)	
$CO_2\underline{C}H_3$				21.2	2.05 (3H, s)				
$\underline{C}O_2CH_3$				171.2					

[a] For ^{1}H, 500 MHz, ppm [integration, mult., J (Hz)]; for ^{13}C, 125 MHz.

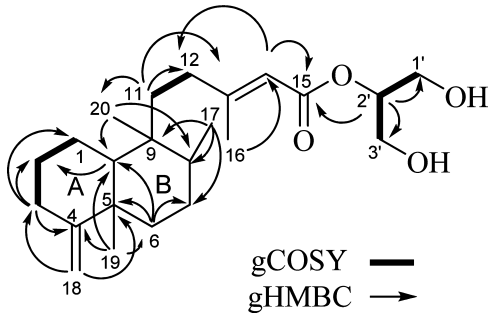

Figure 1. Key gHMBC and gCOSY correlations of palmadorin A (**1**).

tions of H-3a (δ_H 2.09) with C-4, C-5, and C-18 were consistent with this assignment. Ring A could be completed by the addition of the two methylenes adjacent to C-3 on the basis of HMBC correlations of H-3a to C-1 (δ_C 21.6), H-3b (δ_H 2.27) to C-2 (δ_C 28.6), and H-10 (δ_H 1.03) to C-2. The attachment of the singlet methyl group H_3-19 (δ_H 1.02) to the A/B ring junction at C-5 was based on HMBC correlations of H_3-19 with C-4, C-5, and C-10.

Two methyl groups, doublet H_3-17 (δ_H 0.79) and singlet H_3-20 (δ_H 0.71), showed HMBC correlations to C-8 (δ_C 36.7) and C-9 (δ_C 39.3), while H_3-20 alone correlated with C-10 (δ_C 48.7) and H_3-17 alone correlated to C-7 (δ_C 27.4), suggesting that they are located on adjacent carbons, C-8 and C-9, on ring B. Both δ_H 1.44 (H-7a) and δ_H 1.47 (H-7b) exhibited HMBC connectivities to C-8 and C-5, completing the assignment of the B ring of palmadorin A (**1**).

The HMBC spectrum of palmadorin A (**1**) further established a broad singlet at δ_H 5.68 (H-14) correlating to the ester carbonyl (C-15, δ_C 166.7), as well as methylene (C-12, δ_C 34.6) and singlet methyl (C-16, δ_C 19.5) groups. The latter methyl signal, H_3-16 (δ_H 2.14), in turn exhibited HMBC correlations to C-13 (δ_C 163.7), C-14 (δ_C 114.3), and C-15. These assignments are consistent with a trisubstituted olefin attached to an ester carbonyl. Both H-12a (δ_H 1.85) and H-12b (δ_H 1.96) showed distinct HMBC correlations to C-13, C-14, C-16, and C-11 (δ_C 36.1). Further, H-11a (δ_H 1.33) and H-11b (δ_H 1.44) showed HMBC connectivity to carbons C-8, C-9, C-10, C-12, C-13, and C-20 (δ_C 18.0), unambiguously defining the side chain as originating at C-9.

The remaining three carbons could be assigned as a glyceride moiety on the basis of the gCOSY correlation of two chemically equivalent protons H-1′/H-3′ (δ_H 3.83) with H-2′ (δ_H 4.91) and the HMBC correlations of H-1′/H-3′ with C-2′ (δ_C 74.4). The HMBC correlation observed between H-2′ and the ester carbonyl, C-15 (δ_C 166.7), established that it is connected to the terpenoid residue via an ester linkage between C-15 and C-2′.

The axial orientation of H_3-19 and H_3-20 was confirmed by the observation of strong nuclear Overhauser enhancement (NOE) between them when protons of both methyl groups were irradiated separately (Figure 2). Neither NOE irradiation of H_3-20 nor the ROESY spectrum could identify proximity to H-10, leading to the assignment of that proton as axial and defining it as a *trans*-decalin. H_3-20 showed further correlation with H-11b, H-12b, and H_3-17, establishing all three decalin methyl groups as on the same face. That H_2-12 and H-14 display ROESY correlations defines the C-13 olefin as *E*.

