

Article

## Sesquiterpene and Acetogenin Derivatives from the Marine Red Alga *Laurencia okamurai*

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**Abstract:** In addition to 13 known compounds, four new bisabolane sesquiterpenes, okamurenes A–D (1–4), a new chamigrane derivative, okamurene E (5), and a new C<sub>12</sub>-acetogenin, okamuragenin (6), were isolated from the marine red alga *Laurencia okamurai*. The structures of these compounds were determined through detailed spectroscopic analyses. Of these, okamurenes A and B (1 and 2) are the first examples of bromobisabolane sesquiterpenes possessing a phenyl moiety among *Laurencia*-derived sesquiterpenes, while okamuragenin (6) was the first acetogenin aldehyde possessing a C<sub>12</sub>-carbon skeleton. Each of the isolated compounds was evaluated for the brine shrimp (*Artemia salina*) lethal assay and 7-hydroxylaurene displayed potent lethality with LD<sub>50</sub> 1.8 μM.

**Keywords:** marine alga; *Laurencia okamurai*; bisabolane sesquiterpene; C<sub>12</sub>-acetogenin; brine shrimp lethality

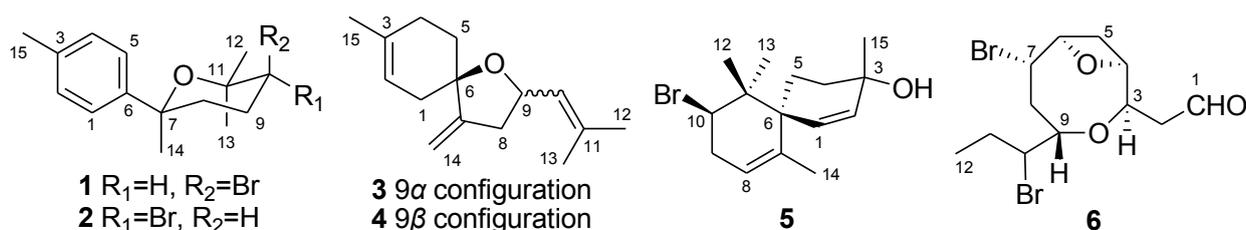
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### 1. Introduction

Marine red algae of the genus *Laurencia* are prolific sources of diversified secondary metabolites, predominantly sesquiterpenoids, diterpenoids, and nonterpenoid C<sub>15</sub>-acetogenins [1]. The red alga *Laurencia okamurai*, widely distributed along the coast of China, mainly yields sesquiterpenes and C<sub>15</sub>-acetogenins [2]. These compounds, with structurally diverse skeletons, have attracted much

attention for total syntheses [3] as well as chemotaxonomic research [4–6]. In the past five years, we have systematically conducted chemical investigation towards eight *Laurencia* species, which have resulted in the isolation of more than 30 new compounds [2,7–11]. In the course of our phytochemical studies on *Laurencia okamurai*, a new, rearranged chamigrane sesquiterpene, laurenokamurin, was previously characterized [10]. Continuous effort on the chemical investigation of this algal species collected from Weihai coastline resulted in the isolation and identification of five new sesquiterpenes, okamurenes A–E (**1–5**), one new C<sub>12</sub>-acetogenin, okamuragenin (**6**) (Figure 1), as well as nine known sesquiterpenes and four known C<sub>15</sub>-acetogenins. We present herein the isolation, structure elucidation, and bioactivity of these compounds.

**Figure 1.** Structures of the isolated new compounds **1–6** from *L. okamurai*.



## 2. Results and Discussion

### Structure Elucidation of the New Compounds

Okamurene A (**1**) was obtained as a colorless oil and its molecular formula was established by HRESIMS to be C<sub>15</sub>H<sub>21</sub>BrO, corresponding to five degrees of unsaturation. The <sup>1</sup>H NMR spectrum of **1** (Table 1) exhibited resonances for a *para*-substituted phenyl unit, four methyl groups, and a brominated or oxygenated methine group. There were also four signals for two diastereotopic methylene protons. The <sup>13</sup>C NMR and DEPT spectroscopic data (Table 1) revealed the presence of 15 carbon signals including six aromatic carbons (corresponding to a *para*-substituted phenyl unit) and nine aliphatic carbons (corresponding to four methyls, two methylenes, one brominated methine, and two oxygenated quaternary carbons). These units accounted for 4 degrees of unsaturation, requiring one additional ring to be present in **1**.

