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# Oxygenated Cembrene Diterpenes from *Sarcophyton convolutum:* Cytotoxic Sarcoconvolutum A–E

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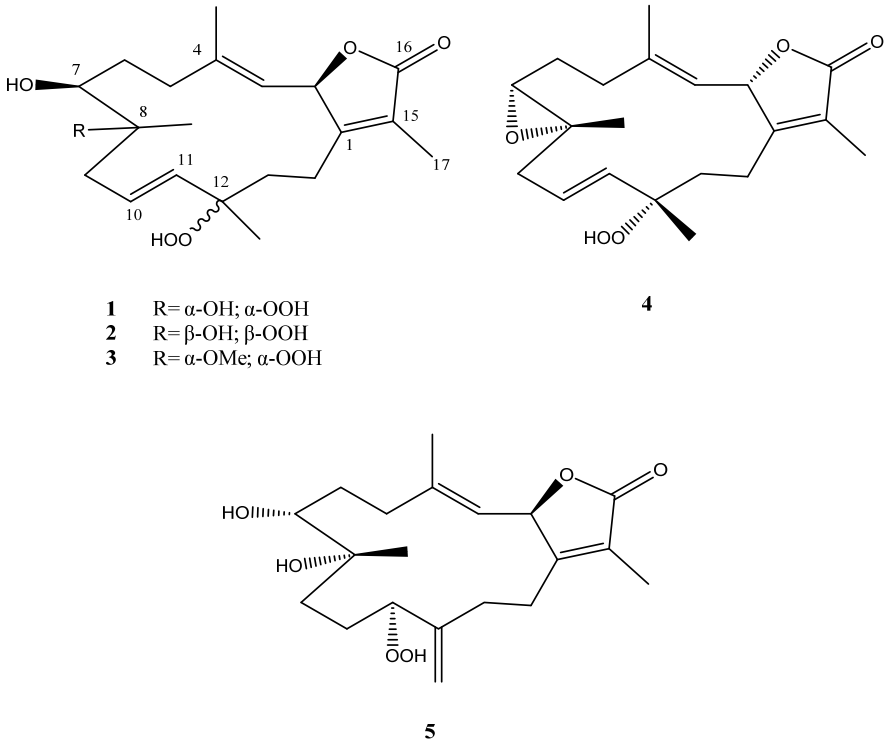
**Abstract:** The soft coral genus *Sarcophyton* contains the enzymatic machinery to synthesize a multitude of cembrene-type diterpenes. Herein, highly oxygenated cembrenoids, sarcoconvolutum A–E (**1**–**5**) were purified and characterized from an ethyl acetate extract of the red sea soft coral, *Sarcophyton convolutum*. Compounds were assemblies according to spectroscopic methods including FTIR, 1D- and 2D-NMR as well as HRMS. Metabolite cytotoxicity was tested against lung adenocarcinoma, cervical cancer, and oral-cavity carcinoma (A549, HeLa and HSC-2, respectively). The most cytotoxic compound, (**4**) was observed to be active against cell lines A549 and HSC-2 with $IC_{50}$ values of 49.70 and 53.17 μM, respectively.

**Keywords:** *Sarcophyton convolutum*; sarcoconvolutum A–E; cembrenoids; cytotoxicity

## 1. Introduction

Natural products and structural analogues are key for drug discovery, especially for pharmacotherapies for cancers and infectious diseases [1–5]. Such biologically active metabolites are characterized by scaffold diversity and structural complexity. For example, within *Sarcophyton* soft coral, isolated metabolites include cembrenoids [2,6–9], diterpene dimers [10,11], sesquiterpenes [12,13], ceramides [14], steroids [15,16], and prostaglandins [17,18]. Cembrenoids in particular exhibit a significant number of $sp^3$ carbon centers and oxygen atoms with no nitrogen or halogen atoms. Higher numbers of H-bond acceptors and donors confer hydrophilicity and ring structures provide significant molecular rigidity. Cembrenoids also exhibit promising biological activities including anticancer [6–8], anti-inflammatory [8], antifouling [19,20], ichthyotoxic [21], antifeedant [12], antiviral [22], and neuroprotective [23] properties.

In a pursuit of novel metabolites with biological activity, polyoxygenated cembrene-type diterpenoids (Figure 1) have been isolated from the Red Sea soft coral *S. convolutum*. Metabolite antiproliferative activity was assayed against a non-small-cell lung adenocarcinoma, a cervical cancer, and an oral-cavity squamous-cell carcinoma, A549, HeLa, and HSC-2, respectively.




**1** R= α-OH; α-OOH
**2** R= β-OH; β-OOH
**3** R= α-OMe; α-OOH

**Figure 1.** Sarcoconvolutum A–E (**1**–**5**) isolated from *S. convolutum*.

## 2. Results and Discussion

Sarcoconvolutum A (**1**) was isolated as yellow oil with a positive optical rotation (+10.6) in a MeOH solvent. The HRCIMS molecular ion peak at *m*/*z* 367.2122 $[M + H]^+$ was assigned predicting a molecular formula of $C_{20}H_{30}O_6$ (calcd. 367.2121, $[M + H]^+$) with six degrees of unsaturation suggesting a bicyclic skeleton. FTIR spectrum exhibited bands at 3465 $cm^{-1}$ and 1765 $cm^{-1}$ corresponding to hydroxyls and carbonyl groups. From $^1H$ NMR spectrum (Table 1), two oxygenated protons at $\delta_H$ 3.26 br d ($J$ = 10.8 Hz) and 5.53 d ($J$ = 10.4 Hz), three olefinic protons at $\delta_H$ 4.84 br d ($J$ = 10.4 Hz), 5.48 br d ($J$ = 16.3 Hz), and 5.50 m, in addition to four methyls were at $\delta_H$ 1.28 s, 1.37 s, 1.83 s, and 1.87 s were characterized. Twenty carbon resonances were predicted from $^{13}C$ NMR (Table 1) and categorized into 6 quaternary (including carbonyl at $\delta_C$ 175.3; three olefinics at $\delta_C$ 123.1, 143.9 and 162.3; alongside of two oxygenated carbons at $\delta_C$ 74.5 and 84.1), five methines (comprising two oxygenated carbons at $\delta_C$ 71.6 and 79.4; as well as three olefinic at $\delta_C$ 121.1, 127.8, and 134.8), five methylenes, four methyls ($\delta_C$ 8.8, 16.5, 20.3, and 24.2) depending upon DEPT and HSQC experiments. The above analyses of 1D NMR of **1** revealed the cembrene-based diterpenoid [6,7,24]. The 1D NMR of 1 was similar to 12-hydroperoxylsarcoph-10-ene isolated previously from *S. glaucum* [24]. Observed signal differences for the newly isolated compound included a down field shift of H-7 and C-7 by 0.73 and 12.1 ppm, respectively; a down field shift of 15.3 ppm for C-8; and a down field shift of 6 ppm for Me-19 consistent with the substitution of a hydroxyl group at C-7 instead of an epoxide ring coupling C-7 and C-8. The proposed structure was confirmed by $^1H$ $^1H$ COSY and HMBC spectral analysis. H-2 [$\delta_H$ 5.53 d ($J$ = 10.4 Hz)]/C-4 ($\delta_C$ 143.9, $J^3$), and $^1H$ $^1H$ COSY of H-2/H-3 [$\delta_H$ 4.84 br d ($J$ = 10.4 Hz)] indicated the location of $\Delta^{3(4)}$. The HMBC correlation of $H_3$-18 ($\delta_H$ 1.87 s) and $C_{H2}$-5 ($\delta_C$ 35.2, $J^3$) followed by $^1H$ $^1H$ COSY correlations of $H_2$-5 [$\delta_H$ 2.18 br d ($J$ = 12.7 Hz)]/ H-6 ($\delta_H$ 1.77 br d ($J$ = 13.4 Hz)], H-6/H-7 ($\delta_H$ 3.26 br d ($J$ = 10.8 Hz) confirmed a hydroxyl group at C-7. HMBC correlation of $H_3$-19 ($\delta_H$ 1.28 s)/C-7 ($\delta_C$ 71.6, $J^3$), $H_3$-19/C-8 ($\delta_C$ 74.5, $J^3$), and H-7/C-8 ($J^2$) established hydroxylation of the C-8 quaternary carbon. Subsequently, HMBC correlation of $H_3$-19/$C_{H2}$-9 ($\delta_C$ 42.9, $J^3$), $H_3$-20 ($\delta_H$ 1.37 s)/$C_{H2}$-11 ($\delta_C$ 134.8, $J^3$), in addition to $^1H$ $^1H$ COSY of $H_2$-9 [$\delta_H$ 2.34 m]/H-10 ($\delta_H$ 5.50 m), H-10/H-11 [($\delta_H$ 5.48 br d ($J$ = 16.3 Hz)] established $\Delta^{10(11)}$. The down field