To establish the absolute configuration of palmadorin A (**1**), ozonolysis was conducted, resulting in diketone **4** (Figure 3). The ^{13}C NMR spectrum of **4** showed two quaternary carbon signals, at δ_C 216.3 (C-4) and 209.0 (C-13), indicating the presence of two ketone groups. The former carbon resonance showed distinct HMBC connectivites with H-3a (δ_H 2.23) and H_3-16 (δ_H 1.15), resulting in its assignment as C-4 (Figure 3). The latter quaternary carbon was correlated to proton signals H_3-14 (δ_H 2.16) and H-12b (δ_H 2.34), providing further evidence for the presence of another ketone

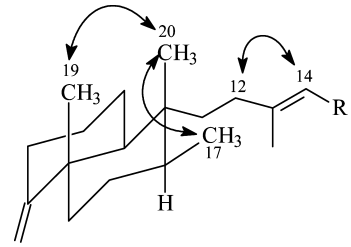

Figure 2. Key NOE and ROESY correlations of palmadorin A (**1**).

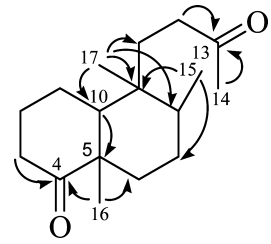

Figure 3. Key gHMBC correlations of diketone **4**.

at C-13. On the basis of the HMBC correlations of the methyl groups H_3-15 (δ_H 0.82) and H_3-17 (δ_H 0.73) and methine proton H-10 (δ_H 1.20), the structure of the product was confirmed as **4**. A negative Cotton effect ($\Delta\varepsilon$ −0.2) in the circular dichroism spectrum of **4** is consistent with an absolute configuration for palmadorin A of 5*S*, 8*S*, 9*R*, and 10*S* based on the analogy with *trans*-10-methyl-1-decalone.[19]

Analysis of the HRFABMS data of palmadorin B (**2**) gave an accurate mass consistent with the molecular formula $C_{25}H_{40}O_5$ ([M + H]$^+$ *m/z* 421.2964). The ^{13}C NMR spectrum of compound **2** exhibited 25 carbon signals in agreement with the HRFABMS data. A complete structure elucidation of palmadorin B was accomplished by an extensive analysis of its HMBC, HSQC, and COSY data and indicated that it has the same planar structure as palmadorin A, for the rings A and B and the side chain terminating at the C-15 carbonyl.

The ester carbonyl at δ_C 166.8 (C-15) of compound **2** exhibited a HMBC correlation to the multiplet at δ_H 5.06 (H-2′). Two doublet signals observed in the ^{1}H NMR spectrum (Table 1), δ_H 3.72 (H-3′) and 4.26 (H-1′), showed COSY correlations to H-2′, establishing the presence of the glyceride moiety. H-1′ showed HMBC correlations not only with C-2′ (δ_C 72.0) and C-3′ (δ_C 62.2) but also with a second ester carbonyl at δ_C 171.2. The ester carbonyl displayed a HMBC correlation to a methyl singlet at δ_H 2.05 (COOC$\underline{H}_3$), defining the presence of an acetyl group. All these observations are consistent with a glyceride moiety attached to a carbonyl via an ester linkage at C-2′ that is acetylated at C-1′.

The carbon and proton NMR spectroscopic data of the A and B rings of palmadorins A (**1**) and B (**2**) showed significant similarities, suggesting that they have similar relative configurations. The stereochemical analysis of palmadorin B, using ROESY and one-dimensional selective NOE experiments, indicated that it had the identical *trans*-decalin configuration to that of palmadorin A. Hence, by analogy with the stereochemical features of palmadorin A, the absolute configuration of palmadorin B was assigned as 5*S*, 8*S*, 9*R*, and 10*S* based on similar NMR data, optical rotation, and biosynthetic considerations.

Palmadorin C (**3**) displayed a prominent nominal mass spectrometric peak at *m/z* 377.4, indicative of [M − H_2O]$^+$, in the LRFABMS. The [M + H]$^+$ peak at *m/z* 395.4 supported the molecular formula of $C_{23}H_{38}O_5$. As seen in the ^{13}C NMR spectrum of palmadorin A (**1**), the spectrum from **3** showed 22 signals, due to the coincidence of the hydroxymethylene carbons. An additional hydroxymethine was observed at δ_C 73.9.

