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New naturally occurring cembranoid diterpene derivatives from the soft corals family Alcyoniidae, 2016 - 2017

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Abstract

Work reviews the new isolated isolated cembranoid diterpene derivatives from species belonging to the family Alcyoniidae, which comprises the genera *Sarcophyton*, *Sinularia*, and *Lobophytum* as well as their biological properties, during 2016–2017. The compilation permitted to conclude that much more new cembranoid diterpenes were found in the soft corals of the genus *Sarcophyton* sp. (33 new compounds) than in those belonging to the genera *Lobophytum* (17) or *Sinularia* (8). Several methods have been used for identifying these new compounds, after extraction with organic solvents and fractionation. The fractions obtained, in some cases, were followed by TLC, and again subjected to chromatographic procedures, including semi-preparative HPLC. Beyond the chemical composition, the biological properties were also evaluated, namely anti-microbial against several Gram-positive and Gram-negative bacteria and fungi, anti-inflammatory and anti-tumoral against several types of cancer cells. Although the biological activities detected in almost all samples, they were not outstanding ones.

Keywords: *Sarcophyton*; *Sinularia*; *Lobophytum*; new compounds; anti-microbial; anti-inflammatory; anti-tumoral.

Contents

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1. Abbreviation

^{1}D-NMR: One dimensional nuclear magnetic resonance
^{2}D-NMR: Two dimensional nuclear magnetic resonance
^{1}H-^{1}H COSY: Correlation Spectroscopy
^{1}H-NMR: *Proton* nuclear magnetic resonance
^{13}C-NMR: Carbon-*13* nuclear magnetic resonance
A549: Adenocarcinomic human alveolar basal epithelial
BEAS-2B: Human bronchial epithelial
Caco-2: Heterogeneous human epithelial colorectal adenocarcinoma
CL-15: Lung adenocarcinoma
DFT : Density Functional Theory
EC_{50}: Half maximal effective concentration
ESI-MS: Electrospray ionization mass spectrometry
fMLP/CB: formyl-Met-Leu-Phe/cytochalasin B
FTIR: Fourier-transform infrared
H520: Squamous cell carcinoma
HCT116: Human colon cancer
HepG2: Liver hepatocellular carcinoma
HL-60: Human promyelocytic leukemia
HMBC: Heteronuclear Multiple-Bond Correlation Spectroscopy
HMQC: Heteronuclear Multiple-Quantum Correlation
HPLC: High Performance Liquid Chromatography
HR-ESI/FTMS: High Resolution-Electrospray Ionization/Fourier Transform Mass spectrometry
HRESIMS: High Resolution Electrospray Mass Spectrometry
HRMS: High resolution mass spectrometry
IC_{50}: Half maximal inhibitory concentration
IFN-γ: Interferon-γ
IG_{50}: Inhibition growth 50
IL-12: Interleukin 12
IR: Infrared
K-562: Human erythroleukemia
LC_{50}: Concentration that originates 50% mortality
LCMS-IT-TOF: Liquid chromatography–mass spectrometry-ion trap-time-of-flight)
LD_{50}: Median lethal dose
LPS: Lipopolysaccharide
MBC: Minimum Bactericidal Concentration
MIC: Minimum Inhibitory Concentration
MOLT-4: Acute lymphoblastic leukemia
MS: Mass spectrometry
NO: Nitric oxide
NOESY: Nuclear Overhauser Effect Spectroscopy
OPLS: Orthogonal projection to latent structures-discriminant analysis
PCA: Principal component analysis
PTP1B: Protein tyrosinase phosphatase 1B
qNMR: quantitative nuclear magnetic resonance
RAW 264.7: Murine macrophage
TDDFT/ECD: Time-Dependent Density Functional Theory Electronic Circular Dichroism
TLC: Thin layer chromatography
UPLC-MS: Ultra-performance liquid chromatography-mass spectrometry

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2. Introduction

The cembrane skeleton is isoprenoid and consists of a fourteen-membered carbocyclic ring with an isopropyl residue at position 1 and three methyl groups at positions 4, 8 and 12 (**Figure 1**). The basic structure of this diterpene usually presents cyclic ether, lactone, or furan moieties around the macrocyclic ring. There are also cembranoids variants which contain a 12 or 13- membered carbon skeleton (González et al., 2015; Weinheimer et al., 1979; Xi et al., 2013).

In nature, this class of diterpenoids has been found in marine invertebrates, lower and higher plants, insects (termites), and even paracloacal glands of male alligators (*Alligator sinensis*) (Polastro et al., 2016; Vasamsetty et al., 2014). Cembranoids from marine invertebrates are particularly isolated from soft corals of the genera *Sinularia*, *Lobophytum*, *Eunicea*, *Clavularia*, and *Sarcophyton*, and from the gorgonian octocorals, mainly of the genera *Pseudopterogorgia*, *Leptogorgia*, and *Lophogorgia* (Fattorusso et al., 2011; González et al, 2015; Li and Pattenden, 2011a).

Soft corals (phylum, Cnidaria; class, Anthozoa; subclass, Octocorallia; order, Alcyonaceae; family, Alcyoniidae) have been the target of study since the nineteenth century. The subclass Octocorallia includes soft corals, gorgonians, and sea pens. Most soft corals belong to the order Alcyonacea, which is comprised of the families Xeniidae, Nephtheidae, and Alcyoniidae. This family contains the genera *Sarcophyton*, *Sinularia*, and *Lobophytum* (Aratake et al., 212). Soft corals are found in Indo Pacific reefs whereas Gorgonian octocorals dominate the biomass in coral reef environments of the north-western Atlantic Ocean and in the Caribbean Sea (Li and Pattenden, 2011a).

In nature, cembranoids may act as chemical defence compounds against fish predators and/or competing for reef organisms, bacteria, parasites, that is for their protection and

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survival (Li and Pattenden, 2011a; Lai et al., 2017). Multiple biological properties of cembranoids of marine origin have been reported such as anti-inflammatory, anti-tumoral, anti-bacterial, anti-viral, neuroprotective, antiarthritic, calcium-antagonistic and cytotoxic (Lai et al., 2017; Yang et al., 2012a).

Yang et al. (2012a) review all the metabolites of cembrane diterpenes either from terrestrial or marine organisms up to 2010, divided into several different families according to the variety of ring sizes and oxidation patterns, as well as the respective biological activities. Several other reviews have been made regarding new compounds and their biological activities that have been isolated from marine microorganisms and phytoplankton, green, brown and red algae, sponges, cnidarians, bryozoans, molluscs, tunicates, echinoderms, mangroves and other intertidal plants, since 2013 until 2017 (Blunt et al., 2015, 2016, 2017, 2018). Marine invertebrates isolated from soft corals of the genera *Sinularia*, *Lobophytum*, *Eunicea*, and *Sarcophyton* are also included in these reviews.

Liang and Guo (2013), in a review on the terpenes from the soft coral of the genus *Sarcophyton* (*S. elegans*, *S. glaucum*, *S. ehrenbergi*, *S. trocheliophorum*, *S. molle*, *S. mililatensis*, *S. crassocaule*, *S. latum*, *S. cherbonnieri*, *S. stolidotum*, *S. tortuosum*, *S. infundibuliforme*, *S. flexuosum*, *S. solidum*, and some undefined species) from different geographical origins, reported 165 diterpenes, 29 biscembranoids, among other terpene compounds, during the period 1995 - July 2011. Some of these compounds possessed biological properties. In their review, Liang and Guo (2013), reported the work of Grkovic et al. (2011) who identified one cembranoid diterpene isolated from *Sarcophyton* sp..

The present work will review the new cembranoid diterpenes isolated from species belonging to the family Alcyoniidae, which contains the genera *Sarcophyton*, *Sinularia*, and *Lobophytum* as well as their biological properties, during 2016 and 2017. For this review, the *Web of Science* was used as a database for research, utilizing the keywords *cembrane*, and *cembranoid*.

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3. Chemical structure of cambranoids from marine origin

According to Rodríguez et al. (1993), the cembrane skeleton of marine origin is derived from the cyclization of geranylgeranyl pyrophosphate. This hypothesis is based on the fact that the double bonds of the cembrane skeleton have the geometry *E*, such as is observed in geranylgeraniol.

Cembrane diterpenoids have many structural variations with a multitude of functional groups (lactone, epoxide, furan, ester, aldehyde, hydroxyl, carboxyl moieties) and cyclizations, which permit to group them in several families (Blunt et al., 2011; Yang et al., 2012a).

According to the review of Yang et al. (2012a), the cembrane-type diterpenoids (**Figure 2**) may be classified as follows:

- Simple cembrene: isopropyl cembranes (e.g. sarcophytol M), isopropenyl cembranes (e.g. sinulariol), isopropyl/isopropenyl acid cembranes (e.g. flexibilisin A)
- Cembranolides: 5-membered lactone (e.g. deacetyldeepoxy lobolide), 6-membered lactone (e.g. manaarenolide A), 7-membered lactone (sinuladiterpene), 8-membered lactone (e.g. echinodolide)
- Furanocembranoids (e.g. bipinnatin A)
- Biscembranoids (e.g. lobophytone A)
- Special cembranes: *seco*cembranes (e.g. mayolide A), 13-membered carbocyclic cembranoids (e.g. sartol acetate), cembrane glycosides (e.g. calyculaglycoside A), cembrane-africanane (e.g. polymaxenolide), other cembranes(e.g. planaxool).

Cembranolides possess a 14-membered carbocyclic nucleus, generally fused to a 5-, 6- or 7-membered lactone ring. Furanocembranoids possess a 14-membered carbocyclic nucleus as well as a furan heterocycle. They also have a butenolide moiety involving C_{10}-C_{12}, as well as C_{20}. Biscembranoids possess a 14-6-14 membered tricyclic backbone of tetraterpenoids.

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(Yang et al., 2012a). The structure of polymaxenolide comprises a 14-membered cembranoid skeleton linked via a spiro ring system, to an africanane skeleton (**Figure 2**) (Kamel et al., 2009).