**Table 1.** <sup>1</sup>H- and <sup>13</sup>C-NMR data of compounds **1** and **2** in CDCl<sub>3</sub> <sup>a</sup>.

No.	<b>1</b> (CDCl <sub>3</sub> )		<b>2</b>	
	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$
1/5	7.34, d (8.0)	124.8, CH	7.34, d (8.1)	126.0, CH
2/4	7.11, d (8.0)	128.7, CH	7.12, d (8.1)	128.6, CH
3		136.1, C		136.4, C
6		146.0, C		143.2, C
7		74.6, C		74.4, C
8 <sub>eq</sub>	2.16, m	34.1, CH <sub>2</sub>	2.56, m	36.0, CH <sub>2</sub>
8 <sub>ax</sub>	2.10, m		2.18, m	
9 <sub>eq</sub>	2.28, m	28.2, CH <sub>2</sub>	2.27, m	29.4, CH <sub>2</sub>
9 <sub>ax</sub>	2.25, m		1.82, m	

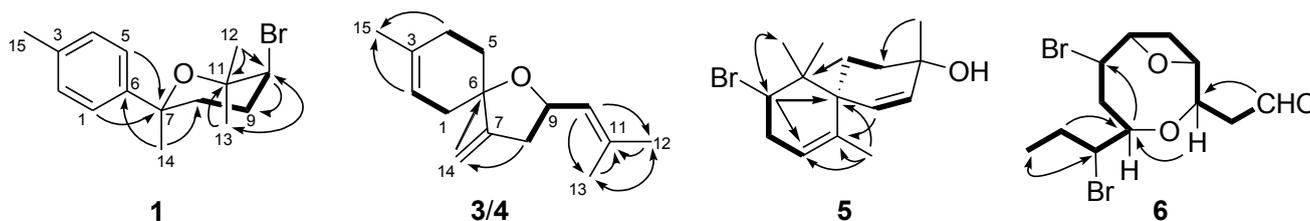
**Table 1.** Cont.

10	4.05, dd (7.9, 4.4)	59.1, CH	4.04, dd (12.1, 4.1)	59.0, CH
11		75.2, C		76.4, C
12	1.47, s	27.8, CH <sub>3</sub>	1.35, s	22.5, CH <sub>3</sub>
13	1.14, s	29.4, CH <sub>3</sub>	0.78, s	30.8, CH <sub>3</sub>
14	1.50, s	31.8, CH <sub>3</sub>	1.36, s	35.8, CH <sub>3</sub>
15	2.23, s	20.9, CH <sub>3</sub>	2.34, s	21.0, CH <sub>3</sub>

<sup>a</sup> Measured at 500 MHz for <sup>1</sup>H and 125 MHz for <sup>13</sup>C.

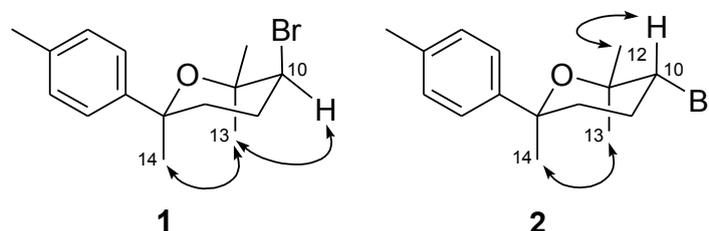
The structure of the non-phenyl portion of **1** was determined by analysis of 2D NMR data (<sup>1</sup>H–<sup>1</sup>H COSY, HSQC, and HMBC). The <sup>1</sup>H–<sup>1</sup>H COSY experiment established the connectivity for a –CH<sub>2</sub>–CH<sub>2</sub>–CH– unit (C-8 through C-10, Figure 2). The C-10 methine of this unit was connected to CH<sub>3</sub>-12 and CH<sub>3</sub>-13 via the oxygenated quaternary carbon C-11 (δ<sub>C</sub> 75.2) as evidenced by the observed HMBC correlations from the methyl protons H<sub>3</sub>-12 and H<sub>3</sub>-13 to C-10 and C-11, while the C-8 methylene was linked to the CH<sub>3</sub>-14 via the oxygenated quaternary carbon C-7 (δ<sub>C</sub> 74.6) as supported by the observed HMBC correlation from the methyl protons H<sub>3</sub>-14 to C-8 (Figure 2). Given the fact that only one oxygen atom existed in the structure, the linkage of C-7/O/C-11 could be constructed, leading to the formation of a tetrahydropyran moiety, which accounted for the remaining degree of unsaturation. Thus, the planar structure of **1** was assigned.