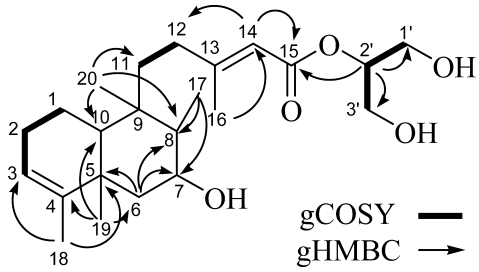

Figure 4. Key HMBC and COSY correlations of palmadorin C (**3**).

Analysis of the HMBC data (Figure 4) from palmadorin C (**3**) showed that the exomethylene group present in the A ring of palmadorins A (**1**) and B (**2**) at C-18 has been rearranged to a trisubstituted double bond between C-3 (δ_C 120.0) and C-4 (δ_C 145.2). The vinylic proton at δ_H 5.13 (H-3) did not show HMBC correlations. H_3-18 (δ_H 1.60) displayed HMBC correlations to C-3, C-4, and C-5 (δ_C 37.8), which affirmed the above assignment. An additional singlet methyl group, H_3-19 (δ_H 1.27), a methyl group on C-5, could be assigned on the basis of HMBC correlations to C-4, C-5, C-6 (δ_C 43.1), and C-10 (δ_C 46.9). The connectivity from C-3 to C-10 via two methylenes C-1 (δ_C 18.2) and C-2 (δ_C 27.0) was confirmed by COSY correlations of H-1a (δ_H 1.54), H-2a (δ_H 1.98), and H-3.

Attachment of two methyl groups, H_3-17 (δ_H 1.01) and H_3-20 (δ_H 0.99), at C-8 (δ_C 39.5) and C-9 (δ_C 38.5), respectively, was confirmed by their HMBC correlations. The former correlated to C-7 (δ_C 73.9) and C-8, whereas the latter correlated to C-8, C-9, C-10, and C-11 (δ_C 37.6). Assignment of a secondary hydroxy to C-7 was supported by the HMBC correlation of H-6b (δ_H 2.10) with C-19 (δ_H 22.0), C-7, and C-8. In addition, H-10 (δ_H 1.37) showed HMBC correlations to C-1, C-4, C-5, C-6, C-19, and C-20 (δ_C 20.3), reaffirming the above assignments.

Further analysis of HMBC and COSY data of palmadorin C (**3**) indicated the occurrence of the same side chain observed in palmadorins A (**1**) and B (**2**), which terminates giving rise to an ester carbonyl at C-15 (δ_C 167.0). On the basis of the HMBC correlation observed between H-2′ (δ_H 4.91) and this ester carbonyl, a glyceride moiety could be attached to the diterpene via an ester linkage between H-2′ and C-15. This assignment is in agreement with the observation of COSY correlations between the multiplet at δ_H 4.91 (H-2′) and the 4H doublet at δ_H 3.82 (H-1′/H-3′). Thus, palmadorin C (**3**) shares the same glyceride substitution pattern found on palmadorins A (**1**) and B (**2**). This is relatively uncommon among diterpene glycerides.[4]

The relative configuration of palmadorin C (**3**) was studied by ROESY spectroscopy. The olefinic proton H-3 (δ_H 5.13) displayed strong ROESY correlations to H-2b (δ_H 2.08) and the vinylic methyl at δ_H 1.60 (H_3-18). A correlation between the singlet methyl groups at H_3-19 (δ_H 1.27) and H_3-20 (δ_H 0.99) indicated they both were on the same face of the decalin system. Similarly, H-7 (δ_H 4.02) showed spatial proximity with H-8 (δ_H 1.51) and H-6a (δ_H 1.39) by means of ROESY correlations, and H-6a in turn was correlated to H-10 (δ_H 1.37). H-6a, H-7, H-8, and H-10 were therefore all oriented toward the opposite face of the decalin compared to H_3-19 and H_3-20 (Figure 5).