There are also the polycyclic norcembranoid diterpenes, rare and found exclusively in soft corals of the genus *Sinularia*. These diterpenes are within the family of furanecembranoids which lack a C_{18} carbon substituent in comparison with C_{20}-cembrenoids. They co-occur with 14-membered macrocyclic *nor*cembranoids with a furan heterocycle in which also lacks a C_{18} carbon substituent (Li and Pattenden, 2011b). According to the review made by Yang et al (2012a) with 189 references, the authors reported the source, chemistry and bioactivities of 644 new cembrane diterpenes from both terrestrial and marine organisms up to 2010.

The present work will review the source, chemistry and bioactivities of new cembrane diterpenes from marine organisms since 2016 until 2017. The most important sources of cembrane derivatives found in that period were those species belonging to the genus *Sarcophyton*, immediately followed by the genus *Sinalarina* and *Lobophytum*. For this reason, the brief review aims at identifying the new compounds found in those species belonging to these genera, during that period.

4. New cembrane derivatives from the genus *Sarcophyton*

The genus *Sarcophyton* presents a large number of species. Many of these species have been chemically examined. Some examples of groups of compounds include sesquiterpenes, diterpenes, diterpene dimmers, prostaglandins, steroids, ceramides, which have been extensively reviewed (Blunt et al., 2011; 2015; 2016; 2017; 2018; El-Ezz et al., 2017; Liang and Guo, 2013; Yang et al., 2012a). Among those metabolites, terpenes are the most frequently detected, possessing many biological properties (anti-inflammatory, anti-

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viral, anti-fouling, cytotoxic, neuroprotective) (Liang and Guo, 2013; Yang et al., 2012a and references therein).

Nine works regarding new cembranoid diterpenes from the genus *Sarcophyton* (*S. ehrenbergi*, *S. elegans*, *S. stellatum, S. subviride*, and *S. trocheliophorum*) were found, during the last two years. These species of soft corals were collected at several places of the Red Sea Coast and South China Sea. One sample was collected in the Indian Ocean, at the east coast of Madagascar (Table 1). The interest on researching in this subject did not decay since the number of publications is within the range of the precedent years. Beyond the publications regarding the discovery of new cembrane diterpenoids in the genus *Sarcophyton* as well as their biological properties, three other publications with distinct approaches were found. One of them aimed at examining the effect of oxylipin analogues and wounding on the secondary metabolism of the soft corals *Sarcophyton glaucum* (Farag et al., 2017a). The second one applied the quantitative NMR (qNMR) for assessing the diterpene variation in 16 soft coral specimens in the context of their genotype, origin, and growing habitat. The study revealed higher diterpene amounts in *Sarcophyton* sp. than in *Sinularia* or *Lobophyton* (Farag et al., 2017b). In their publication, Farag et al. (2017b) reported the metabolite profile of the soft coral genus *Sarcophyton* in different habitats along the coastal Egyptian Red Sea, was performed through *Proton* nuclear magnetic resonance (^{1}H-NMR) and ultra-performance liquid chromatography-mass spectrometry (UPLC-MS). At the same time, the authors compared the metabolite profile of these wild soft corals with those growing in aquarium. Generally, wild soft corals presented more bioactive compounds than aquarium grown ones. This discrepancy found between wild and aquarium grown corals were attributed, by the authors, to the less necessity for producing compounds acting as defences against predators, that are absent in tanks.

The large-scale metabolomics analyses were made for the first time in 16 *Sarcophyton* species, comparing MS and NMR results. This approach permitted to simultaneously identify

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120 metabolites including 65 diterpenes, 8 sesquiterpenes, 18 sterols, and 15 oxylipids. Both similarities and differences among samples were achieved after application of principal component analysis (PCA) and orthogonal projection to latent structures-discriminant analysis (OPLS). UPLC-MS revealed to be better tool for a compound based classification of coral species than NMR technique (Farag et al., 2016).

The extraction of cembranoid diterpenes was generally made with organic solvents, followed by the concentration under vacuum. Afterwards, the residue is partitioned between pairs of solvents and further column chromatography eluting with a gradient of solvents with increasing polarity. Different fractions originate the cembranoid compounds, which can be subjected to semi- or preparative HPLC. The identification of compounds is generally made through ^{1}H-NMR, ^{13}C-NMR (Carbon-*13* nuclear magnetic resonance), one dimensional and two dimensional nuclear magnetic resonance (1D-NMR and 2D-NMR) including ^{1}H-1H COSY, HMQC, HMBC, and NOESY spectra (Correlation Spectroscopy, Heteronuclear Multiple-Quantum Correlation, Heteronuclear Multiple-Bond Correlation Spectroscopy, Nuclear Overhauser Effect Spectroscopy, respectively), Time-Dependent Density Functional Theory Electronic Circular Dichroism (TDDFT/ECD), Density Functional Theory (DFT)/NMR calculations, FTIR (Fourier-transform infrared spectrosocopy), single crystal X-ray diffraction, and LCMS-IT-TOF (liquid chromatography–mass spectrometry-ion trap-time-of-flight) (Hegazy et al., 2017; Kamada et al., 2016; Li et al., 2017; Liang et al., 2017; Rahelivao et al., 2017; Shaaban et al., 2016; Sun et al., 2016; Tang et al., 2016; Zubair et el., 2016).

Kamada et al., (2016) from a Malaysian specimen of *Sarcophyton* sp., collected at the Karah Island (West Malaysia), isolated, identified and evaluated the antibacterial activity of 16-hydroxy cembra-1,3,7,11-tetraene (**Figure 3**), a new cembrane, along with the known cembrane diterpenes 15-hydroxycembra-1,3,7,11-tetraene, sarcophine and sarcophytoxide. Extracts were obtained by maceration of fresh soft coral with methanol, for several days and

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at room temperature. For isolating the compounds, the extraction was concentrated and partitioned between pairs of solvents and further column chromatography eluting with a gradient of *n*-hexane and ethyl acetate with increasing polarity. Different fractions originate the cembrane compounds. The identification was made through ^{1}H-NMR, ^{13}C-NMR, LCMS-IT-TOF, and FTIR.

The antimicrobial activity of all compounds was assayed against antibiotic resistant clinical bacterial strains *Staphylococcus aureus* and *Escherichia coli.* Only the new compound presented inhibition against *Staphylococcus aureus*. Its MBC (Minimum Bactericidal Concentration) and MIC (Minimum Inhibitory Concentration) were 75 and 25 µg/mL, respectively (Kamada et al., 2016).

From the South China Sea coral *S. elegans*, Li et al. (2017) isolated two novel biscembranoids, sarelengans A and B, five new cembranoids, sarelengans C–G (**Figure 4**), along with the two known cembranoids (sartrolide E and sarcophelegan B). Sarelengans B and C had moderate inhibitory activity against the lipopolysaccharide (LPS)-induced nitric oxide (NO) production in RAW264.7 macrophages. Their half maximal inhibitory concentration (IC_{50}) values were 18.2 and 32.5 µM, respectively.

Li et al. (2017) isolated those compounds after extraction of samples with ethanol at room temperature and after removal of solvent, the residue obtained was suspended in water and partitioned sequentially with crescent polarity solvents. Afterwards, the separation of the compounds was done through various column chromatographic techniques. The identification and the elucidation of chemical structures of the compounds were obtained using spectroscopic techniques, including 1D-NMR and 2D-NMR including ^{1}H-1H and COSY, HMQC, HMBC, and NOESY spectra, ^{13}C-NMR, high resolution electrospray ionisation mass spectrometry (HRESIMS), and single crystal X-ray diffraction for some compounds. The two novel biscembranoids had a *trans*-fused A/B-ring conjunction between the two cembranoid

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unities, in contrast to all biscembranoids. Such finding led the authors to hypothesize an unusual biosynthetic pathway of these compounds (Li et al., 2017).

Two new biscembranoid-like compounds were obtained from the soft coral *Sarcophyton subviride* from the coast of Xisha, Hainan Province (China) (Sun et al., 2016). They were bissubvilides A and B (**Figure 5**). These compounds did not present any cytotoxic activity against human osteosarcoma MG-63 (IC_{50} > 30 μM) or A549 lung cancer (IC_{50} > 25 μM) cells or Huh7 human hepatology cancer stem cells (IC_{50} > 50 μM).

The structures and absolute configurations of bissubvilides A and B were solved by spectroscopic analysis, TDDFT/ECD and DFT/NMR calculations, after exhaustively extracted with acetone at room temperature. The residue obtained after evaporation was partitioned between ether and water. The organic extract was reduced and subjected a column chromatography on silica gel and eluted with diverse eluents originating diverse fractions, which were again subjected to new chromatographic procedures giving new subfractions which were further purified on preparative HPLC (Sun et al., 2016).

Sarcoehrenbergilid A–C, three new cembane diterpenoids, along with two known cembrane diterpenoids, sarcophine, (+)-7α,8β-dihydroxydeepoxysarcophine (**Figure 6**), among other terpenoids, were isolated and characterized from the Red Sea soft coral *S. ehrenbergi* (Hegazy et al., 2017). Soft coral was extracted with methylene chloride and methanol at room temperature. After concentration, the dried material as subjected to the column chromatography on silica gel column and eluted with a gradient of solvents with increasing polarity. The fractions obtained were subjected to new chromatography technique (HPLC). Chemical structures were elucidated through ^{1}H-NMR, a ^{13}C-NMR, FT-IR (Fourier Transform-Infra Red), HR-ESI/FTMS (High Resolution-Electrospray Ionization/Fourier Transform Mass spectrometry), and X-ray Crystallography Data.

Cytotoxic activity of cembrane diterpenoids was performed using three human tumor cell lines (lung or A549; colon or Caco-2; and liver or HepG2). The compounds

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sarcoehrenbergilid A and C, (+)-7α,8β-dihydroxydeepoxysarcophine, sinulolide A, and sinulolide B were moderately active against A549 and HepG2, with IC_{50} = 43.6 – 98.6 µM (Hegazy et al., 2017).