**Figure 2.** Key COSY (bold lines) and HMBC (arrows) correlations for compounds **1**, **3/4**, **5**, and **6**.



Analysis of the proton coupling constants and NOESY data enabled assignment of the relative configuration of **1**. The appearance of the bromomethine proton H-10 as a double doublet, with coupling constants of 7.9 and 4.4 Hz, suggesting the equatorial orientation of H-10 for **1**. In the NOESY spectrum, NOE correlations of H<sub>3</sub>-13 with both H-10 and H<sub>3</sub>-14 placed the methyl groups CH<sub>3</sub>-13 and CH<sub>3</sub>-14 on the same face (axial or pseudoaxial) of the tetrahydropyran ring (Figure 3). On the basis of the above evidence, the structure of **1** was determined, and the trivial name okamurene A was assigned.

**Figure 3.** Key NOESY correlations for compounds **1** and **2**.



The  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectroscopic data of okamurene B (**2**), an isomer of **1** as established by HRESIMS data, were very similar to those of **1** except for some chemical shift variations of signals corresponding to the C-8, C-9, and C-12 through C-14 (Table 1). Therefore, compound **2** was presumed to be a stereoisomer of **1**. Detailed analysis of the  $^1\text{H}$  and  $^{13}\text{C}$  NMR data as well as  $^1\text{H}$ – $^1\text{H}$  COSY and HMBC correlations supported the conclusion that **2** possesses the same planar structure as **1**. However, comparisons of the  $J$ -value and NOESY data of **2** with those of **1** revealed a difference in relative configuration at C-10. A *trans*-diaxial  $J$ -value for  $\text{H}_{\text{ax}}\text{-10}$  and  $\text{H}_{\text{ax}}\text{-9}$  (12.1 Hz) indicated an equatorial orientation for the Br-atom at C-10. The NOE correlation from H-10 to  $\text{H}_3\text{-12}$  in the NOESY spectrum indicated an equatorial face of  $\text{CH}_3\text{-12}$ , while the NOE correlation from  $\text{H}_3\text{-13}$  to  $\text{H}_3\text{-14}$  placed these two methyl groups in axial orientation (Figure 3). Based on the above data, the structure of compound **2** was identified and it was named okamurene B.

Okamurenes C (**3**) and D (**4**) were obtained as a colorless oily mixture in a 2:1 ratio, as indicated by the  $^1\text{H}$  NMR spectrum. Attempts to separate the mixture by various CC steps using different solvent systems failed. On the other hand, there is no conjugated system in compounds **3** and **4**, making these compounds unsuitable for HPLC separation using the available UV detector. A similar unseparable mixture containing (9*S*)- and (9*R*)-2-bromo-3-chloro-6,9-epoxybisabola-7(14),10-diene from *L. saitoi* was previously described [11]. Most of the NMR signals for compounds **3** and **4** were duplicated or overlapped. By detailed analysis of 1D and 2D NMR data, their structures were determined to be C-9 epimer of 6,9-epoxybisabola-2,7(14),10-triene.