Mosher's method[20] was used to determine the configuration of C-7, taking advantage of the secondary hydroxy group. However, prior to that, the primary hydroxy groups in the glyceride moiety needed to be protected. This was accomplished by selectively acetylating the primary alcohols, leaving the secondary alcohol available for Mosher's method (Figure 6). With C-7 determined as *S*, the absolute configurations of the remaining centers were assigned from the relative configuration described above, resulting in the absolute configuration of palmadorin C (**3**) assigned as 5*S*, 7*S*, 8*R*, 9*S*, and 10*S*.

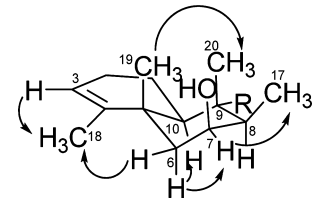

Figure 5. Key ROESY correlations of palmadorin C (**3**).

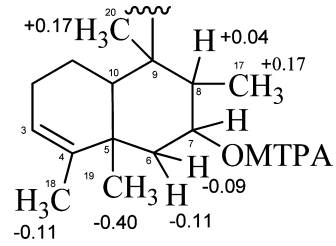

Figure 6. Mosher's analysis of palmadorin C diacetate.

Tropical plants have been a rich source of clerodane-type diterpenes, and *A. kerguelenensis*, collected from the Ross Sea, has been the source of one.[9] The palmadorin diterpenoic acid parent can be found among those tropical plant diterpenes.[21,22] *A. kerguelenensis*, a bright white to yellow shell-less mollusk, stands out from its mostly colorless natural environment, seemingly advertising itself to its predators;[16] palmadorins may offer chemical protection toward some predators.

Experimental Section

General Experimental Procedures. Instrumentation and routine procedures follow those of our recently reported work.[23] CD spectra were obtained with an Aviv Instruments model 215 CD spectrometer. HPLC was performed on a Shimadzu LC-8A multisolvent delivery system connected to a Shimadzu SPD-10A UV−vis tunable absorbance detector and using a Phenomenex SiO_2 column (5 μm, 10 mm) for normal-phase chromatography and a YMC-Pack ODS-AQ C-18 column for reversed-phase chromatography. EM Science silica gel 60 of 230−400 mesh was used in flash column chromatography. TLC was carried out on Whatman K6F silica gel 60 Å TLC plates with 0.25 mm thickness. They were visualized by spraying with 5% phosphomolybdic acid in EtOH and heating.

Animal Material. *Austrodoris kerguelenensis* was collected by scuba from the vicinity of Palmer Station, Antarctica (64°46′ S, 64°03′ W), in March and April 2000 and November and December 2001. Taxonomic identification was conducted by one of the authors (K.B.I.), and voucher specimens are held at the University of South Florida.

Extraction and Isolation. Following freeze-drying, the nudibranchs were extracted 3× with $CHCl_3$ for 24 h. After concentration under reduced pressure, a gummy, reddish-brown solid (2.5 g) was obtained. This $CHCl_3$ extract was further purified into 12 fractions by elution of increasing polarity gradient of EtOAc in *n*-hexane on silica gel. Fractions 7−11 showed ^{1}H NMR signals indicative of terpene glycerides. Further purification of fraction 9 by HPLC on C_{18} by isocratic elution of MeCN−H_2O, 7.5:2.5, afforded palmadorin A (**1**, 24 mg). Separation of fraction 7 by HPLC, first on SiO_2 using EtOAc−*n*-hexane, 4:6, and then on C_{18}, with MeCN−H_2O, 7.5:2.5, gave palmadorin B (**2**, 7 mg). Fraction 10 was separated by HPLC first on SiO_2 using EtOAc−*n*-hexane, 6:4, and subsequently on C_{18} with isocratic elution of MeCN−H_2O, 7.5:2.5, to obtain palmadorin C (**3**, 7 mg).

Palmadorin A (1): colorless oil; $[\alpha]^{25}_D$ +18 (*c* 0.05, MeOH); UV (MeOH) λ_{max} (ε) 215 (1001), 248 (499), 266 (484) nm; IR (thin film) 3400 (br), 2969, 2864, 1712, 1640, 1488, 1382, 1281, 1147, 1100, 1040, 975 cm^{-1}; ^{1}H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 379.3 (52, [M + H]$^+$), 361.2 (10); HRFABMS *m/z* 379.2842 [M + H]$^+$ ($C_{23}H_{39}O_4$ requires 379.2848).