The chemical composition and biological properties of *S. ehrenbergi* from the South China Sea were studied by Tang et al. (2016). This study led to the isolation and identification of eight cembrane diterpenoids, including the five new sarcophytonoxides A-E, and three known ones, (2*S*,11*R*,12*R*)-isosarcophytoxide, (+)-isosarcophine, and 8-hydroxyisosarcophytoxide-6-ene (**Figure 6**). The extraction of these compounds was made using acetone as an extraction solvent. Column chromatography, semi-preparative HPLC were the methods used for obtaining the fractions and subfractions were those metabolites were present. The identification of compounds was made through ^{1}H-NMR, a ^{13}C-NMR, IR and ESI-MS. All cembranoids were inactive (IC_{50} > 25 µM) against the human ovarian cancer cell line A2780 (Tang et al., 2016).

The antimicrobial activity of two new cembranoid diterpenes (sarcotrocheldiol A and B) and one new tetracyclic biscembrane hydrocarbon (trocheliane) (**Figure 7**) isolated from the Red Sea soft coral *Sarcophyton trocheliophorum* was evaluated by Zubair et al. (2016). Along with this new compounds, the known diterpene cembrene C was also isolated and identified from the same natural source. The coral material was exhaustively extracted with chloroform and methanol. The residue was partitioned between methylene chloride and water. The organic phase was further subjected to column chromatography using mixtures of eluents with increasing polarity. The fractions obtained were purified by preparative thin layer chromatography (TCL) on glass supported silica gel plates using adequate eluents. The structures of the compounds were elucidated using spectroscopic analysis such as 1D-NMR and 2D-NMR including ^{1}H-1H COSY, HMQC, HMBC, and NOESY spectra and ^{13}C-NMR (Zubair et al., 2016). Trocheliane was active against the two multidrug-resistant bacteria

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Acinobacter baumannii and *Staphylococcus aureus*. The MIC of this compound ranged from 4 to 6 µM for all the tested bacteria (*A. baumannii*, *S. aureus*, *S. epidermidis*, *Streptococcus pneumoniae*, *Escherichia coli*, *Klebsiella pneumonia*, and *Pseudomonas aeruginosa*) (Zubair et al., 2016).

Along with the known compounds sarcotrocheliol acetate, (+)-sarcophytol A, and (−)-sarcophytonin A (**Figure 7**), Shaaban et al. (2016) isolated and identified 9-hydroxy-10,11-dehydro-sarcotrocheliol, a new pyrane-based cembranoid diterpene, from the organic extract of the Red Sea soft coral *S. trocheliophorum*. The structures of the compounds were elucidated using ^{1}H-1H COSY, HMQC, HMBC, and NOESY spectra, and ^{13}C-NMR, as well as ESI-MS (electrospray ionization mass spectrometry) and HRMS (high resolution mass spectrometry) spectra, after extraction of samples reported by the authors (Shaaban et al., 2016) in previous publications (reported in Shaaban et al., 2016). All compounds isolated by the authors from the soft coral *S. trocheliophorum* did not possess any antimicrobial activity towards *Bacillus subtilis, S. aureus, Streptomyces viridochromogenes* (Tü 57), *Escherichia coli, Candida albicans, Mucor miehei, Chlorella vulgaris, Chlorella sorokiniana, Scenedesmus subspicatus, Rhizoctonia solani*, and *Pythium ultimum,* at 40 µg per disk. The cytotoxicity of the four compounds against brine shrimp was also absent (Shaaban et al., 2016).

Nine new cembranoids, sarcophytrols M–U (**Figure 7**), were isolated from the South China Sea soft coral *S. trocheliophorum*, along with one already known. Such new compounds possess diverse types of cyclized rings: furan rings in sarcophytrols M-P, pyran rings in sarcophytrols, oxepane and peroxyl rings in sarcophytrols T and U, respectively. Sarcophytrols R and S had a rare bicyclic skeleton of the decaryiol-type, as reported for the first time for the same genus of soft coral *S. decaryi* (Liang et al., 2017). The identification of these compounds was obtained after analysing them by several methods, including ^{1}H-1H

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COSY, HMQC, HMBC, and NOESY spectra, and ^{13}C-NMR, as well as HRMS spectra, polarimetry and infrared (IR). The extraction solvent used was acetone. The fractionation of samples was made using silica gel column chromatography and eluted with increasing polarity eluents. The bioassay for evaluating the capacity for inhibiting human protein tyrosinase phosphatase 1B (PTP1B) enzyme, important for the treatment of type-2 diabetes and obesity, all compounds isolated from the soft coral *S. trocheliophorum* did not provide positive results. Cytotoxicity against the human tumor cell lines HL-60 (Human promyelocytic leukemia cells) and K-562 (human erythroleukemia cells), as well as the antibacterial activity of the same compounds against *P. aeruginosa* also revealed negative (Liang et al., 2017).

Rahelivao et al. (2017) investigated three soft corals (*S. stellatum*, *Capnella fungiformis* and *Lobophytum crassum*) and the sponge *Pseudoceratina arabica* from the coast of Madagascar. Concerning *S. stellatum*, the authors reported a new (+)-enantiomer of the cembranoid (1*E*, 3*E*)-7,8-epoxycembra-1,3,11,15-tetraene produced by this organism, which structure was deduced from 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY). More three cembranoids were isolated and identified, by the authors, from *S. stellatum*: (+)-(7*S*,8*S*)-epoxy-7,8-dihydrocembrene C, (+)-(7*R*,8*R*,14*S*,1*Z*,3*E*,11*E*)-14-acetoxy-7,8-epoxycembra-1,3,11-triene, and (−)-(2*R*,7*R*,8*R*)-sarcophytoxide (**Figure 8**). The biological properties, particularly antiplasmodial activity against the FCM29 strains of *Plasmodium falciparum* and antimicrobial was only studied with some extracts of the sponge *Pseudoceratina arabica*. Only one extract of *S. stellatum*, the methanolic one, was biologically evaluated against *P. falciparum*. It presented only a moderate inhibition activity (IC_{50} = 35.20 µg/mL) (Rahelivao et al., 2017).

From dominant soft coral species of the genus *Sarcophyton* sp. on the reef at Mahengetang Island (Indonesia), Januar et al. (2017) isolated a new compound (2-hydroxy-crassocolide E) alongside with 5 known cembranoid compounds (sarcophytoxide, sarcrassin

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E, 3,7,11-cembreriene-2,15-diol, 11,12-epoxy-Sarcophytol A, and sarcophytol A (**Figure 9**). The structures were elucidated using 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY), and IT-TOF. All compounds inhibited the growth of human breast tumor cell lines MCF-7, being the IG_{50} (inhibition growth 50) value of 18.3 ppm for the new compound (Januar et al., 2017).

5. New cembrane derivatives from the genus *Sinularia*

Soft coral *Sinularia* consists of more than 150 species (Lai et al., 2011). As aforementioned for *Sarcophyton* sp., reviews have also been made regarding the discovery of new compounds and their biological activities up to 2015 (Blunt et al., 2011; 2015; 2016; 2017). During the period 2016-2017, only three publications could be found in the *Web of Science* utilizing the words *cembrane*, *cembranoid* and *Sinularia* (one study in 2016 and two studies in 2017). The species reported in these 3 works were *S. erecta*, *S. compacta* and *S. flexilibis*. These soft corals were collected predominantly in the South China Sea (Table 1). Publications regarding new cembranoid diterpenes produced by the genus *Sinularia* decreased comparatively to the years 2012 and 2013, in which in the same database (*Web of Science*), eight (Cheng et al., 2012; Lin et al., 2012; Mingli et al., 2012; Shen et al., 2012; Shih et al., 2012; Wright et al., 2012; Yang et al., 2012b; Yen et al., 2012) and seven (Aboutabl et al., 2013; Chen et al., 2013; Hu et al., 2013a, Hu et al., 2013b; Lin et al., 2013; Su et al., 2013; Yen et al., 2013) publications could be found, respectively.

Species of the genus *Sinularia* are rich in bioactive cembranoids and norcembranoids (Huang et al., 2016). These authors reported for the first time two new norcembranoids (sinulerectol A and B) (**Figure 10**), a new cembranoid (sinulerectol C) and a new degraded cembranoid sinulerectadione, alongside some known isoprenoids (norcembrene, sinularectin and ineleganolide) isolated from an extract of the marine soft coral *Sinularia erecta* from South China Sea (off the coast of Dongsha Atoll) (Huang et al., 2016). The structures of the

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compounds were determined by ^{1}H-NMR and ^{13}C-NMR, 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY) after extraction with ethyl acetate and further column chromatography over silica gel and adequate eluents with increasing polarity. Several fractions were obtained being subjected to a new chromatographic process, generally reversed-phase HPLC and further normal phase HPLC with the objective to obtain subfractions from where the compounds were isolated and identified.

Sinulerectadione exhibited inhibitory activity against myelogenous leukemia (K-562) and acute lymphoblastic leukemia (MOLT-4) cell lines with IC_{50} values of 8.6 and 9.7 µM, respectively, whereas sinulerectol C was effective against MOLT-4 cell lines (9.2 µM).

The anti-inflammatory activity of sinulerectols A and B on neutrophil pro-inflammatory responses was potent when evaluated by measuring the capacity for suppressing formyl-Met-Leu-Phe/cytochalasin B (fMLP/CB)-induced superoxide anion generation (IC_{50} = 0.9 and 3.8 µM, respectively) and elastase release in human neutrophils (113 and 93% inhibition at the same concentration, respectively) (Huang et al., 2016).

In the first report about chemical constituents of *S. compacta*, Wang et al. (2017a) reported three new compounds in the genus *Sinularia* (lobomichaolide, michaolide, and 20-acetylsinularolide), along with more eight compounds already found in the same genus of soft coral [presinularolide B, 14-acetoxy-3,4-epoxycembra-7,11,15-trien-17,2-olide, sinularolide C, 5-*epi*-sinuleptolide, 1-*iso*propyl-4,8,12-trimethyl-cyclotetradeca-2,4,7,11-tetraene, (1*R*,4*R*,2*E*,7*E*,11*E*)-cembra-2,7,11-trien-4-ol, sinulariol B, and sinulariol D] (**Figure 11**), all of them are 14-membred cembranoid diterpenes. The first six cembranoid diterpenes possess a α-methylene-γ-lactone moiety, make them cytotoxic, as well as anti-HIV and antituberculosis (Wang et al., 2017a). However, only 5-*epi*-sinuleptolide exhibited cytotoxic activity against the tumor cell lines HCT-116 and A-549 (IC_{50} values of 10.1 and 14.7 µM, respectively). Michaolide and 20-acetylsinularolide were lethal toward brine shrimp *Artemia*

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salina with lethal ratios of 90.5% and 90.0%, respectively, at a concentration of 50 µg/mL (Wang et al., 2017a).