shift of C-12 by ca. 7 ppm [24] as well as the HMBC correlation of H-11/C-12 ($\delta_C$ 84.1, $J^3$), $H_3$-20/C-12 ($J^2$), and $H_3$-20/C-13 ($\delta_C$ 34.4 $J^3$) indicated the presence of a hydroperoxide group at C-12. The C-1/C-2 included lactone ring was established via HMBC of H-2/C-16 (keto group, $\delta_C$ 175.3, $J^3$), H-2/C-15 ($\delta_C$ 123.1, $J^3$), $H_3$-17 ($\delta_H$ 1.83 s)/C-16 ($J^3$), $H_3$-17/C-1 ($\delta_C$ 162.3, $J^3$), and $H_3$-17/C-15 (Figure 2).

The relative configuration of 1 is based on coupling constants and NOESY data (Figure 3). From the coupling constant of H-2 (10.4 Hz) along with the vicinal coupling of H-3 (10.4 Hz), the *cis* orientation and α configuration of H-2 was established [7,24,25]. Starting from this point, the NOESY correlations of H-2α [$\delta_H$ 5.53 d ($J$ = 10.4 Hz)]/ $H_3$-18 [$\delta_H$ 1.87 s], $H_3$-18/ H-5α [$\delta_H$ 2.18 br d ($J$ = 12.7 Hz)], $H_3$-18/ H-6α [$\delta_H$ 1.77 br d ($J$ = 13.4)], H-5α/ H-6α, and H-6α/ H-7 [$\delta_H$ 3.26 br d ($J$ = 10.8 Hz)] established an α orientation for H-7. NOESY correlations of H-6β [$\delta_H$ 1.41 ddd ($J$ = 13.7, 12.7, 10.9 Hz)]/ $H_3$-19 ($\delta_H$ 1.28 s), H-9β ($\delta_H$ 2.34 m)/ $H_3$-19, H-9β/ H-11 [$\delta_H$ 5.48 br d ($J$ = 16.3 Hz)], H-11/ H-13β [$\delta_H$ 1.72 br t ($J$ = 13.2 Hz)], H-13β/ $H_3$-20 [$\delta_H$ 1.37 s], H-11/ $H_3$-20 indicated β configurations for both Me-19 and Me-20. Based on the described spectral analysis, **1** was identified as 7β,8α-dihydroxy-12α-hydroperoxide-16-keto-cembra-1*E*,3*E*,10*E*-triene (sarcoconvolutum A).