Palmadorin B (2): colorless oil; $[\alpha]^{25}_D$ +24 (*c* 0.05, MeOH); UV (MeOH) λ_{max} (ε) 214 (1063), 248 (537), 264 (524) nm; IR (thin film) 3360 (br), 2933, 2359, 1718, 1642, 1448, 1383, 1219, 1145, 110, 909

cm^{-1}; ^{1}H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 421.4 (36, [M + H]$^+$), 379.4 (8), 321.3 (8), 287.3 (90); HRFABMS *m/z* 421.2964 [M + H]$^+$ ($C_{25}H_{41}O_5$ requires 421.2954).

Palmadorin C (3): colorless oil; $[\alpha]^{25}_D$ +8 (*c* 0.05, MeOH); UV (MeOH) λ_{max} (ε) 216 (1002), 248 (635) nm; IR (thin film) 3389 (br), 2840, 1648, 1408, 1228, 1219, 1109, 1015, 894 cm^{-1}; ^{1}H and ^{13}C NMR data, see Table 1; LRFABMS (+) *m/z* 395.4 (3, [M + H]$^+$), 377.4 (15, [M + H − H_2O]$^+$), 309.1 (14), 195.1 (15), 153.1 (45), 135 (45).

Ozonolysis of Palmadorin A (1). Compound **1** (5 mg, 0.01 mmol) was dissolved in 2 mL of CH_2Cl_2 and allowed to react with O_3 for 25 min at −80 °C. Dimethylsulfide (5 drops) was then added and allowed to react for 1.5 h while allowing the reaction mixture to warm to room temperature. The solvent was evaporated under reduced pressure to obtain 8 mg of a crude product as a colorless solid. This crude product was purified by chromatography over silica using 30% EtOAc/hexane, yielding the ozonolysis product **4** as a white, amorphous solid (1 mg, 0.004 mmol, 40%).

Palmadorin A Oxidation Product (4): colorless solid; $[\alpha]^{25}_D$ +0.6 (*c* 0.05, MeOH); CD (MeOH) $\Delta\varepsilon$ −0.2; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H 2.58 (1H, m, H-3b), 2.34 (1H, m, H-12b), 2.23 (1H, m, H-3a), 2.16 (3H, s, H_3-14), 2.13 (1H, m, H-12a), 1.72 (1H, m, H-11b), 1.60 (1H, m, H-6b), 1.56 (1H, m, H-6a), 1.53 (1H, m, H-7b), 1.46 (1H, m, H-2b), 1.44 (1H, m, H-11a), 1.28 (1H, m, H-7a), 1.24 (1H, m, H-2a), 1.20 (1H, m, H-10), 1.15 (3H, s, H_3-16), 0.82 (3H, d, 6.8, H_3-15), 0.73 (3H, s, H_3-17); ^{13}C NMR (125 MHz, $CDCl_3$) δ_C 216.3 (C-4), 209.0 (C-13), 50.0 (C-10), 40.6 (C-9) 39.4 (C-5), 37.7 (C-12), 37.6 (C-3), 36.6 (C-8), 33.1 (C-6), 30.8 (C-11), 30.6 (C-14), 26.5 (C-2), 26.5 (C-7), 18.8 (C-15), 17.00 (C-17), 15.5 (C-16); EIMS *m/z* 175 (22), 122 (10), 95 (20), 43 (100), 41 (45), 28 (30).

Acetylation of Palmadorin C (3). Palmadorin C (5 mg, 0.01 mmol) was dissolved in CH_2Cl_2 (1 mL) and reacted with Ac_2O (30 μL, 0.025 mmol) in the presence of DMAP (3 mg) and Et_3N (30 μL) for 24 h at 25 °C. The reaction was quenched by the addition of a few drops of MeOH. Upon evaporation of the solvent by reduced pressure, the crude product was separated by chromatography over silica gel (EtOAc−hexane gradient elution) to obtain palmadorin C diacetate (**5**) (6 mg, 0.01 mmol, 100%).