The structures of all compounds were elucidated by ^{1}H-NMR, ^{13}C-NMR, and ESI-MS spectral data and by comparing with those previously reported elsewhere, after extraction with ethanol and then with methylene chloride and methanol. The fractions were obtained by column chromatography on silica gel and/or Sephadex LH-20 (Wang et al., 2017a).

Seven cembrane diterpenes were isolated from the soft coral of *S. flexibilis* from China (Sanya Bay, Hainan Island) [epoxycembrane A, sinularin, sinulariolide, (1*R*,13*S*,12*S*,9*S*,8*R*,5*S*,4*R*)-9-acetoxy-5,8:12,13-diepoxycembr-15(17)-en-16,4-olide, 11-dehydrosinulariolide, (-)14-deoxycrassin, and dihydrosinularin] (**Figure 12**). Epoxycembrane A was for the first time reported in *S. flexibilis* (Wang et al., 2017b)

Tributyltin and copper are antifouling largely used in order to deter marine fouling organisms on the surfaces of artificial structures submerged in the sea; nevertheless, they present some drawbacks particularly due to their adverse environmental impacts (Wang et al., 2017b). For this reason, several attempts have been made for finding more environmental friendly compounds. Wang et al. (2017b) assayed the antifouling activity on the larvae of the bryozoan *Bugula neritina* and the barnacle *Balanus albicostatus* of all the cembranoid diterpenes isolated from *S. flexibilis*. With the exception of sinularin, all remaining ones presented activity, and particularly (-)-14-deoxycrassin had the highest antifouling activity against both *Bugula neritina* and barnacle *Balanus albicostatus* [the concentrations of the compound that inhibited settlement by 50% relative to the control (EC_{50}) were 3.90 µg/mL and 21.26 µg/mL, respectively], and low toxicity against *B. albicostatus* larvae [the concentration that originates 50% mortality) (LC_{50}) > 100 µg/mL]. According to the authors (Wang et al., 2017b), the antifouling activity of epoxycembrane A 1*R*,13*S*,12*S*,9*S*,8*R*,5*S*,4*R*)-

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9-acetoxy-5,8:12,13-diepoxycembr-15(17)-en-16,4-olide, 11-dehydrosinulariolide, (-)14-deoxycrassin, and dihydrosinularin was reported for the first time.

The compounds were extracted from the soft coral with methanol and then fractionated between methylene chloride and water. The organic phase was subjected to a column chromatography and eluted with diverse solvents. The fractions obtained by this procedure were afterwards subjected to a reverse phase – HPLC to obtain the compounds. The structure of the compounds was elucidated by ^{1}H-NMR, ^{13}C-NMR and ESIMS.

6. New cembrane derivatives from the genus *Lobophytum*

The genus *Lobophytum* is rich in cembranoids and more than 250 different structures had been isolated from the genus *Lobophytum* (Lai et al., 2017). The number of publications about the structure of new cembranoid diterpenes and/or their biological properties remains without fluctuations during the last years. In 2014, three publication in *Web of Science*, using the terms *cembrane*, *cembranoid* and *Lobophytum* could be found (Cueng et al., 2014; Cuong et al., 2014; Thao et al., 2014), as well as in 2015 (Thao et al., 2015a; Thao et al., 2015b; Thao et al., 2015c), and 2016 (Al-Footy et al., 2016; Roy et al., 2016; Zhao et al., 2016). In 2017, the number of publications was 5 (Farag et al., 2017a; Lai et al., 2017, Lin et al. 2017; Mohamed et al., 2017; Rahelivao et al., 2017).

During 2016-2017, the most studied of soft coral species was *L. crassum*, either in terms of biological properties of known cembranoids or research of new cembranoid diterpenes. This species was collected in several places (Table 1). Species *L. crassum* is well known to produce oxygenated cembranoids. The structural variety of these metabolites is often correlated with geographic variation and environmental conditions (Zhao et al., 2016). The soft coral *L. crassum* from the South China Sea was studied by Zhao et al. (2016) and from this study, the authors isolated and identified nine new cembranoids [locrassumin A,B, D-G, (-)-laevigatol, (-)-*iso*surcophine, and (–)-7*R*,8*S*-dihydroxydeepoxysarcophytoxide], a

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diterpene possessing a tetradecahydrobenzo[3,4]cyclobuta[1,2][8]annulene ring system (locrassumin C) , and eight known cembranoids [(-)-sarcophytoxide, *ent*-sarcophine, sarcophytonolide O, sartrolide G, emblide, sarcrassin D, ketoemblide, and methyl sarcotroate B] (**Figure 13**). The known compounds were identified by comparison of their ^{1}H- and ^{13}C-NMR, MS spectroscopic data and specific rotations with those previously reported, whereas the elucidation of the new compounds were also made using ^{1}H-NMR and ^{13}C-NMR, 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY), and HRESIMS spectra (Zhao et al., 2016). The extraction was made with ethanol and after partitioned between ethyl acetate and water, the organic residue obtained was subjected to silica gel vacuum column chromatography using several gradient of a binary system constituted by ethyl acetate and petroleum ether. One of the fractions obtained were newly submitted to column chromatography but using other gradients of the same binary mixture. The fractions obtained were purified by semi-preparative HPLC. Some fractions were also submitted to Sephadex LH-20 column to obtain some of the cembranoid diterpenes (Zhao et al., 2016).

The anti-inflammatory activity of all compounds was evaluated, after measuring the lipopolysaccharide (LPS)-induced NO (nitric oxide) production in mouse peritoneal macrophages. Compounds locrassumin A, locrassumin D, *ent*-sarcophine, sarcophytonolide O, and ketoemblide exhibited moderate inhibition against LPS-induced NO production with IC_{50} values of 8–24 μM (Table 1). The remaining metabolites did not present inhibitory effect (IC_{50} > 30 μM) (Zhao et al., 2016).

From wild soft coral of *L. crassum*, collected around 8 m off the coast of Pingtung, Taiwan, was isolated and identified two new compounds (lobophyolide A and B), and the known cembranoid diterpenes [16-methoxycarbonyl-cembrene A, sinarone, sinulariol D, 16-acetyl-sinulariol D, and (*E*,*E*,*E*)-6,10,14-trimethy-3-methylene-*trans*-3*a*,4,7,8,11,12,15,15*a*-octahydrocy clotetradeca[*β*]furan-2(3H)-one) (**Figure 13**) (Lai et al., 2017). The elucidation

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of the structures of the compounds was made using ^{1}H-NMR and ^{13}C-NMR, 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY), and HRESIMS spectra (Lai et al., 2017). The extraction of the compounds was based on maceration with ethyl acetate and then the fractions were obtained by column chromatography, normal-phase and reverse-phase HPLC (Lai et al., 2017).

The anti-inflammatory activity of the cembranoid compounds was evaluated studying the effect of these compounds on the LPS-induced interleukin 12 (IL-12) release and NO production in dendritic cells (Lai et al., 2017). The results showed that lobophyolide A, 16-methoxycarbonyl-cembrene A and sinulariol D (<50 µg/mL) presented a potent inhibitory effect of IL-12 and NO release (86.1-96.2%). Moreover, the same compounds also had considerable cytotoxicity (Table 1) (Lai et al., 2017).

Mohamed et al. (2017) isolated from *L. crassum*, collected off the coast of Dongsha Atoll (South China Sea), three new cembrenoids (lobophylins F-H), together with three known ones (lobophylins A-C) (**Figure 13**). These compounds were isolated after extraction with ethyl acetate and fractionated by column chromatography on silica gel using *n*-hexane–ethyl acetate mixtures with increasing polarity. Some fractions obtained were further subjected to reverse-phase-HPLC. The elucidation of the structures was carried out by spectroscopic methods, namely IR, ^{1}H-NMR and ^{13}C-NMR, 1D and 2D-NMR measurements (COSY, HMBC, and NOESY), and ESIMS spectra.

Rahelivao et al. (2017) did not isolate cembranoids from the soft coral *L. crassum* extract from the coast of Madagascar, only reported the moderate activity of the crude methanol extract against the malarial parasite FCM29 strain of *Plasmodium falciparum* (IC_{50} value of 33.15 µg/mL). In other work, Lin et al. (2017) studied the anticancer ability of lobocrassin B (**Figure 13**), a natural cembrane diterpenoid previously isolated from the soft coral *L. crassum*. The authors reported that this compound exerted cytotoxic effects for

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concentrations < 10 µM on lung cancer CL-15 and H520 cells lines, not only by decreasing cell viability but also by inducing apoptosis, oxidative stress and mitochondrial dysfunction (increased level of Bax, cleaved caspase-3, -9 and -8, and suppression of Bcl-2). Nevertheless, much higher concentration was necessary to add to the normal human bronchial epithelium (BEAS-2B) for exerting cytotoxic effect (> 25-50 µM), which means that lobocrassin B preferably causes cell death of carcinogenic cells than normal cells.

Al-Footy et al. (2016) isolated diverse secondary metabolites (sesquiterpenes, steroid type compounds and only one known cembrane diterpene) from the soft coral *Lobophytum* sp. were collected off the Red Sea Coast, at Jeddah, Saudi Arabia. The isolated cembrane diterpene was cembrene A. This cembrane diterpene showed moderate antibacterial activity against *Acinetobacter* sp., *E.coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pneumonia*, nevertheless, presented high toxicity against brine shrimp *A. salina* and antitumor activity against Erhlich carcinoma cells with median lethal dose (LD_{50}) values of 25 and 50 µg/mL, respectively.