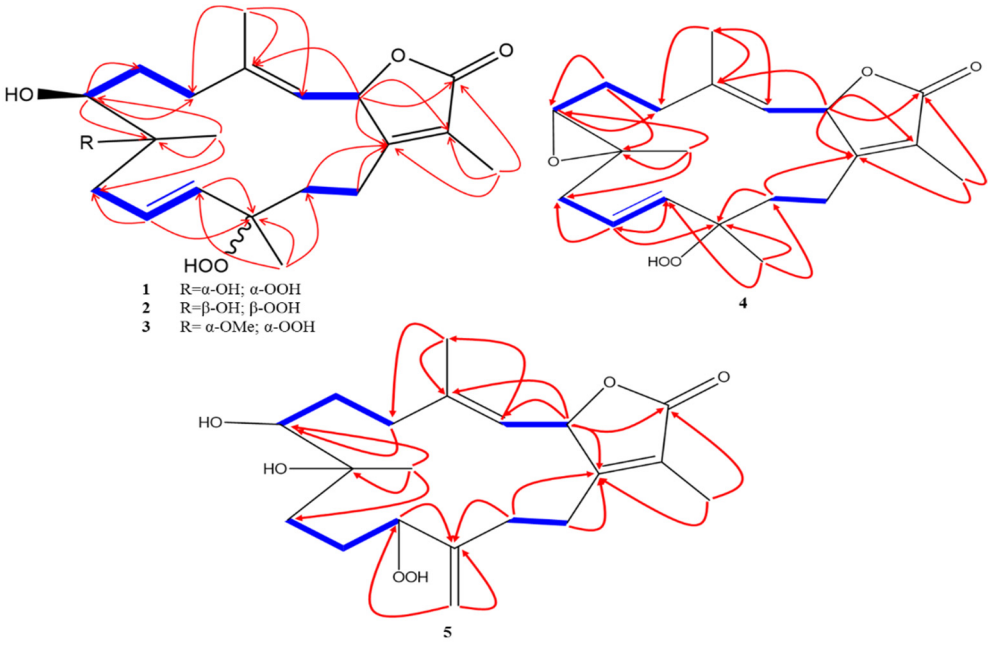



**Figure 2.** Key HMBC and $^1$H $^1$H COSY of sarcoconvolutum A–E (**1**–**5**).

Sarcoconvolutum B (**2**) was isolated as yellow oil with a positive optical rotation (+43.1) in MeOH. From the HRCIMS molecular ion peak at *m/z* 367.2122 $[M + H]^+$, the molecular formula of $C_{20}H_{30}O_6$ (calcd. 367.2121, $[M + H]^+$) a bicylic skeleton with six degrees of unsaturation was predicted. The FTIR broad bands corresponding to hydroxyls and carbonyl groups were detected at 3462 $cm^{-1}$ and 1768 $cm^{-1}$. The $^1$H NMR spectrum (Table 1) displayed two oxygenated protons at $\delta_H$ 3.32 d ($J$ = 10.9 Hz) and 5.45 d ($J$ = 10.2 Hz), three olefinic protons at $\delta_H$ 4.88 d ($J$ = 10.1 Hz), 5.50 d ($J$ = 16.1 Hz), and 5.57 dt ($J$ = 16.1, 7.2 Hz), along with four methyls were at $\delta_H$ 1.28 s, 1.42 s, 1.82 s, 1.85 s. From the observed twenty signals in the $^{13}$C NMR spectrum (Table 1), 6 quartenary (including carbonyl at $\delta_C$ 174.8; three olefinics at $\delta_C$ 123.5, 143.5 and 161.9; alongside of two oxygenated carbons at $\delta_C$ 74.5 and 84.0), 5 methenes (comprising two oxygenated carbons at $\delta_C$ 78.7 and 71.1, as well as three olefinic signals at $\delta_C$ 121.6, 126.9, 134.6), 5 methylenes, 4 methyls ($\delta_C$ 8.9, 16.1, 23.6, and 24.1) were identified based on DEPT and HSQC signals. A down field shift of $\delta_C$ 84.0 by ca. 7 ppm suggested the presence of a hydroperoxyl unit at C-12 [24] which was consistent with mass spectral data. A $^{13}$C NMR comparison of the two isolated compounds indicated a down field shift of Me-20 for **2**. The 2D NMR comparisons showed otherwise

similar $^{1}$H $^{1}$H COSY and HMBC signals (Figure 2). A $^{13}$C NMR down field shift of 3.3 ppm for Me-20 indicated the opposite stereochemistry at C-12.

Relative stereochemistry was determined based on coupling constants and NOESY data (Figure 3). The coupling constant of H-2 (10.2 Hz) in addition to the vicinal coupling of H-3 (10.1 Hz) suggested a *cis* orientation and *α* configuration of H-2 [7,24,25]. The NOESY correlations of H-2*α* [$\delta_H$ 5.45 d ($J$ = 10.2 Hz)]/ $H_3$-18 [$\delta_H$ 1.82 s], H-18/ H-5 [$\delta_H$ 2.37 td ($J$ = 13.2t, 2.8 Hz)], H-18/ H-6 [$\delta_H$ 1.49 td ($J$ = 13.7t, 3.6 Hz)], H-5*α*/ H-6, H-6/ H-7 [$\delta_H$ 3.32 br d ($J$ = 10.9 Hz)], H-7/ H-19 [$\delta_H$ 1.28 s] and H-13β [$\delta_H$ 1.95 td ($J$ = 13.1, 4.1 Hz)]/ $H_3$-19 indicated a H-7*α* Me-19 and Me-20orientation. Based upon these spectral data, **2** was identified as 7β,8α-dihydroxy-12β-hydroperoxide-16-keto-cembra-1*E*,3*E*,10*E*-triene (sarcoconvolutum B).

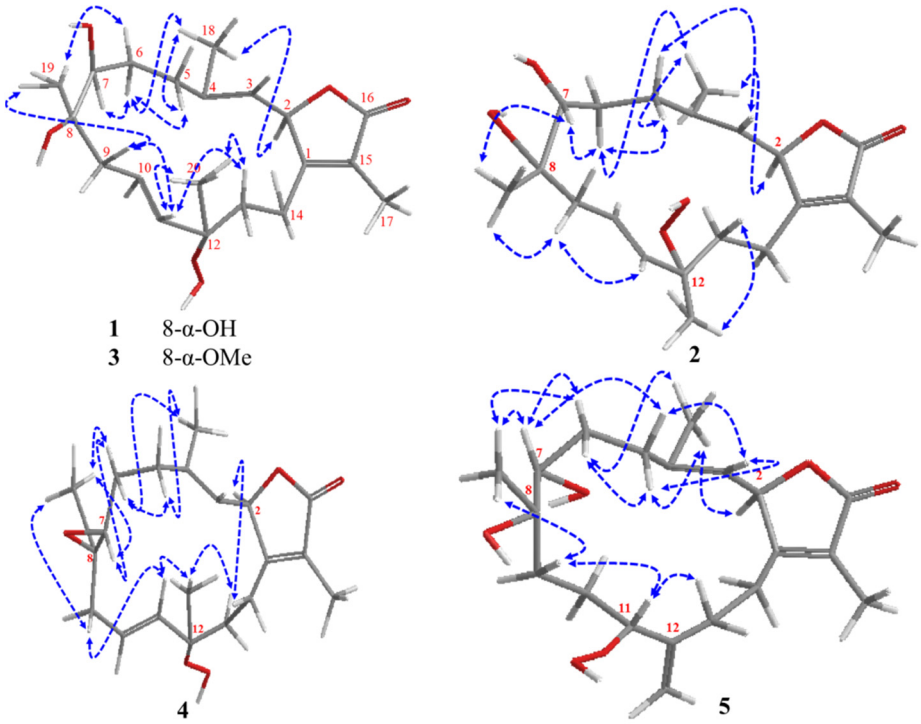



**Figure 3.** Significant NOESY of sarcoconvolutum A–E (**1**–**5**).

Sarcoconvolutum C (**3**) was isolated as yellow oil with a positive optical rotation (+33.1, *c* 0.003, $CH_3OH$). The molecular formula was deduced as $C_{21}H_{32}O_6$ (calcd. 364.2250, [M-OH + H]$^+$) based on a HREIMS molecular ion peak at $m/z$ 364.2258 [M-OH + H]$^+$ indicating a bicylic skeleton with six degrees of unsaturation. FTIR broad bands corresponded to hydroxyls and carbonyl groups were detected at 3457 cm$^{-1}$ and 1761 cm$^{-1}$. From $^{1}$H NMR spectrum (Table 1), two oxygenated protons at $\delta_H$ 3.27 br d ($J$ = 10.9 Hz) and $\delta_H$ 5.49 d ($J$= 9.9 Hz), three olefinic protons at $\delta_H$ 4.86 br d ($J$ = 9.9 Hz), $\delta_H$ 5.44 br d ($J$ = 16.1 Hz) and $\delta_H$ 5.56 dt ($J$ = 15.6, 8.1t Hz), four methyls as well as one methyl of methoxy group at $\delta_H$ 3.23 s were assigned. Based on the $^{13}$C NMR (Table 1), twenty-one carbon resonances were observed and were categorized by DEPT and HSQC analysis as 6 quaternary carbons (including carbonyl at $\delta_C$ 174.8; three olefinics at $\delta_C$ 123.3, 144.2 and 161.4; alongside of two oxygenated carbons at $\delta_C$ 78.3 and 84.4), 5 methines (comprising two oxygenated carbons at $\delta_C$ 78.3 and 71.9; as well as three olefinic at $\delta_C$ 121.2, 128.2, 134.0), 5 methylenes, 5 methyls ($\delta_C$ 9.0, 16.2, 18.2, 21.2, and 49.3). As with **1** and **2**, down field shift at $\delta_C$ 84.4 ppm of ca. 7 ppm along with a mass shift of [M-$H_2O$], suggested the addition of a hydroperoxy group [24]. The compound is similar to **1** and **2** except for a down field shift of $\delta_H$ 78.3 by 3.8 ppm associated with C-8 and a new methoxyl signal at $\delta_H$ 3.23 s and $\delta_C$ 49.3. HMBC correlations of a methyl proton at $\delta_H$ 3.23 and the C-12 at $\delta_H$ 78.3

confirm the localization of the methoxylation to C-12 (Figure 2). Thus, **3** was identified as 7-hydroxy-8-methoxy-12-hydroperoxide-16-keto-cembra-1*E*,3*E*,10*E*-triene.