Palmadorin C diacetate (5): colorless solid; $[\alpha]^{25}_D$ +12 (*c* 0.05, MeOH); IR (thin film) 3389 (br), 2834, 1648, 1408, 1228, 1210, 1040, 1109, 1015, 894 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H 5.65 (1H, s, H-14), 5.26 (1H, m, H-2′), 4.23 (4H, m, H-1′/3′), 4.03 (1H, m, H-7), 2.15 (3H, s, H_3-16), 2.08 (1H, m, H-6b), 2.06 (3H, s, $COOCH_3$), 2.05 (1H, m, H-2b), 1.98 (1H, m, H-2b), 1.93 (1H, m, H-12b), 1.90 (1H, m, H-12a), 1.60 (3H, s, H_3-18), 1.52 (1H, m, H-8), 1.48 (1H, m, H-11a), 1.38 (1H, m, H-6a), 1.38 (1H, m, H-10), 1.37 (1H, m, H-11), 1.27 (3H, s, H_3-19), 1.02 (3H, d, 7, H_3-17), 1.00 (3H, s, H_3-20); ^{13}C NMR (125 MHz, $CDCl_3$) δ_C 170.8 ($COOCH_3$), 165.7 (C-15), 163.3 (C-13), 145.2 (C-4), 120.0 (C-3), 114.7 (C-14), 73.9 (C-7), 68.2 (C-2′), 62.7 (C-1′/C-3′), 54.8 (OCH_3), 46.9 (C-10), 43.2 (C-6), 39.5 (C-8), 38.0 (C-9), 37.8 (C-5), 37.5 (C-11), 35.1 (C-12), 26.9 (C-2), 21.8 (C-19), 20.2 (C-20), 19.7 (C-16), 12.7 (C-17); LRESIMS (−) *m/z* 477.2 (78, [M − 1]$^+$), 478.2 (20, [M]$^+$).

Preparation of *R*-MTPA Esters of Palmadorin C Diacetate (6). Palamadorin A diacetate (**5**) (1 mg, 0.002 mmol) was dissolved in CH_2Cl_2 (300 μL) and allowed to react with *S*-MTPA Cl (50 mg, 0.07 mmol) in the presence of Hunig's base (100 μL) and DMAP (1 mg) for 48 h. Conversion of the starting material to the products was monitored by TLC. Evaporation of the solvent gave a crude product of 20 mg. Further purification of this mixture by chromatography on silica gel (EtOAc−hexane gradient elution) gave compound **7** (1 mg, 0.001 mmol, 50%).

Palmadorin C diacetate *R*-MTPA (6): colorless solid; $[\alpha]^{25}_D$ −5 (*c* 0.02, MeOH); IR (thin film) 3342 (br), 2905, 1658, 1408, 1340, 1235, 1048, 1109, 1020, 870 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H (7.59 (1H, m, Ph), 7.53 (2H, m, Ph), 7.37 (2H, m, Ph), 5.61 (1H, s, H-14), 5.24 (1H, m, H-2′), 4.22 (4H, m, H-1′/3′), 5.44 (1H, m, H-7), 3.59 (3H, s, OCH_3), 2.13 (3H, s, H_3-16), 2.07 (1H, m, H-2b), 2.05 (3H, s, $COOCH_3$), 2.05 (1H, m, H-2b), 1.98 (1H, m, H-2b), 2.06 (1H, m, H-6b), 1.90 (1H, m, H-2a), 1.90 (1H, m, H-12b), 1.86 (1H, m, H-12a), 1.65 (1H, m, H-8), 1.54 (3H, s, H_3-18), 1.48 (1H, m, H-6a), 1.46 (1H, m, H-11b), 1.35 (1H, m, H-10), 1.26 (1H, m, H-11a), 0.75 (3H, d, 6.9, H_3-17), 0.63 (3H, s, H_3-20) 0.94 (3H, s, H_3-19); ^{13}C NMR (125 MHz, $CDCl_3$) δ_C 170.8 ($COOCH_3$), 167.0 (COOMTP), 165.7 (C-15), 163.3 (C-13), 144.1 (C-4), 129.7 (Ph), 128.6 (Ph), 128.0 (Ph), 127.6 (Ph), 127.6 (Ph), 120.0 (C-3), 114.7 (C-14), 73.9 (C-7), 68.2 (C-2′), 62.7 (C-1′/C-3′), 46.6 (C-10), 43.2 (C-6), 39.5 (C-8), 38.1 (C-9), 37.5 (C-11), 37.4 (C-5), 35.1 (C-12), 26.9 (C-2), 20.8 (C-19), 20.2 (C-20), 19.7 (C-16), 12.7 (C-17); HRESIMS *m/z* 717.3220 [M + Na]$^+$ ($C_{37}H_{49}O_9F_3Na$ requires 717.3226).