Roy et al. (2016) isolated and identified 7 cembrane-type diterpenes from the coast of Irabu Island (Okinawa, Japan), a soft coral *Lobophytum* sp.: a new rare casbane-tipe diterpenoid 1, two new cembrane diterpenoids (2 and 3); and four known cembrane diterpenoids (4-7). The authors did not attribute names for the structures presented (**Figure 14**). The structures of the compounds were obtained by analysis of spectroscopic data, using IR, ^{1}H-NMR and ^{13}C-NMR, 1D and 2D-NMR measurements (COSY, HSQC, HMBC, and NOESY), and HRESIMS spectra, after extraction with acetone and fractionation by chromatographic processes. The authors reported that the compounds 1-5 showed weak anti-bacterial activity (*Staphylococcus aureus*, *Salmonella enterica* and *E. coli*). Compounds 1-3 showed moderate cytotoxicity against human colon cancer cells (HCT116) with IC_{50} values ranging from 135.57 to 177.11 µM, and anti-inflammatory activity in LPS/IFN-γ

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(LPS/interferon-γ)-stimulated RAW 264.7 macrophages cells (IC_{50} 41.21 – 74.76 μM (Roy et al., 2016).

7. Concluding remarks

The use of increasingly sophisticated equipment has permitted to identify new compounds, including natural compounds of marine origin, generally with the aim to find remarkable biological properties for possible application in Medicine. A huge diversity of chemical structures have been isolated and evaluated in biological terms from marine organisms. The soft corals belonging to the family Alcyoniidae are not an exception, and therefore they are the target of several studies in searching new products with biological properties, particularly antimicrobial, anti-inflammatory and anti-tumoral activities. Cembranoids from soft corals of the genera *Sinularia*, *Lobophytum*, and *Sarcophyton* are the most well studied secondary metabolites of the specimens belonging to these genera. The chemical diversity attenuation of cembranoid diterpenes is remarkable and evident from previous studies, which can be attributed to the differences in environmental conditions between the different localities, such as surface temperature, salinity, nutrient concentrations and turbidity. For this reason, there are teams of researchers who have studied the diterpene variation in diverse soft coral specimens in the context of their genotype, origin, and growing habitat. They also intended to find a tool for a compound based classification of coral species (Farag et al., 2017a,b). Such studies revealed that UPLC-MS is a better tool than NMR spectroscopy. In addition, higher diterpene amounts were found in *Sarcophyton* sp. than in *Sinularia* or *Lobophyton*. Maybe for this reason, much more new compounds were reported in *Sarcophyton* sp., during 2016 and 2017, than in the remaining specimens of the genera *Sinularia* or *Lobophyton*.

8. Conflicts of interest

There are no conflicts to declare.

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9. References

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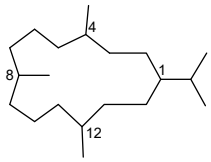

Figure 1. Cembrane skeleton.

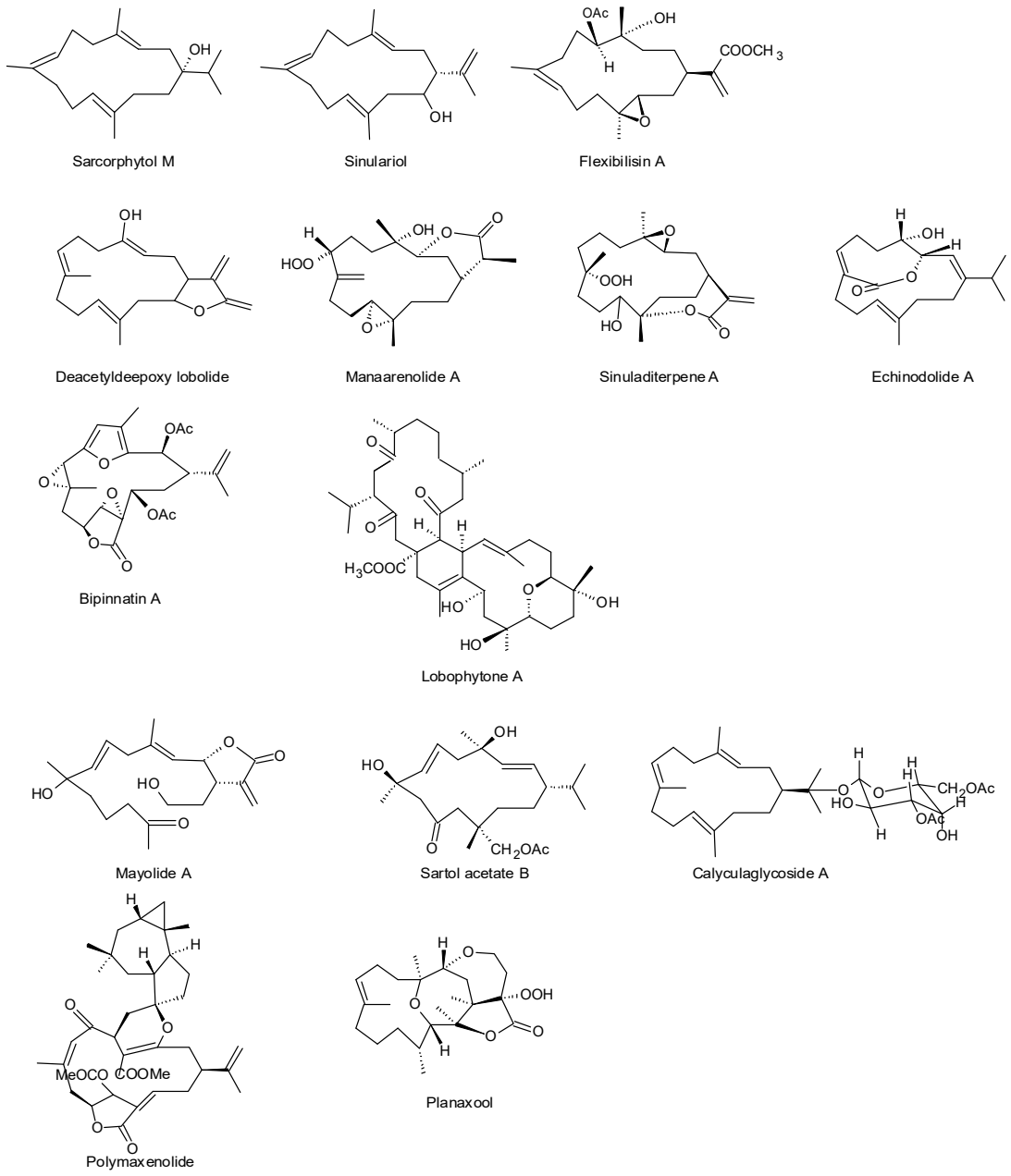

Figure 2. Some structures of marine cembranoids belonging to diverse chemical groups.

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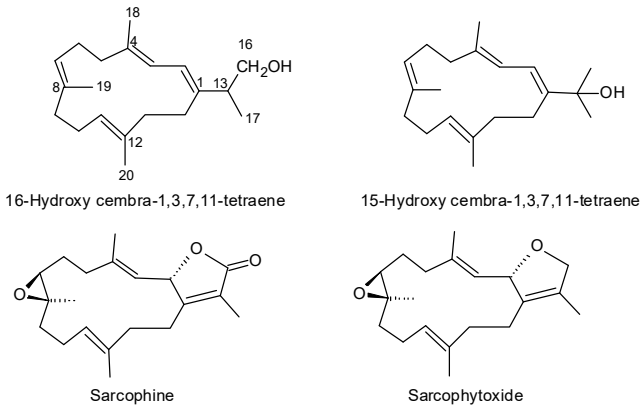

Figure 3. Cembranoid diterpenes isolated from *Sarcophyton* sp., collected at the Karah Island (West Malaysia) Kamada et al. (2016).

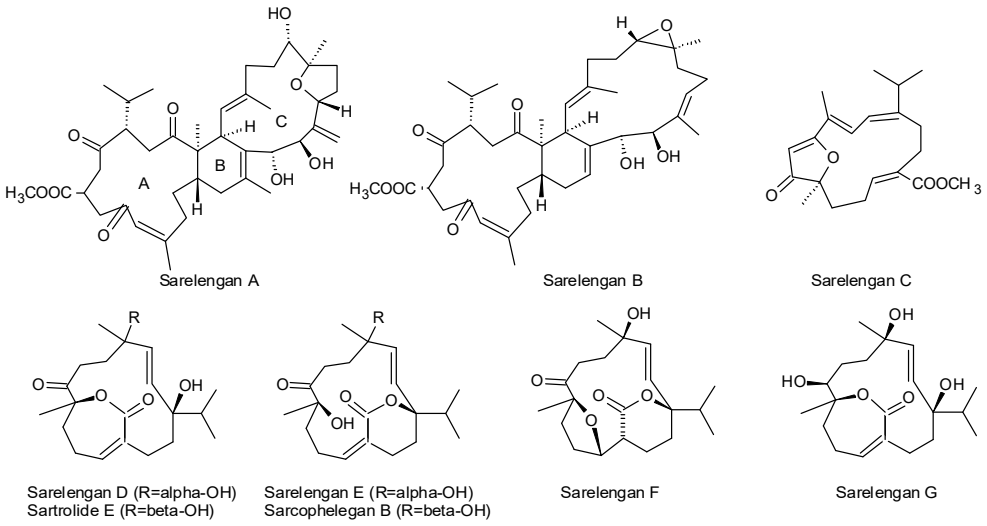

Figure 4. Biscembranoids and cembranoid diterpenes isolated from *Sarcophyton elegans*, collected at Xisha Islands in the South China Sea (Li et al., 2017).

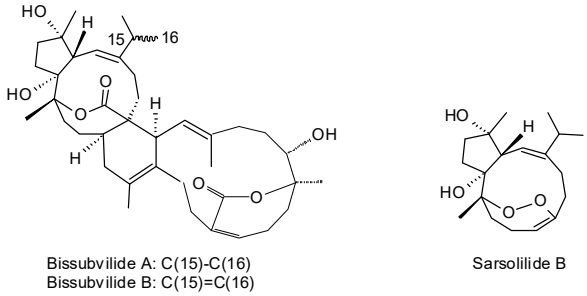

Figure 5. Biscembranoid-like compounds, bissubvilides A and B together with sarsolilide B from the soft coral *Sarcophyton subviride*.