**Table 1.** $^{1}$H and $^{13}$C NMR ($CDCl_3$, 500 Hz) of sarcoconvolutum A–E (**1**–**5**).

| No | Sarcoconvolutum A (1) | | Sarcoconvolutum B (2) | | Sarcoconvolutum C (3) | | Sarcoconvolutum D (4) | | Sarcoconvolutum E (5) | |
|---|---|---|---|---|---|---|---|---|---|---|
| | $^{1}$H NMR (*J* Hz) | $^{13}$C | $^{1}$H NMR (*J* Hz) | $^{13}$C | $^{1}$H NMR (*J* Hz) | $^{13}$C | $^{1}$H NMR (*J* Hz) | $^{13}$C | $^{1}$H NMR (*J* Hz) | $^{13}$C |
| 1 | — | 162.3 | — | 161.9 | — | 161.4 | — | 161.7 | — | 161.8 |
| 2 | 5.53 d (10.4) | 79.4 | 5.45 d (10.2) | 78.7 | 5.49 d (9.9) | 79.0 | 5.45 d (9.4) * | 78.8 | 5.47 d (10.1) | 80.2 |
| 3 | 4.84 br d (10.4) | 121.1 | 4.88 br d (10.1) | 121.6 | 4.86 br d (9.9) | 121.2 | 5.00 d (9.4) | 121.1 | 4.92 br d (10.1) | 119.2 |
| 4 | — | 143.9 | — | 143.5 | — | 144.2 | — | 144.4 | — | 145.9 |
| 5α<br>5β | 2.18 br d (12.7)<br>2.40 t (11.3) | 35.2 | 2.23 dt (13.2, 3.4t)<br>2.37 td (13.2t, 2.8) | 35.3 | 2.18 dt (13.4, 3.4t)<br>2.37 td (13.4, 7.4) | 35.4 | 2.26 m *<br>2.34 td (13.1t, 5.7) | 36.6 | 2.35 dt (13.0, 7.6t)<br>2.10 td (13.0t, 5.1) | 37.9 |
| 6α<br>6β | 1.77 br d (13.4)<br>1.51 ddd (13.7, 12.7, 10.9) | 26.7 | 1.49 td (13.7t, 3.6)<br>1.83 m * | 26.3 | 1.87 td (13.4t, 3.6)<br>1.44 td (15.3t, 4.3) | 26.7 | 1.60 m<br>1.82 m * | 24.1 | 1.75 m *<br>1.25 m * | 30.5 |
| 7 | 3.26 br d (10.8) | 71.6 | 3.32 br d (10.9) | 71.1 | 3.27 br d (10.9) | 71.9 | 2.65 dd (6.8, 3.6) | 57.0 | 3.45 br d (10.5) | 75.0 |
| 8 | — | 74.5 | — | 74.5 | — | 78.3 | — | 60.0 | — | 73.9 |
| 9α<br>9β | 2.34 m *<br>2.34 m * | 42.9 | 2.44 dd (13.7, 7.8)<br>2.28 dd (13.4, 6.6) | 43.4 | 2.34 m *<br>2.34 m * | 37.2 | 2.41 dd (13.1, 5.7)<br>2.46 dd (13.3, 7.1) | 39.0 | 1.57 m<br>1.51 m | 35.1 |
| 10α<br>10β | 5.50 m | 127.8 | 5.57 dt (16.1, 7.2) | 126.9 | 5.56 dt (15.6, 8.1t) | 128.2 | 5.42 m * | 125.8 | 1.57 m<br>1.32 m | 25.2 |
| 11 | 5.48 br d (16.3) | 134.8 | 5.50 br d (16.1) | 134.6 | 5.44 br d (16.1) | 134.0 | 5.47 m * | 135.5 | 4.43 dd (9.0, 5.0) | 89.3 |
| 12 | — | 84.1 | — | 84.0 | — | 84.4 | — | 84.6 | — | 146.0 |
| 13α<br>13β | 1.72 br t (13.2)<br>1.68 td (13.2t, 5.6) | 34.4 | 1.49 td (13.7, 3.6)<br>1.95 td (13.1, 4.1) | 36.4 | 1.80 td (13.4t, 3.5)<br>1.67 td (13.4t, 4.7) | 35.1 | 1.58 m *<br>1.89 m * | 36.0 | 2.02 br t (15.1)<br>2.25 m | 27.8 |
| 14α<br>14β | 2.01 br t (13.2)<br>2.43 br t (13.2) | 22.7 | 2.06 br t (13.7)<br>2.62 td (13.7, 4.0) | 21.6 | 2.05 br t (13.4)<br>2.40 td (13.7, 4.3) | 22.7 | 2.07 br t<br>2.27 td * (13.2t, 5.5) | 22.0 | 2.13 m<br>2.65 td (12.9, 4.9) | 24.5 |
| 15 | — | 123.1 | — | 123.5 | — | 123.3 | — | 123.6 | — | 124.3 |
| 16 | — | 175.3 | — | 174.8 | — | 174.8 | — | 174.8 | — | 174.9 |
| 17 | 1.83 s | 8.8 | 1.85 s | 8.9 | 1.85 s | 9.0 | 1.86 s | 9.1 | 1.86 s | 8.9 |
| 18 | 1.87 s | 16.5 | 1.82 s | 16.1 | 1.83 s | 16.2 | 1.87 s | 15.8 | 1.86 s | 16.4 |
| 19 | 1.28 s | 24.2 | 1.28 s | 24.1 | 1.19 s | 18.2 | 1.31 s | 22.2 | 1.26 s | 24.8 |
| 20 | 1.37 s | 20.3 | 1.42 s | 23.6 | 1.41 s | 21.2 | 1.41 s | 18.8 | 5.07 s, 5.13 s | 113.4 |
| OMe | | | | | 3.23 s | 49.3 | | | | |

* overlapped.

The relative configuration of **3** was deduced via the coupling constants and NOESY analysis (Figure 3). The coupling constant of H-2 (9.9 Hz) and the vicinal coupling of H-3 (9.9 Hz) identified an *α* and *cis* orientation for H-2 [7,24,25]. The NOESY experiments exhibited the same configuration of **1** that elucidated the *α* orientation of H-7 and β and both methyls, Me-19 and Me-20. Based on these spectral observations, **3** was identified as 7β-hydroxy-8α-methoxy-12α-hydroperoxide-16-keto-cembra-1*E*,3*E*,10*E*-triene (sarcoconvolutum C).