The molecular formula of compounds **3** and **4** were determined to be  $\text{C}_{15}\text{H}_{22}\text{O}$  (five degrees of unsaturation) on the basis of HRESIMS data. Examination of the  $^1\text{H}$  and  $^{13}\text{C}$  NMR data (Table 2) revealed that they resembled 9*S*- and/or 9*R*-2-bromo-3-chloro-6,9-epoxybisabola-7(14),10-diene [11], except for the presence of signals for a trisubstituted vinyl group at C-2 and, accordingly, the lack of the resonances due to a brominated methine at C-2 and a chlorinated quaternary carbon at C-3 [11]. The chemical shifts for the vinyl carbons at  $\delta_{\text{C}}$  119.1/119.0 (C-2) and 133.4/133.7 (C-3) as well as for one of the neighboring methylene groups C-4 ( $\delta_{\text{C}}$  27.7/28.0) in the  $^{13}\text{C}$  NMR spectrum of **3** and **4** were very similar to those reported for 8-bromo-chamigra-1,11(12)-dien-9-ol (with C-2 at  $\delta_{\text{C}}$  119.4, C-3 at  $\delta_{\text{C}}$  132.9, and C-4 at  $\delta_{\text{C}}$  27.5) [12], and these data strongly supported the presence of the trisubstituted vinyl group at C-2 in **3/4**. These data indicated that compounds **3** and **4** were the dehalogenated derivatives corresponding to 9*S*- and/or 9*R*-2-bromo-3-chloro-6,9-epoxybisabola-7(14),10-diene [11]. The  $^1\text{H}$ – $^1\text{H}$  COSY and HMBC correlations (Figure 2) further verified the planar structures of **3/4** to be 6,9-epoxybisabola-2,7(14),10-triene. Assignment of the relative configuration at C-6 by NOESY experiment is not applicable for compounds **3** and **4** since there is no proton around C-6 in the tetrahydrofuran ring. However, the C-6 relative configuration was tentatively assigned to be the same as that of 9*S*- and/or 9*R*-2-bromo-3-chloro-6,9-epoxybisabola-7(14),10-diene based on the similar NMR data around the chiral center, as well as on biogenetic consideration [11].

Okamurene E (**5**), a colorless oil, was shown to have the molecular formula of  $\text{C}_{15}\text{H}_{23}\text{BrO}$  by the interpretation of HRESIMS data. The IR absorption at  $3401\text{ cm}^{-1}$  exhibited the presence of a hydroxyl group. The  $^1\text{H}$  NMR spectrum (Table 2) delineated four methyl singlets, one double doublet ascribable to an oxygenated/halogenated methine, and one multiplet and two doublets attributable to three olefinic protons. The  $^{13}\text{C}$  and DEPT NMR spectra (Table 2) displayed four methyls, three methylenes, four methines, and four quaternary carbons. Compared to the reported NMR data for 10-bromo-7 $\alpha$ ,

8 $\alpha$ -epoxychamigr-1-en-3-ol [12], compound **5** exhibited no resonances for the epoxy moiety in the NMR spectra. Instead, it showed additional signals at  $\delta_{\text{H}}$  5.23 (H-8) and  $\delta_{\text{C}}$  139.5 (C-7) and 120.8 (C-8) for a trisubstituted vinyl group, which was positioned at C-7 based on the observed HMBC correlations from H-14 to C-6, C-7, and C-8. Further analysis of the  $^1\text{H}$ - $^1\text{H}$  COSY and HMBC correlations (Figure 2) confirmed the structure of **5** as 10-bromo-1,7-chamigradien-3-ol. The relative configurations at C-3, C-6, and C-10 of **5** were deduced to be same as those of 10-bromo-7 $\alpha$ , 8 $\alpha$ -epoxychamigr-1-en-3-ol [12] by the NOESY correlation between H-5 and H-10 as well as by their similar NMR data.