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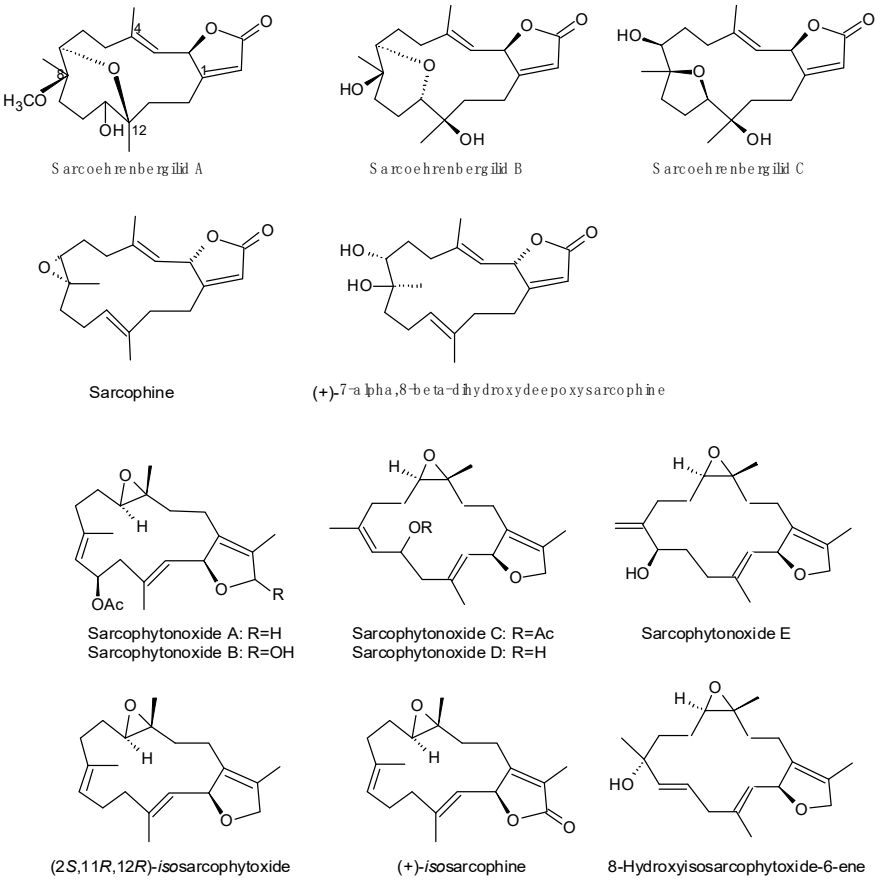

Figure 6. Cembranoid diterpenes isolated from *S. ehrenbergi*, from the Red Sea and South China Sea (Hegazy et al., 2017; Tang et al., 2016).

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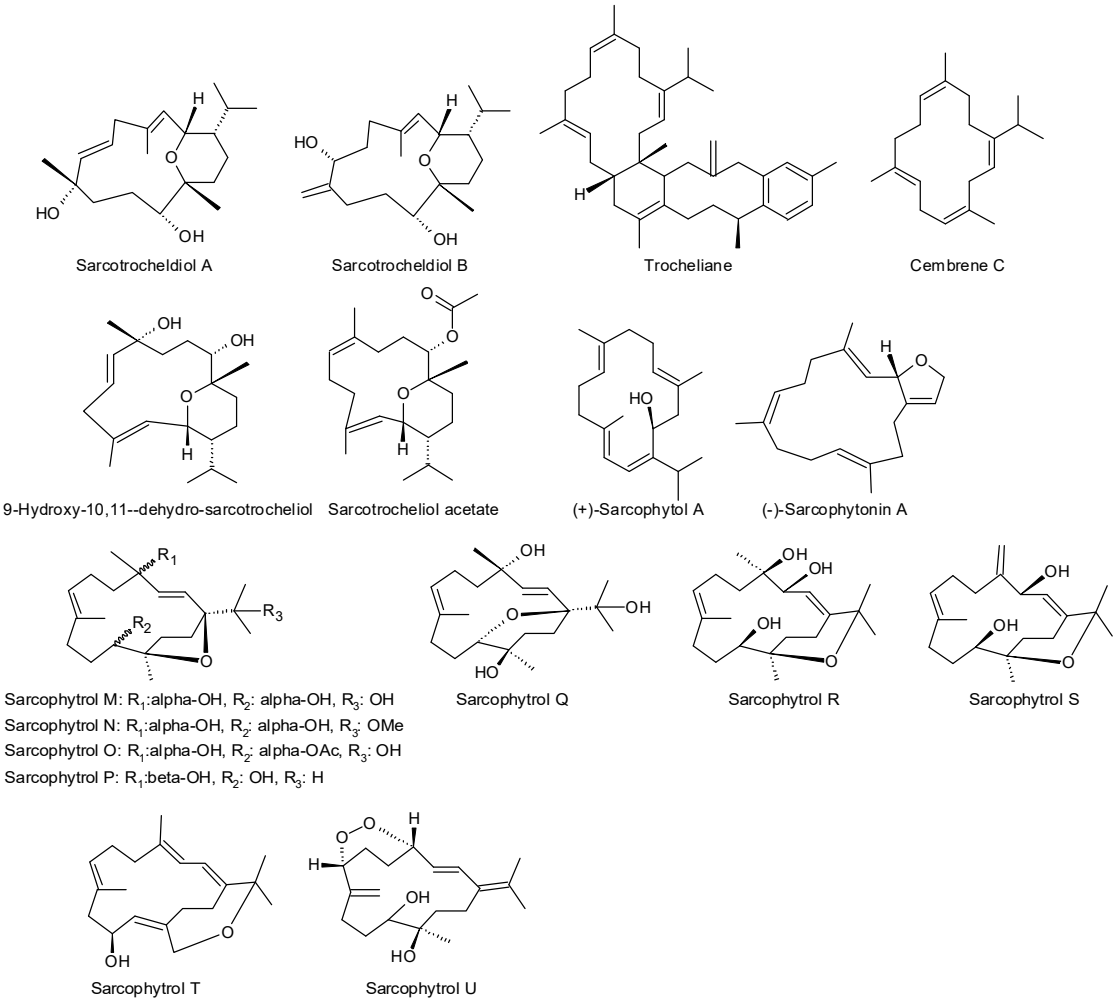

Figure 7. Cembranoid diterpenes isolated from *S. trocheliophorum*, from the Red Sea and South China Sea (Zubair et al., 2016; Shaaban et al., 2016; Liang et al., 2017).

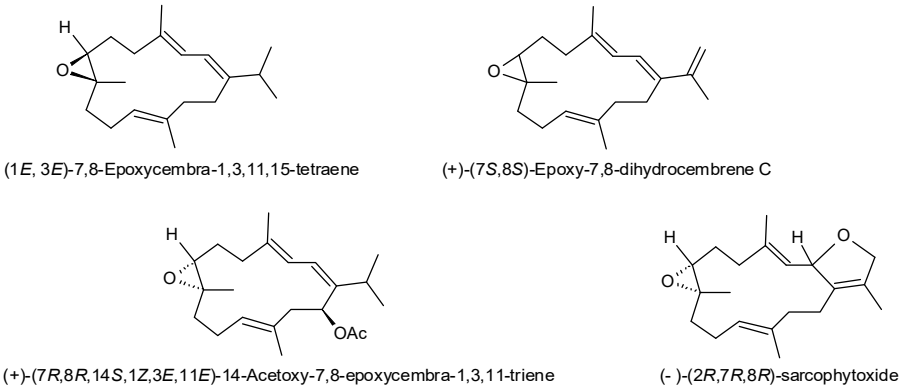

Figure 8. Cembranoid diterpenes isolated from *S. stellatum*, from the coast of Madagascar (Rahelivao et al., 2017).

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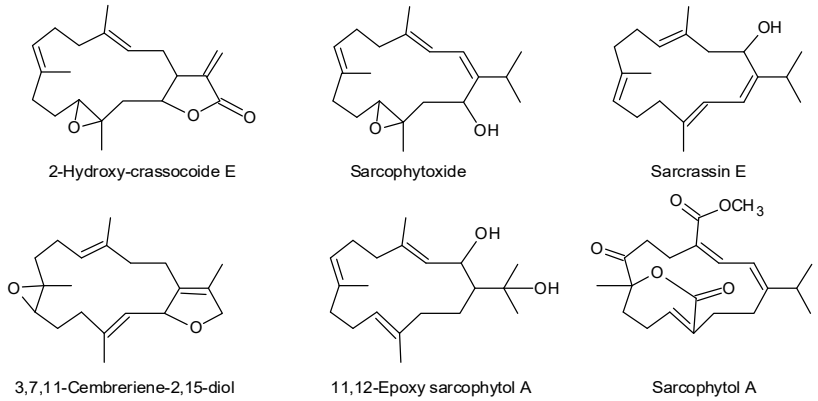

Figure 9. Cembranoid diterpenes isolated from *Sarcophyton* sp. on the reef at Mahengetang Island (Indonesia) (Januar et al., 2017).