Sarcoconvolutum D (**4**) was obtained as dark yellow oil with a positive optical rotation (+37.2, *c* 0.003, $CH_3OH$). The HREIMS molecular ion peak at *m*/*z* 348.1939 $[M]^+$ that predicted a molecular formula of $C_{20}H_{28}O_5$ (calcd. 348.1937, $[M]^+$) with a bicylic skeleton and seven degrees of unsaturation. The corresponding FTIR broad bands to hydroxyls and carbonyl groups at 3456 $cm^{-1}$ and 1763 $cm^{-1}$ were assigned. The $^{1}$H NMR (Table 1) revealed two oxygenated protons at $\delta_H$ 2.65 dd (*J* = 6.8, 3.6 Hz), and 5.45 d (*J* = 9.4 Hz), three olefineic protons at $\delta_H$ 5.00 d (*J* = 9.4 Hz), 5.42 m, and 5.47 m, in addition to four methyls at $\delta_H$ 1.31 s, 1.41 s, 1.86 s, and 1.87 s. From $^{13}$C NMR, twenty carbon resonances were characterized and categorized to six quartenary (consisting of carbonyl at $\delta_C$ 174.8; three olefinics at $\delta_C$ 123.6, 144.4, and 161.7; and two oxygenated carbons at $\delta_C$ 60.0 and 84.6, five methenes (comprising two oxygenated carbons at $\delta_C$ 57.0, and 78.8; and two olefinic carbon (at $\delta_C$ 125.8 and 135.5), five methylenes, four methyls based on DEPT-135 and HSQC analysis. The compound is very similar to **1** except for an up-field shift of $\delta_H$ 2.65 dd (*J* = 6.8, 3.6 Hz) by 0.67 ppm and an up-field shift of $\delta_C$ 57.0 by 14.6 ppm both associated with C-7 and an up-field shift of $\delta_C$ 60 by 14.5 ppm associated with C-8. These chemical shift changes were deduced to be a result of a C-7/C-8 epoxide ring that was consistent with a molecular ion peak at *m*/*z* 348.1939 ($C_{20}H_{28}O_5$). The localization of this group was derived via the HMBC correlations of Me-19 ($\delta_H$ 1.31 s)/C-7 ($J^3$), Me-19/C-8 ($J^2$), H-6 ($\delta_H$ 1.60 m)/C-7 ($J^2$), and H-6/C-8 ($J^3$) (Figure 2).

The relative stereochemistry was identified via coupling constants [7,24,25] and NOESY analysis (Figure 3). Compound **4** exhibited almost same NOESY correlations as **1** except the correlations of Me-19/H-7, and H-6β/H-7 that elucidated the β orientation of H-7. These data confirmed **4** as 12α-hydroperoxide-16-keto-cembra-1*E*,3*E*,10*E*-triene-7α, 8α-epoxide (sarcoconvolutum D).

Sarcoconvolutum E (**5**) was obtained as yellow oil exhibiting a positive optical rotation (+61.7, *c* 0.003, $CH_3OH$). The mass spectroscopy exhibited a HREIMS molecular ion peak at *m*/*z* 367.2130 $[M + H]^+$ that was identified as $C_{20}H_{30}O_6$ (calcd. 367.2042, $[M + H]^+$) with a bicylic skeleton and six degrees of unsaturation. The FTIR broad bands corresponding to hydroxyls and carbonyl groups were detected at 3464 $cm^{-1}$ and 1759 $cm^{-1}$. $^1H$ NMR (Table 1) displayed three oxygenated protons at $\delta_H$ 3.45 br d (*J* = 10.5 Hz), 4.43 dd (*J* = 9.0, 5.0 Hz), and 5.47 d (*J* = 10.1 Hz), one olefineic proton at $\delta_H$ 4.92 br d (*J* = 10.1 Hz), one exomethylene proton at $\delta_H$ 5.07 s, and 5.13 s, and three methyles at $\delta_H$ 1.26 s, 1.86 s (6H). The $^{13}C$ NMR showed twenty carbon resonances. Six quaternary (including carbonyl at $\delta_C$ 174.9; four olefinics at $\delta_C$ 124.3, 145.9, 146.0 and 161.8; and one oxygenated carbon at $\delta_C$ 73.9), four methenes (comprising two oxygenated carbons at $\delta_C$ 70.0 and 89.3; and one olefinic carbon (at $\delta_C$ 119.2), 7 methylenes (comprising exomethylene carbon at $\delta_C$ 113.4), three methyls ($\delta_C$ 8.9, 16.4, and 24.8) that were characterized via DEPT-135 and HSQC experiments. NMR spectral data of **5** were similar to **1** and **2** except for the following signals: (i) a down-field shift of $\delta_C$ 75.0, by ca. 3.5 ppm associated with C-7; (ii) an downfield shift of $\delta_H$ 3.45 brd (*J* = 10.5 Hz) by 0.2 ppm associated with H-7; and (iii) an up-field shift of $\delta_H$ 4.43 dd (*J* = 9.0, 5.0 Hz) by 1 ppm and a shift of ca. 45 ppm of $\delta_C$ 134.0 to 89.0 associated with C-11 indicating the oxygenation of this carbon; (iv) the appearance of $\delta_H$ 1.32 (m, 1.57) associated with H-10 and C-10 (at $\delta_C$ 25.0) indicating a disappearance of $\Delta^{10(11)}$; and (v) the presence of $\delta_C$ 146.0 associated with C-12 and signals $\delta_H$ 5.07 s, 5.13 s and $\delta_C$ 113.4 associated with an exomethylene carbon at C-20 indicating the presence of $\Delta^{12(20)}$. These new functional group assignments based on spectral data were consistent with 2D HMBC spectral signals including $H_3$-19 [$\delta_H$ 1.26 s]/C-7 [$\delta_C$ 75.0, $J^3$], $H_3$-19/C-7 [$\delta_C$ 73.9, $J^2$] confirmed the hydroxylation of both C-7 and C-8. The HMBC correlations of $H_2$-20 [at $\delta_H$ 5.07 s, 5.13 s]/C-12 [at $\delta_C$ 146.0, $J^2$], $H_2$-20/C-11 [at $\delta_C$ 89.0, $J^3$], and $H_2$-20/C-13 [at $\delta_C$ 27.8, $J^3$], H-11 [at $\delta_H$ 4.43 dd (*J* = 9.0, 5.0 Hz)]/C-12 [$J^2$], H-11/C-13 [$J^3$] confirmed the $\Delta^{12(20)}$ and the oxygenation of C-11 (Figure 2). Consistent with the mass spectral data and the observed up-field shift of C-11, the oxygenation of C-11 was identified as a hydroperoxide group.

The relative stereochemistry assignment was based the coupling constant of H-2 (10.1 Hz) and the vicinal coupling of H-3 (10.1 Hz) the indicated a *α* and *cis* orientation of H-2 [7,24,25]. NOESY correlations (Figure 3) of H-2α [$\delta_H$ 5.47 d (*J* = 10.1 Hz)]/ $H_3$-18 [$\delta_H$ 1.86 s], H-3α [$\delta_H$ 4.92 br d (*J* = 10.1 Hz)]/ H-5α [$\delta_H$ 2.35 dt (*J* = 13.0, 7.6t Hz)], and H-5α/ H-6 [$\delta_H$ 1.75 m], $H_3$-18/ H-6, H-5/ H-7 [$\delta_H$ 3.45 br d (*J*= 10.5 Hz)], H-6/ H-7, H-7/ $H_3$-19 [$\delta_H$ 1.26 s], $H_3$-19β/ H-9β [$\delta_H$ 1.51 m], H-9β/ H-11 [$\delta_H$ 4.43 dd (*J* = 9.0, 5.0 Hz)], H-13β [$\delta_H$ 2.25 m]/ H-identified a β orientation for H-7, H-11 and Me-19. Therefore, **5** was assigned as 7α,8α-dihydroxy-11α-hydroperoxide-16-keto-cembra-1E,3E,12(20)-triene (sarcoconvolutum E). 1D, 2D-NMR, HRMS and FTIR for all isolated secondary metabolites (1–5) were deposited in the supplementary files (see Figures S1–S50).