**Table 2.**  $^1\text{H}$ - and  $^{13}\text{C}$ -NMR data of compounds **3–6** in  $\text{CDCl}_3$  <sup>a</sup>.

No.	<b>3</b>		<b>4</b>		<b>5</b>		<b>6</b>	
	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (J in Hz)	$\delta_{\text{C}}$
1	2.25, m	38.1, CH <sub>2</sub>	2.18, m	36.8, CH <sub>2</sub>	5.54, d (10.4)	131.2, CH	9.80, br s	199.3, CH
2a	5.34, m	119.1, CH	5.34, m	119.0, CH	5.85, d (10.4)	136.5, CH	2.67, dd (17.5, 6.2)	42.4, CH <sub>2</sub>
2b							3.06, dd (17.3, 7.9)	
3		133.4, C		133.7, C		67.4, C	4.34, t (6.5)	72.7, CH
4a	1.93, m	27.7, CH <sub>2</sub>	1.93, m	28.0, CH <sub>2</sub>	1.56, m	28.5, CH <sub>2</sub>	4.65, dd (8.7, 5.0)	81.6, CH
4b	2.22, m		2.22, m		1.99, m			
5a	1.58, m	31.5, CH <sub>2</sub>	1.66, m	34.5, CH <sub>2</sub>	1.78, m	36.3, CH <sub>2</sub>	2.75, m	21.7, CH <sub>2</sub>
5b	1.82, m		1.75, m				2.91, m	
6		80.9, C		80.8, C		47.4, C	4.97, m	80.9, CH
7		156.3, C		156.5, C		139.5, C	4.21, m	50.4, CH
8a	2.38, m	40.1, CH <sub>2</sub>	2.38, m	40.3, CH <sub>2</sub>	5.23, m	120.8, CH	2.42, dd (14.1, 5.8)	41.7, CH <sub>2</sub>
8b	2.71, dd (15.7, 9.7)		2.61, dd (15.6, 9.5)				2.61, m	
9	4.63, m	71.8, CH	4.63, m	72.8, CH	2.58, m	36.1, CH <sub>2</sub>	4.50, dd (7.4, 3.5)	74.4, CH
10	5.22, m	126.0, CH	5.22, m	126.2, CH	4.64, dd (10.6, 6.4)	61.4, CH	3.80, dt (11.5, 3.5)	64.1, CH
11a		136.2, C		135.5, C		41.6, C	1.77, m	27.4, CH <sub>2</sub>
11b							1.88, m	
12	1.69, s	18.2, CH <sub>3</sub>	1.70, s	18.3, CH <sub>3</sub>	1.02, s	18.1, CH <sub>3</sub>	1.07, t (7.7)	12.8, CH <sub>3</sub>
13	1.71, s	25.8, CH <sub>3</sub>	1.71, s	25.8, CH <sub>3</sub>	1.11, s	26.3, CH <sub>3</sub>		
14a	4.78, br s	103.5, CH <sub>2</sub>	4.78, br s	103.8, CH <sub>2</sub>	1.57, s	21.9, CH <sub>3</sub>		
14b	4.90, br s		4.91, br s					
15	1.66, s	23.4, CH <sub>3</sub>	1.66, s	23.4, CH <sub>3</sub>	1.31, s	28.8, CH <sub>3</sub>		

<sup>a</sup> Measured at 500 MHz for  $^1\text{H}$  and 125 MHz for  $^{13}\text{C}$ .

Okamuragenin (**6**), isolated as a colorless oil, was assigned the molecular formula  $\text{C}_{12}\text{H}_{18}\text{Br}_2\text{O}_3$  on the basis of HRESIMS, consistent with three degrees of unsaturation. The IR spectrum exhibited strong absorptions at 2762 and 1728  $\text{cm}^{-1}$ , indicating the existence of an aldehyde group. In accordance with the IR signals, the  $^1\text{H}$  and  $^{13}\text{C}$  NMR data (Table 2) also indicated the presence of an aldehyde group at  $\delta_{\text{H}}$  (9.80, H-1) and  $\delta_{\text{C}}$  199.3 (CH, C-1). The  $^1\text{H}$ - $^1\text{H}$  COSY spectrum revealed that the aldehyde

group was extended to a straight spin system consisting of six methines, four methylenes, and terminated by a methyl group (Figure 2). Compound **6** was deduced to be bicyclic, since no other unsaturated functionalities were indicated by the NMR data (Table 2). The connectivity of C-3/O/C-9 was deduced by the correlation from H-3 to C-9 in the HMBC spectrum (Figure 2). Taking into account the downfield chemical shifts of C-4 ( $\delta_C$  81.6) and C-6 ( $\delta_C$  80.9) and the calculated 3 degrees of unsaturation, C-4 and C-6 had to be linked through an oxygen atom. Finally, the two remaining Br-atoms indicated by the molecular formula could only be located at C-7 and C-10 based on the chemical shifts [13]. The relative configuration was determined by NOESY experiment. The same orientation of CH<sub>2</sub>-2, H-4, and H-9 was evidenced by the NOE correlations of H-2 to H-4 and H-9, while H-9 was *syn* to H-7 based on the NOE correlation between them. The above data established the structure of **6**, trivially named okamuragenin.