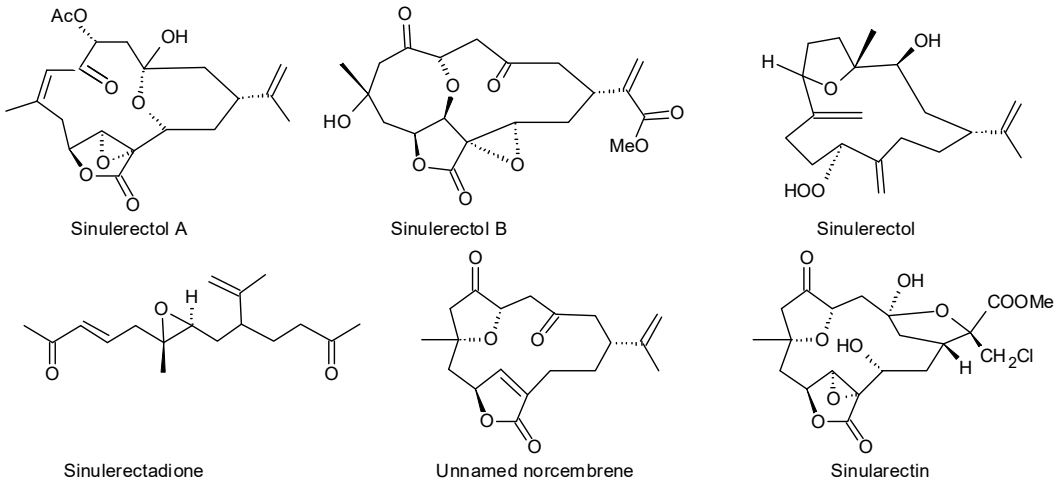

Figure 10. Cembranoid diterpene derivatives isolated from *Sinularia erecta* from South China Sea (Huang et al., 2016).

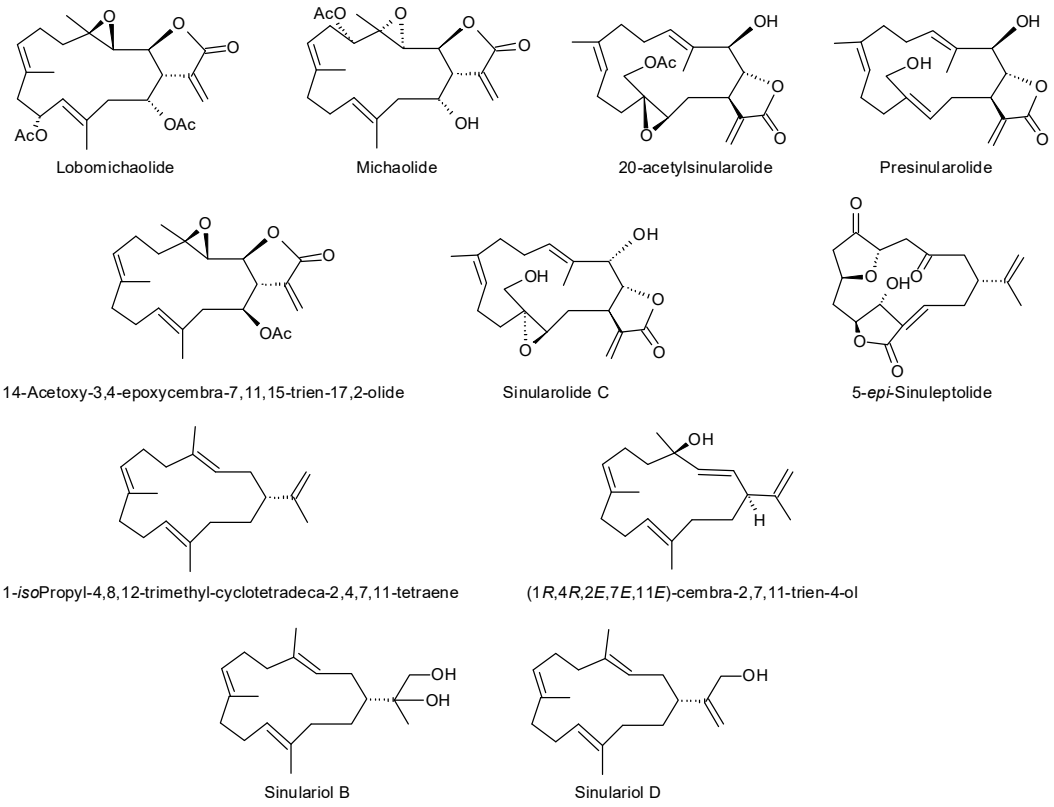

Figure 11. Cembranoid diterpene derivatives isolated from *Sinularia compacta* from the South China Sea (Tongguling National Nature Reserve of Coral Reefs) (Wang et al., 2017a).

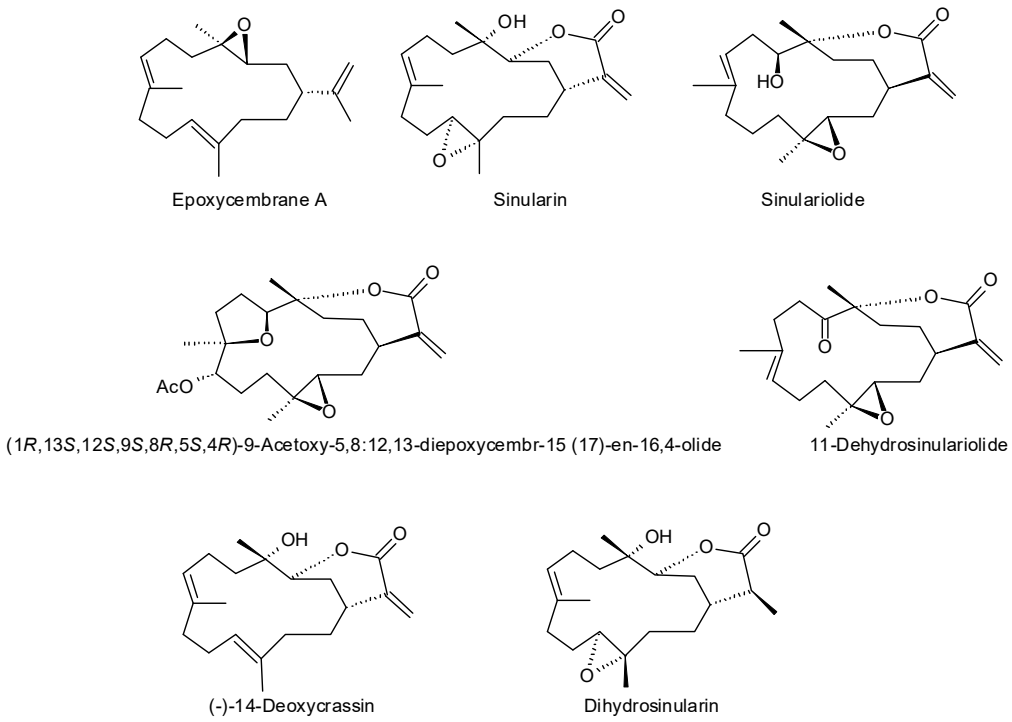

Figure 12. Cembranoid diterpenes from *Sinularia flexibilis* from China (Sanya Bay, Hainan Island) (Wang et al., 2017b).

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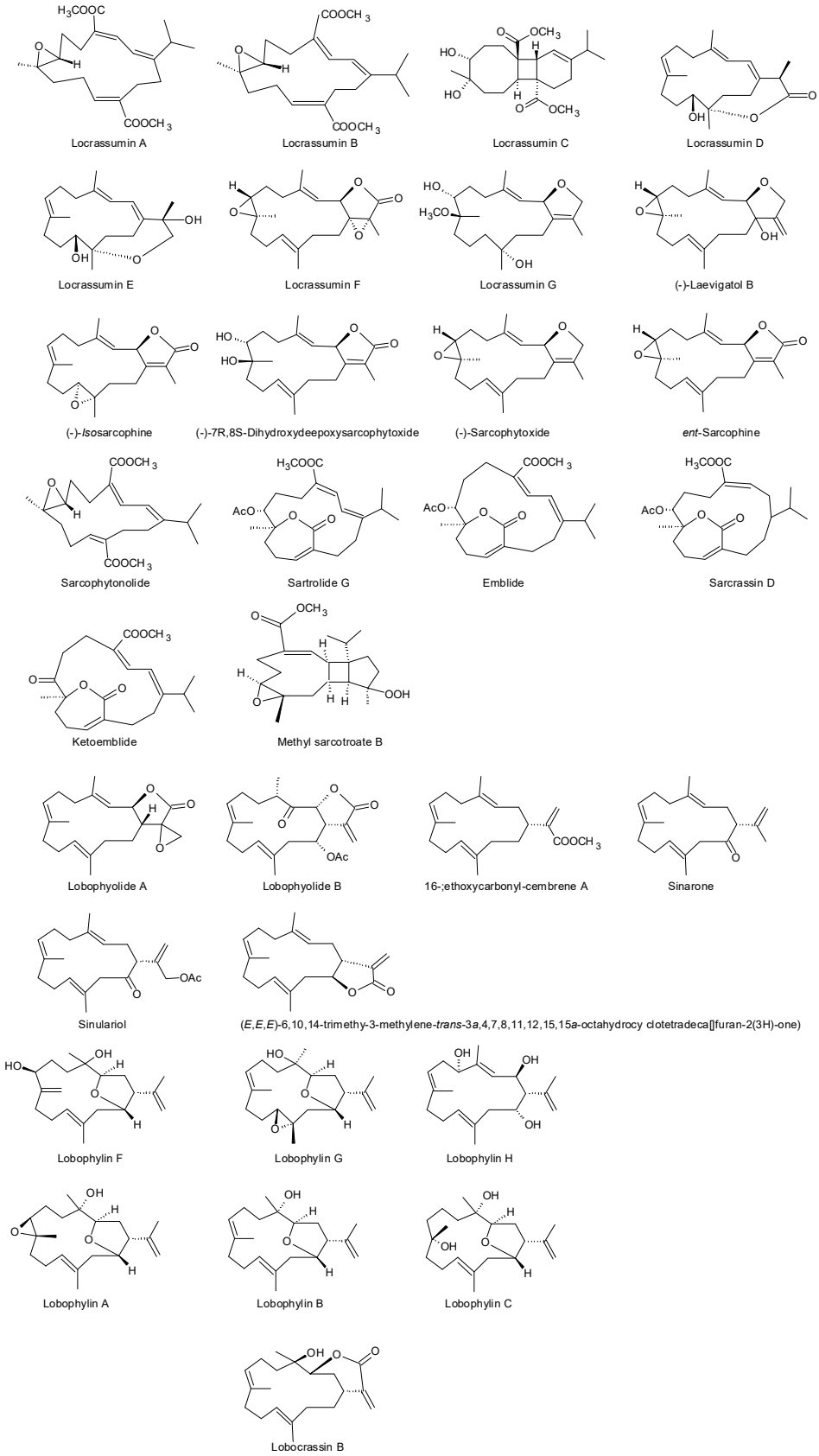

Figure 13. Cembranoid diterpenes from Lobophytum crassum from China (South China Sea and Twain) (Lai et al., 2017; Lin et al., 2017; Mohamed et al., 2017; Zhao et al., 2016).