The initial screening of **1**–**5** (Figure 4) for cytotoxic activity was performed using three cancer cell lines (A549, HeLa and HSC-2) with concentrations of 100, 10 and 1 μM (Figure 4A). Compound **4** showed cytotoxic activity against cell lines A549 (Figure 4B) and HSC-2 (Figure 4C). The dose-dependent toxicity with concentrations range (0.001–100 μg/mL) was examined against A549 and HSC-2 cell lines (Figure 4). Compound **4** showed $IC_{50}$ values of 49.70 and 53.17 μM against A549 and HSC-2 cells, respectively (Table 2, Figure 4A,B).

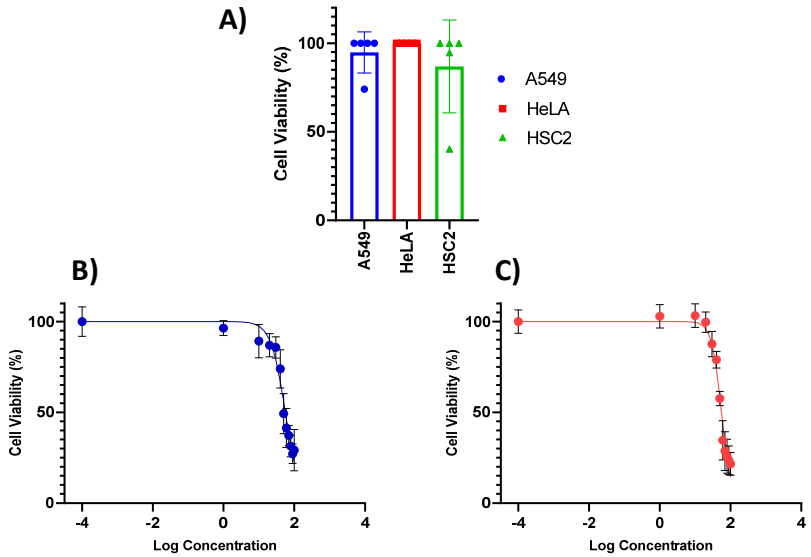



**Figure 4.** Cytotoxic activity: (**A**) Screening cytotoxic activity against A549, HeLa and HSC2 cell lines; (**B**) Dose-dependent toxicity with concentrations in a range from 0.001 to 100 μM for compounds **4** against A549; (**C**) Dose-dependent toxicity with concentrations in a range from 0.001 to 100 μM for compound 4 against HSC2.

**Table 2.** Cytotoxicity of 1-5 against cancer cell lines.

| Cell Lines | ($IC_{50}$, μM) | | | | | |
|---|---|---|---|---|---|---|
| | **1** | **2** | **3** | **4** | **5** | **Doxorubicin** |
| **A549** | >100 | >100 | >100 | 49.70 ± 0.05 | >100 | 0.42 ± 0.3 |
| **HeLa** | >100 | >100 | >100 | 91.98 ± 0.15 | >100 | 1.35 ± 0.16 |
| **HSC2** | >100 | >100 | >100 | 53.17 ± 0.03 | 91.39 ± 0.17 | 0.50 ± 2.6 |

## 3. Materials and Methods

### *3.1. General Experimental Procedures*

A JASCO P-2300 polarimeter (Tokyo, Japan) and shimazu FTIR-8400S instrument (Columbia, MD 21046, USA) was operated for optical rotation and IR spectra, respectively. Then, 1D and 2D NMR spectra were recorded on a Bruker 600 or 500 Hz NMR spectrometer (MA, USA). HR-MS spectra were obtained on a JEOL JMS-700 instrument (Tokyo, Japan). For Chromatography: column chromatography (CC) [silica gel 60 (Merck, 230–400 mesh, Merck, Darmstadt, Germany)]; TLC analysis: [precoated silica gel plates (Merck, Kieselgel 60 $F_{254}$, 0.25 mm, Merck, Darmstadt, Germany]. High-performance liquid chromatography (HPLC) equipped with a Jasco PU-980 pump, a Jasco UV-970 intelligent UV/VIS detector at 210 nm and a semi preparative reversed-phase column (Cosmosil $C_{18}$ column 250 × 10 mm, 5 μm).

### *3.2. Coral Material*

The animal soft coral *Sarcophyton convolutum* was collected from the Egyptian Red Sea (the coast of Hurghada) in March 2017 and was recognized by M Al-Hammady with a voucher specimen (08RS1071). It was deposited in the National Institute of Oceanography and Fisheries, marine biological station, Hurghada, Egypt.

### *3.3. Extraction and Separation*

The frozen soft coral (2.5 kg, total wet weight) was chopped into small pieces and extracted with ethyl acetate at room temperature (3 L × 5 times). The collective extracts were concentrated *in vacuo* to a brown gum. The dried material (95 g) was subjected to a silica gel column (6 × 120 cm) eluting with *n*-hexane (2000 mL), followed by a gradient of

*n*-hexane-EtOAc up to 100% EtOAc and EtOAc –MeOH up to 50% MeOH (3000 mL each of the solvent mixture). Fractions were collected (SC1-SC6) and monitored via TLC. Fraction (SC3; 1.4 g) was eluted step gradient with *n*-hexane-EtOAc over silica gel column afforded three main sub fractions (SC3A-C). Sub-fraction SC3A was re-purified by reversed-phase HPLC using MeOH/$H_2O$ (6.5:3.5), 3.5 mL/min, to afford **1** (6.1 mg, $t_R$ = 27 min), and **4** (11.6 mg, $t_R$ = 23 min). The sub-fraction SC3B was subjected to reversed-phase HPLC using MeOH/$H_2O$ (3:2), 3.5 mL/min, afforded **2** (10 mg, $t_R$ = 28 min). Fraction (SC4; 1.2 g) was further fractionated over silica gel column chromatography eluted by *n*-hexane/EtOAc step gradient afforded two main sub-fractions (SC4A and B). Sub-fraction SC4A was eluted with MeOH/$H_2O$ (1:1) over reversed-phase HPLC, 3 mL/min, to afford **3** (7.3 mg, $t_R$ = 31 min), and **5** (9.6 mg, $t_R$ = 32 min).

### *3.4. Spectral Data of Sarcoconvolutum A–E (1–5)*

7,8-Dihydroxy-12-hydroxy-16-keto-cembra-1E,3E,10E-triene (Sarcoconvolutum A, **1**): yellow oil; $[\alpha]_D^{25}$ +43.0 (c 0.03, $CH_3OH$); FT-IR (KBr) $\nu_{max}$: 3465, 2961, 1765, 1435, and 987 $cm^{-1}$; $^1H$ and $^{13}C$ NMR data ($CDCl_3$, 500 Hz), see Table 1; HRCIMS *m*/*z* 367.2122 (100, $[M+H]^+$); (calcd. 367.2121, for $C_{20}H_{31}O_6$).