Preparation of *S*-MTPA Esters of Palmadorin C Diacetate (7). Palmadorin A diacetate (3 mg, 0.006 mmol) was dissolved in CH_2Cl_2 (500 μL) and allowed to react with *R*-MTPA Cl (50 mg, 0.07 mmol) in the presence of Hunig's base (100 μL) and DMAP (1 mg) for 48 h. Conversion of the starting material to the products was monitored by TLC. Evaporation of the solvents gave a crude product of 20 mg. Further purification of this mixture by chromatography on silica gel (EtOAc/hexane gradient elution) gave compound **6** (1.5 mg, 0.0022 mmol, 40%).

Palmadorin C Diacetate *S*-MTPA (7): colorless solid; $[\alpha]^{25}_D$ +10 (*c* 0.05, MeOH); IR (thin film) 3401 (br), 2825, 1662, 1510, 1340, 1235, 1032, 1006, 860 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H 7.53 (2H, m, Ph), 7.35 (3H, m, Ph), 5.62 (1H, s, H-14), 5.25 (1H, m, H-2′), 4.22 (4H, m, H-1′/3′), 5.35 (1H, m, H-7), 3.59 (3H, s, OCH_3) 2.13 (3H, s, H_3-16), 2.05 (3H, s, $COOCH_3$), 2.05 (1H, m, H-2b), 1.98 (1H, m, H-2b), 1.97 (1H, m, H-6b), 1.90 (1H, m, H-12b), 1.88 (1H, m, H-12a), 1.43 (3H, s, H_3-18), 1.69 (1H, m, H-8), 1.48 (1H, m, H-11a), 1.37 (1H, m, H-6a), 1.37 (1H, m, H-11), 1.31 (1H, m, H-10), 0.92 (3H, d, H_3-17), 0.80 (3H, s, H_3-20), 0.53 (3H, s, H_3-19); ^{13}C NMR (125 MHz, $CDCl_3$) δ_C 170.8 ($COOCH_3$), 166.9 (COOMTP), 165.7 (C-15), 162.7 (C-13), 145.2 (C-4), 129.7 (Ph), 128.5 (Ph), 127.36 (Ph), 120.6 (C-3), 114.9 (C-14), 78.0 (C-7), 68.3 (C-2′), 62.7 (C-1′/C-3′), 55.8 (OCH_3), 46.4 (C-10), 40.4 (C-6), 39.1 (C-8), 38.2 (C-9), 37.3 (C-5), 37.1 (C-11), 34.9 (C-12), 26.8 (C-2), 21.8 (C-19), 20.2 (C-20), 19.6 (C-16), 12.0 (C-17); HRESIMS *m/z* 717.3220 [M + Na]$^+$ ($C_{37}H_{49}O_9F_3Na$ requires 717.3226).

Acknowledgment. We are indebted to a number of S-022 divers for collecting specimens, including D. Martin, K. Peters, J. Hubbard, C. Petrie, and A. Mahon. We would like to acknowledge the assistance of the staff of Raytheon Polar Services Company and the Antarctic support services of the National Science Foundation for providing logistical support. This research was facilitated by funding from the Office of Polar Programs of the National Science Foundation (OPP-9814538 and -0125181 to C.D.A. and J.B.M., OPP-9901076 and -0125152 to B.J.B.).

Supporting Information Available: ^{1}H, ^{13}C, gCOSY, gHSQC, gHMBC, and ROESY NMR spectra of **1**−**3** and the CD spectrum of **4**. This material is available free of charge via the Internet at http://pubs.acs.org.

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NP900617M