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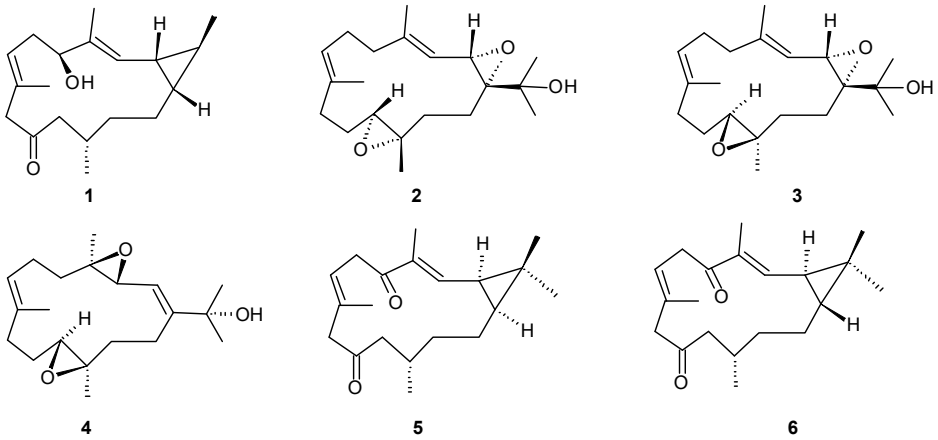

Figure 14. Cembrane-type diterpenes from a soft coral *Lobophytum* collected in the coast of Irabu Island (Okinawa, Japan) (Roy et al., 2016).

[1] **Table 1**. Harvesting locations of the soft corals of the genera *Sarcophyton*, *Sinularia*, and *Lobophytum*, new compounds identified and their biological properties

Soft coral	New bioactive cembranoid diterpene	Biological activities	Location	Reference
Genus *Sarcophyton*				
			Red Sea Coast	
Sarcophyton trocheliophorum	Trocheliane	Activity against the two multidrug resistant bacteria *Acinobacter baumannii* and *Staphylococcus aureus* (MIC=4.2 and 4.0 µM, respectively)	North of Jeddah, Saudi Arabia, Red Sea Coast (21º29'31''N, 39º11´24''E)	Zubair et al. (2016)
Sarcophyton trocheliophorum	9-Hydroxy-10,11-dehydro-sarcotrocheliol	- Inactive against *Bacillus subtilis, Staphylococcus aureus, Streptomyces viridochromogenes* (Tü 57), *Escherichia coli, Candida albicans, Mucor miehei, Chlorella vulgaris, Chlorella sorokiniana, Scenedesmus subspicatus, Rhizoctonia solani*, and *Pythium ultimum* at 40 µg per disk. - no cytotoxicity on against brine shrimp at a concentration 10 µg/mL (24 h)	Red Sea	Shaaban et al. (2016)
Sarcophyton ehrenbergi	Sarcoehrenbergilid A–C	Moderate anti-proliferative activities against two human tumor cell lines: lung (A549) (IC_{50}=50.1-76.4 µM), and liver (HepG2) (IC_{50}=53.8 µM, only for Sarcoehrenbergilid C), and weak activity against colon (Caco-2) (IC_{50}>100 µM).	Hurghada (Egyptian Red Sea costal)	Hegazy et al. (2017)
			South China Sea	
Sarcophyton sp.	6-Hydroxycembra-1,3,7,11-tetraene	Antibacterial activity against *Staphylococcus aureus* (MBC and MIC values were 75 µg/mL and 25 µg/ mL, respectively. The MBC/MIC ratio was calculated to be 3.0 which indicated that the compound exhibits bactericidal	Karah Island, Terengganu, West Malaysia (5º35'52.6''N,103º03'47.0E)	Kamada et al. (2016)

		activity.		
Sarcophyton elegans	- The biscembranoids sarelengans A and B - The cembranoids sarelengans C–G	Sarelengans B and sarelengans C showed moderate inhibitory activities on nitric oxide production in RAW264.7 macrophages, with IC_{50} values being at 18.2 and 32.5 μM, respectively	Coast of Xisha Island	Li et al. (2017)
Sarcophyton subviride	The biscembranoid-like compounds bissubvilides A and B	These two molecules did not exert any cytotoxicity against human osteosarcoma MG-63 (IC_{50} > 30 μM) or A549 lung cancer (IC_{50} > 25 μM) cells or Huh7 human hepatology cancer stem cells (IC_{50} > 50 μM)	Coast of Xisha Island	Sun et al. (2016)
Sarcophyton ehrenbergi	Sarcophytonoxides A–E	All of the cembranoids were inactive against the human ovarian cancer cell line A2780 (IC_{50} > 25 μM)	North Reef (Beijiao), Xisha Islands	Tang et al. (2016)
Sarcophyton trocheliophorum	The bicyclic cembranoids sarcophytrols M–U	No inhibitory activity against human protein tyrosine phosphatase 1B (PTP1B) enzyme, target for the treatment of type 2 diabetes and obesity. No cytotoxicities against the human tumor cell lines HL-60 and K-562, nor antibacterial activity against *Pseudomonas aeruginosa.*	Yalong Bay, Hainan Province	Liang et al (2017)
			Indian Ocean	
Sarcophyton stellatum	(+)-Enantiomer of the cembranoid (1*E*,3*E*,11*E*)-7,8-epoxycembra-1,3,11,15-tetraene	Not determined, only the crude methanol extract. This showed moderate antimalarial activity (FCM29 strain of *Plasmodium falciparum*): IC_{50} = 35.20 μg/mL	Inner reef of Mohambo, Tamatave province, the east coast of Madagascar (17º29'15.0''S, 49º28'32.1''E)	Rahelivao et al. (2017)
		Genus *Sinularia*		
			South China Sea	
Sinularia compacta	Lobomichaolide, michaolide F, 20-acetylsinularolide B	Michaolide F and 20-acetylsinularolide B exhibited lethality toward brine shrimp *Artemia salina* with lethal ratios of 90.5% and 90.0% at a concentration of 50	Tongguling National Nature Reserve of Coral Reefs	Wang et al. (2017a)

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		µg/mL, respectively		
Sinularia erecta	The two norcembranoids sinulerectols A and B, a cembranoid sinulerectol C, and a degraded cembranoid sinulerectadione	- Sinulerectadione exhibited cytotoxicity toward K-562 and MOLT-4 cancer cell lines with IC50 values of 8.6 and 9.7 ± 2.9 µM, respectively. Sinulerectol C showed cytotoxicity toward the K-562 cell line with an IC50 value of 9.2 µM. - Sinulerectols A and B exhibited potent anti-inflammatory activities in the inhibition of superoxide generation and elastase release. - Sinulerectol C only exhibited significant activity in inhibiting elastase release	Coast of Dongsha Atoll	Huang et al. (2016)
Sinularia flexibilis	Epoxycembrane A	Antifouling activity against the bryozoan *Bugula neritina* and the barnacle *Balanus albicostatus* (EC_{50} = 21.37 and 30.60 µg/mL, respectively)	Sanya Bay, Hainan Island	Wang et al. (2017b)
		Genus *Lobophytum*		
			Indian Ocean	
Lobophytum crassum	No cembranoids were isolated	Moderate activity of the crude methanol extract against the malarial parasite FCM29 strain of *Plasmodium falciparum* (IC_{50} value of 33.15 µg/mL)	Inner reef of Mohambo, Tamatave province, the east coast of Madagascar (17º29'15.0''S, 49º28'32.1''E)	Rahelivao et al. (2017)
			Red Sea Coast	
Lobophytum sp.	Cembrene A (this is not new). This cembrane diterpene showed	Moderate antibacterial activity against *Acinetobacter* sp., *E.coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pneumonia* - High toxicity against brine shrimp *Artemia salina* (LD_{50} = 25 µg/mL) - Antitumor activity against Erhlich carcinoma cells (LD_{50} = 50 µg/mL, respectively	Saudi Arabia Red Sea Coast at Jeddah	Al-Footy et al. (2016)

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			South China Sea	
Lobophytum crassum	Lobophylins F-H	Not evaluated	Coast of Dongsha Atoll	Mohamed et al. (2017)
Lobophytum crassum	Lobophyolide A and B	- Both (<50 µg/mL) presented a potent inhibitory effect on IL-12 and NO release (inhibition rates of > 90%) in LPS-activated dendritic cells - Lobophyolide A and B also had considerable cytotoxicity with survival percentage of dendritic cells, under the concentration of 50 µg/mL, of 76 and 52, respectively.	Coast of Pingtung, Taiwan	Lai et al. (2017)
Lobophytum crassum	Locrassumins A–G, (–)-laevigatol B, (–)-*iso*sarcophine, (–)-7*R*,8*S*-dihydroxydeepoxysarcophytoxide	Locrassumins A and G showed moderate inhibition against LPS-induced NO production in mouse peritoneal macrophages with IC50 values of 17 and 13 µM, respectively. No inhibitory effect was observed for the other compounds (IC50 > 30 µM)	Inner coral reef of Meishan, Hainan Province	Zhao et al. (2016)
			East China Sea	
Lobophytum sp.	Compound 1 (a new rare casbane-tipe diterpenoid), two new cembrane diterpenoids (Compounds 2 and 3)	- Weak anti-bacterial activity (*Staphylococcus aureus*, *Salmonella enterica* and *E. coli*) - Moderate cytotoxicity against human colon cancer cells (HCT116) with IC_{50} values ranging from 135.57 to 177.11 µM - Anti-inflammatory activity in LPS/IFN-γ (LPS/interferon-γ)-stimulated RAW 264.7 macrophages cells (IC_{50} 41.21 – 74.76 µM)	Irabu Island, Okinawa, Japan	Roy et al. (2016)

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