7,8-Dihydroxy-12-hydroxy-16-keto-cembra-1E,3E,10E-triene (Sarcoconvolutum B, **2**): yellow oil; $[\alpha]_D^{25}$ +10.5 (c 0.03, $CH_3OH$); FT-IR (KBr) $\nu_{max}$: 3461, 2957, 1759, 1439, and 991 $cm^{-1}$; $^1H$ and $^{13}C$ NMR data ($CDCl_3$, 500 Hz), see Table 1; HRCIMS *m*/*z* 367.2122 (100, $[M+H]^+$); (calcd. 367.2122, for $C_{20}H_{31}O_6$).

7β-Hydroxy-8α-methoxy-12α-hydroperoxide-16-keto-cembra-1E,3E,10E-triene (sarcoconvolutum C, **3**): yellow oil; $[\alpha]_D^{25}$ +37.2 (c 0.03, $CH_3OH$); FT-IR (KBr) $\nu_{max}$: 3456, 2951, 1763, 1446, and 993 $cm^{-1}$; $^1H$ and $^{13}C$ NMR data ($CDCl_3$, 500 Hz), see Table 1; HRCIMS *m*/*z* 364.2258, $[M\text{-}OH+H]^+$]; (calcd. 364.2250, $[M\text{-}OH+H]^+$, for $C_{21}H_{32}O_6$).

12α-Hydroperoxide-16-keto-cembra-1E,3E,10E-triene-7α,8α-epoxide (sarcoconvolutum D, **4**): yellow oil; $[\alpha]_D^{25}$ +37.2 (c 0.03, $CH_3OH$); FT-IR (KBr) $\nu_{max}$: 3456, 2958, 1763, 1442, and 996 $cm^{-1}$; $^1H$ and $^{13}C$ NMR data ($CDCl_3$, 500 Hz), see Table 1; HREIMS *m*/*z* 348.1939 $[M]^+$; (calcd. 348.1937, for $C_{20}H_{28}O_5$).

7α,8α-Dihydroxy-11α-hydroperoxide-16-keto-cembra-1E,3E,12(20)-triene (sarcoconvolutum E, **5**): yellow oil; $[\alpha]_D^{25}$ +10.5 +61.7 (c 0.03, $CH_3OH$); FT-IR (KBr) $\nu_{max}$: 3464, 2955, 1759, 1433, and 997 $cm^{-1}$; $^1H$ and $^{13}C$ NMR data ($CDCl_3$, 500 Hz), see Table 1; HREIMS *m*/*z* 367.2130 (100, $[M+H]^+$); (calcd. 367.2042, for $C_{20}H_{31}O_6$).

### *3.5. Cell Culture and Treatment Conditions*

Human cancer cell lines of non-small cell lung adenocarcinoma (A549), squamous cell carcinoma of the oral cavity (HSC-2), and human cervical cancer cell (HeLa). All cell lines were purchased from American Type Culture Collection (ATCC®) and were maintained as monolayer culture in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% FBS, 4 mM l-glutamine, 100 U/mL penicillin, and 100 µg/mL streptomycin sulfate. Monolayers were passaged at 70–90% confluence using a trypsin-EDTA solution. All cell incubations were maintained in a humidified $CO_2$ incubator with 5% $CO_2$ at 37 °C. All materials and reagents for the cell cultures were purchased from Lonza (Verviers, Belgium).

### *3.6. Cytotoxicity Assay*

A modified MTT (3-[4,5,[4,5-dimethylthiazole-2-yl]-2,5-diphenyltetrazolium bromide) assay was used for cytotoxicity investigation were performed using based on a previously published method [26]. A549, HeLa and HSC2 cells (5000–10,000 cells/well) were seeded onto 96-well plates and incubated for 24 h with 5% $CO_2$ at 37 °C. The prepared tested compounds (**1**–**5**) were screened and the dose-dependent toxicity with concentrations range (0.001–100 µg/mL) were investigated according to our protocol [26]. Doxorubicin (2 mg/mL) was used as positive control.

### *3.7. Anti-Proliferation Quantitative Analysis*

GraphPad Prism® v6.0 software (GraphPad Software Inc., San Diego, CA, USA) were used to calculate the concentration-response curve fit to the non-linear regression for $IC_{50}$ values.

## 4. Conclusions

Five compounds, sarcoconvolutum A–E were isolated from an organic extract of *Sarcophyton convolutum* tissue. Chemical structures were elucidated based upon spectroscopic analyses. The cytotoxic activity of the isolated compounds was screened against three cancer cell lines (A549, HeLa and HSC2) and **4** was found to be the most active compound against cell lines A549 and HSC-2 with $IC_{50}$ values of 49.70 and 53.17 μM, respectively.

**Supplementary Materials:** The following are available online https://www.mdpi.com/article/10.3390/md19090519/s1, Figures S1–S50: spectra for compounds **1**–**5**.

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## References

1. Ahmed, A.A.; Hegazy, M.-E.F.; Hassan, N.M.; Wojcinska, M.; Karchesy, J.; Pare, P.W.; Mabry, T.J. Constituents of Chrysothamnus viscidiflorus. *Phytochemistry* **2006**, *67*, 1547–1553. [CrossRef]
2. Hegazy, M.E.F.; Mohamed, T.A.; Alhammady, M.A.; Shaheen, A.M.; Reda, E.H.; Elshamy, A.I.; Aziz, M.; Paré, P.W. Molecular architecture and biomedical leads of terpenes from red sea marine invertebrates. *Mar. Drugs* **2015**, *13*, 3154–3181. [CrossRef] [PubMed]
3. Khalifa, S.A.M.; Elias, N.; Farag, M.A.; Chen, L.; Saeed, A.; Hegazy, M.-E.F.; Moustafa, M.S.; El-Wahed, A.; Al-Mousawi, S.M.; Musharraf, S.G. Marine natural products: A source of novel anticancer drugs. *Mar. Drugs* **2019**, *17*, 491. [CrossRef] [PubMed]
4. Nweze, J.A.; Mbaoji, F.N.; Li, Y.-M.; Yang, L.-Y.; Huang, S.-S.; Chigor, V.N.; Eze, E.A.; Pan, L.-X.; Zhang, T.; Yang, D.-F. Potentials of marine natural products against malaria, leishmaniasis, and trypanosomiasis parasites: A review of recent articles. *Infect. Dis. Poverty* **2021**, *10*, 1–19. [CrossRef]
5. Hegazy, M.-E.F.; Mohamed, T.A.; Abdel-Latif, F.F.; Alsaid, M.S.; Shahat, A.A.; Pare, P.W. Trochelioid A and B, new cembranoid diterpenes from the Red Sea soft coral Sarcophyton trocheliophorum. *Phytochem. Lett.* **2013**, *6*, 383–386. [CrossRef]
6. Hegazy, M.-E.F.; Elshamy, A.I.; Mohamed, T.A.; Hamed, A.R.; Ibrahim, M.A.A.; Ohta, S.; Paré, P.W. Cembrene diterpenoids with ether linkages from Sarcophyton ehrenbergi: An anti-proliferation and molecular-docking assessment. *Mar. Drugs* **2017**, *15*, 192. [CrossRef]
7. Hegazy, M.-E.F.; Mohamed, T.A.; Elshamy, A.I.; Hamed, A.R.; Ibrahim, M.A.A.; Ohta, S.; Umeyama, A.; Paré, P.W.; Efferth, T. Sarcoehrenbergilides D–F: Cytotoxic cembrene diterpenoids from the soft coral Sarcophyton ehrenbergi. *RSC Adv.* **2019**, *9*, 27183–27189. [CrossRef]
8. Lin, W.-Y.; Su, J.-H.; Lu, Y.; Wen, Z.-H.; Dai, C.-F.; Kuo, Y.-H.; Sheu, J.-H. Cytotoxic and anti-inflammatory cembranoids from the Dongsha Atoll soft coral Sarcophyton crassocaule. *Bioorg. Med. Chem.* **2010**, *18*, 1936–1941. [CrossRef] [PubMed]
9. Liu, X.; Zhang, J.; Liu, Q.; Tang, G.; Wang, H.; Fan, C.; Yin, S. Bioactive cembranoids from the South China Sea soft coral Sarcophyton elegans. *Molecules* **2015**, *20*, 13324–13335. [CrossRef]