In addition to the six new compounds, the other nine sesquiterpenes including isobromocuparene [14], 7-hydroxylaurene [15], laurene [16], filiformin [17], debromofiliformin [18], 6-bromo-filiformin [19], deoxyprepacifenol [20], 2-bromo-3-chloro-2,7-epoxy-9-chamigren-8 $\alpha$ -ol [11], and 2,10-dibromo-3-chloro-7-chamigren-9-ol [21], together with four C<sub>15</sub>-acetogenins including 3*E*, 12*Z*-laurediol [22], neolaurallene [23], *E*-stereoisomer of neoisoprelaurefucin [24], and 3*Z*-laurentin [25], were all identified by comparison of their spectral data with those previously reported.

The isolated compounds were evaluated for the brine shrimp (*Artemia salina*) lethal activity [26,27]. Among them, 7-hydroxylaurene was found to possess potent lethality with LD<sub>50</sub> 1.8  $\mu$ M, which is more active than that of 7-hydroxylaurene acetate, allolaurinterol acetate, and laurene [12]. Analysis of structure-activity relationship showed that the 7-hydroxyl group in laurene sesquiterpenes may play a key role in the brine shrimp toxicity, and the activity reduced significantly after acetylation. The above data suggested that 7-hydroxylaurene may be a potent chemical defensive agent with cytotoxicity, although the hatchability test was not performed [27]. The other tested compounds only displayed moderate or weak activity (data not shown).

### 3. Experimental Section

#### 3.1. General

IR spectra were measured on a Nicolet NEXUS 470 FT-IR spectrophotometer. Optical rotations were recorded on an Atago Polax-L polarimeter. UV spectra were determined on a Spectrumlab 54 UV-visible spectrophotometer. HRESIMS were run on a VG Autospec 3000 mass spectrometer. 1D and 2D NMR spectra were obtained at 500 and 125 MHz for <sup>1</sup>H and <sup>13</sup>C, respectively, on a Bruker Advance 500 MHz NMR spectrometer in CDCl<sub>3</sub> with TMS as internal standard. Column chromatography (CC) was performed on Si gel (200–300 mesh, Qingdao Haiyang Chemical Co., Qingdao, China) and Sephadex LH-20 (Sigma). TLC was carried out with precoated Si gel plates (GF-254, Qingdao Haiyang Chemical Co., Qingdao, China).

#### 3.2. Algal Material

The marine red alga *Laurencia okamurai* Yamada was collected along Weihai coastline in Shandong Province, China, in May, 2007, and was identified by B.-M. Xia, Institute of Oceanology,

Chinese Academy of Sciences (IOCAS). A voucher specimen (HZ0705) has been deposited at the Key Laboratory of Experimental Marine Biology of IOCAS.

### 3.3. Extraction and Isolation

The dried and powdered alga *L. okamurai* (3.8 kg) was extracted with a mixture of CHCl<sub>3</sub> and MeOH (1:1, v/v). The concentrated extracts were partitioned between H<sub>2</sub>O and EtOAc. The EtOAc-soluble fraction was loaded to Si gel column, eluting with a step gradient of increasing EtOAc (0%–100%) in petroleum ether (PE) to give eight fractions I–VIII. Fraction II eluted with PE/EtOAc 100:1 and was further purified by preparative TLC to afford a mixture of **3** and **4** (5.6 mg). Fraction IV eluted with PE/acetone 100:1 and was further separated by preparative TLC to afford **1** (3.7 mg), **2** (4.7 mg), **6** (13.1 mg). Fraction VI eluted with PE/acetone 30:1 and was further separated by Sephadex LH-20 (MeOH) CC and preparative TLC to afford **5** (10.7 mg).