10. Jia, R.; Guo, Y.-W.; Chen, P.; Yang, Y.-M.; Mollo, E.; Gavagnin, M.; Cimino, G. Biscembranoids and their probable biogenetic precursor from the Hainan soft coral Sarcophyton tortuosum. *J. Nat. Prod.* **2007**, *70*, 1158–1166. [CrossRef]
11. Liang, L.F.; Guo, Y.W. Terpenes from the soft corals of the genus Sarcophyton: Chemistry and biological activities. *Chem. Biodivers.* **2013**, *10*, 2161–2196. [CrossRef] [PubMed]
12. Yasumoto, M.; Mada, K.; Ooi, T.; Kusumi, T. New terpenoid components from the volatile oils of the soft corals Clavularia viridis and Sarcophyton acutangulum. *J. Nat. Prod.* **2000**, *63*, 1534–1536. [CrossRef]
13. Anjaneyulu, A.S.R.; Rao, V.L.; Sastry, V.G.; Rao, D.V. Trocheliophorin: A novel rearranged sesquiterpenoid from the Indian Ocean soft coral Sarcophyton trocheliophorum. *J. Asian Nat. Prod. Res.* **2008**, *10*, 597–601. [CrossRef] [PubMed]
14. Cheng, S.-Y.; Wen, Z.-H.; Chiou, S.-F.; Tsai, C.-W.; Wang, S.-K.; Hsu, C.-H.; Dai, C.-F.; Chiang, M.Y.; Wang, W.-H.; Duh, C.-Y. Ceramide and cerebrosides from the octocoral Sarcophyton ehrenbergi. *J. Nat. Prod.* **2009**, *72*, 465–468. [CrossRef]
15. Anjaneyulu, A.S.R.; Krishna Murthy, M.V.R.; Gowri, P.M. Novel epoxy steroids from the Indian ocean soft coral Sarcophyton crassocaule. *J. Nat. Prod.* **2000**, *63*, 112–118. [CrossRef]
16. Wang, Z.; Tang, H.; Wang, P.; Gong, W.; Xue, M.; Zhang, H.; Liu, T.; Liu, B.; Yi, Y.; Zhang, W. Bioactive polyoxygenated steroids from the South China sea soft coral, Sarcophyton sp. *Mar. Drugs* **2013**, *11*, 775–787. [CrossRef]
17. Cheng, Z.-B.; Deng, Y.-L.; Fan, C.-Q.; Han, Q.-H.; Lin, S.-L.; Tang, G.-H.; Luo, H.-B.; Yin, S. Prostaglandin derivatives: Nonaromatic phosphodiesterase-4 inhibitors from the soft coral Sarcophyton ehrenbergi. *J. Nat. Prod.* **2014**, *77*, 1928–1936. [CrossRef]
18. Sekhar, V.C.; Rao, C.B.; Ramana, H.; Kuwar, M.M.K.; Rao, D.V. New Prostaglandins from the soft coral Sarcophyton ehrenbergi marengeller of Andaman and Nicobar Islands of Indian ocean. *Asian J. Chem.* **2010**, *22*, 5353–5358.
19. Li, Y.-X.; Wu, H.-X.; Xu, Y.; Shao, C.-L.; Wang, C.-Y.; Qian, P.-Y. Antifouling activity of secondary metabolites isolated from Chinese marine organisms. *Mar. Biotechnol.* **2013**, *15*, 552–558. [CrossRef]
20. Sun, X.-P.; Wang, C.-Y.; Shao, C.-L.; Li, L.; Li, X.-B.; Chen, M.; Qian, P.-Y. Chemical constituents of the soft coral Sarcophyton infundibuliforme from the South China Sea. *Nat. Prod. Commun.* **2010**, *5*, 1171–1174. [CrossRef]
21. Iwagawa, T.; Nakamura, S.; Masuda, T.; Okamura, H.; Nakatani, M.; Siro, M. Irregular cembranoids containing a 13-membered carbocyclic skeleton isolated from a soft coral, Sarcophyton species. *Tetrahedron* **1995**, *51*, 5291–5298. [CrossRef]
22. Badria, F.A.; Guirguis, A.N.; El-Naggar, W.A. Antibacterial and antifungal agents from Egyptian marine organisms. *Int. J. Pharmacogn.* **1997**, *35*, 284–287. [CrossRef]
23. Badria, F.A.; Guirguis, A.N.; Perovic, S.; Steffen, R.; Müller, W.E.G.; Schröder, H.C. Sarcophytolide: A new neuroprotective compound from the soft coral Sarcophyton glaucum. *Toxicology* **1998**, *131*, 133–143. [CrossRef]
24. Hegazy, M.-E.F.; Eldeen, A.M.G.; Shahat, A.A.; Abdel-Latif, F.F.; Mohamed, T.A.; Whittlesey, B.R.; Paré, P.W. Bioactive hydroperoxyl cembranoids from the Red Sea soft coral Sarcophyton glaucum. *Mar. Drugs* **2012**, *10*, 209–222. [CrossRef] [PubMed]
25. Elkhateeb, A.; El-Beih, A.A.; Gamal-Eldeen, A.M.; Alhammady, M.A.; Ohta, S.; Paré, P.W.; Hegazy, M.-E.F. New terpenes from the Egyptian soft coral Sarcophyton ehrenbergi. *Mar. Drugs* **2014**, *12*, 1977–1986. [CrossRef] [PubMed]
26. Swapana, N.; Tominaga, T.; Elshamy, A.I.; Ibrahim, M.A.A.; Hegazy, M.-E.F.; Singh, C.B.; Suenaga, M.; Imagawa, H.; Noji, M.; Umeyama, A. Kaemgalangol A: Unusual seco-isopimarane diterpenoid from aromatic ginger Kaempferia galanga. *Fitoterapia* **2018**, *129*, 47–53. [CrossRef] [PubMed]