### 3.4. Computational Details

*Okamurene A (1)*: Colorless oil;  $[\alpha]_D^{18} +2.3$  (*c* 0.11, MeOH); UV (MeOH)  $\lambda_{\max}$  (log  $\epsilon$ ) 221 (3.56) nm; IR (KBr)  $\nu_{\max}$  3065, 2964, 2857, 1514, 1479, and 1205 cm<sup>-1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Table 1; HRESIMS *m/z* 297.0748 [M + H]<sup>+</sup> (calcd for C<sub>15</sub>H<sub>22</sub><sup>79</sup>BrO, 297.0854).

*Okamurene B (2)*: Colorless oil;  $[\alpha]_D^{18} +3.6$  (*c* 0.06, MeOH); UV (MeOH)  $\lambda_{\max}$  (log  $\epsilon$ ) 221 (3.66) nm; IR (KBr)  $\nu_{\max}$  3068, 2964, 2857, 1514, 1477, and 1208 cm<sup>-1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Table 1; HRESIMS *m/z* 319.0726 [M + Na]<sup>+</sup> (calcd for C<sub>15</sub>H<sub>21</sub>BrONa, 319.0673).

*Okamurenes C (3)* and *D (4)*: Colorless oil; IR (KBr)  $\nu_{\max}$  3096, 2924, 2854, 1637, 1457, and 1024 cm<sup>-1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Table 2; HRESIMS *m/z* 219.1757 [M + H]<sup>+</sup> (calcd for C<sub>15</sub>H<sub>23</sub>O, 219.1749).

*Okamurene E (5)*: Colorless oil;  $[\alpha]_D^{18} +7.6$  (*c* 0.09, MeOH); IR (KBr)  $\nu_{\max}$  3401, 2971, 2928, 1549, 1447, 1367 and 1121 cm<sup>-1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Table 2; HRESIMS *m/z* 281.0846 [M – H<sub>2</sub>O + H]<sup>+</sup> (calcd for C<sub>15</sub>H<sub>22</sub><sup>79</sup>Br, 281.0905), and 283.0860 [M – H<sub>2</sub>O + H]<sup>+</sup> (calcd for C<sub>15</sub>H<sub>22</sub><sup>81</sup>Br, 283.0884).

*Okamuragenin (6)*: Colorless oil;  $[\alpha]_D^{18} +11.2$  (*c* 0.18, MeOH); IR (KBr)  $\nu_{\max}$  3060, 2926, 2854, 2762, 1728, 1421, and 1134 cm<sup>-1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Table 2; HRESIMS *m/z* 385.9926 [M + NH<sub>4</sub>]<sup>+</sup> (calcd for C<sub>12</sub>H<sub>22</sub>N<sup>79</sup>Br<sub>2</sub>O<sub>3</sub>, 385.9966), 387.9986 [M + NH<sub>4</sub>]<sup>+</sup> (calcd for C<sub>12</sub>H<sub>22</sub>N<sup>79</sup>Br<sup>81</sup>BrO<sub>3</sub>, 387.9946).

### 3.5. Brine Shrimp Toxicity

Brine shrimp (*Artemia salina*) toxicity of crude extract and pure compounds was determined as detailed previously [26,27].

## 4. Conclusions

Four new bisabolane sesquiterpenes, okamurenes A–D (**1–4**), a new chamigrane derivative, okamurene E (**5**), and a new C<sub>12</sub>-acetogenin, okamuragenin (**6**), together with 13 known related metabolites, were isolated from the marine red alga *L. okamurai*. Among them, okamurenes A and B

(1 and 2) are first examples of bromobisabolane sesquiterpenes possessing a phenyl moiety among *Laurencia*-derived sesquiterpenes, while okamuragenin (6) was the first acetogenin aldehyde possessing a C<sub>12</sub>-carbon skeleton. Each of the isolated compounds was evaluated for the brine shrimp (*Artemia salina*) lethal assay and 7-hydroxy laurene displayed potent lethality with LD<sub>50</sub> 1.8 μM.

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