ANTIMICROBIAL, ANTIVIRAL AND CYTOTOXIC ACTIVITIES OF SELECTED MARINE ORGANISMS COLLECTED FROM THE COASTAL AREAS OF MALAYSIA

Yik Sin Chan¹, Cheong Wei Ong¹, Bee Ling Chuah¹, Kong Soo Khoo², Fook Yee Chye³, and Nam Weng Sit¹

Key words: antibacterial, antifungal, seaweed, sponge, soft coral.

ABSTRACT

Many marine organisms have developed the capability of producing unique metabolites and thus are highly likely to contain anti-infective agents. This study was conducted to investigate extracts of three seaweeds (Caulerpa racemosa, Caulerpa serrulatioides, Kappaphycus alvarezii), two soft corals (Lobophytum microlobulatum, Sarcophyton auritum) and a marine sponge (Spheciospongia vagabunda) collected from Malaysian coast for antibacterial, antifungal, antiviral and cytotoxic activities. The samples were subjected to sequential solvent extraction in order to obtain hexane, chloroform, ethyl acetate, ethanol, methanol and water extracts. The antibacterial and antifungal activities were studied using a colorimetric broth microdilution method. The hexane extract of L. microlobulatum had the strongest antibacterial activity and exhibited the lowest minimum inhibitory concentration (0.04 mg/mL) and minimum bactericidal concentration (0.08 mg/mL) against Staphylococcus aureus and Bacillus cereus, respectively. For antifungal activity, the lowest MIC and minimum fungicidal concentration values were produced by the hexane extract of S. auritum against the dimorphic yeast Cryptococcus neoformans, both with 0.04 mg/mL. None of the extracts were active against the filamentous fungus Aspergillus fumigatus. Only the hexane and ethanol extracts of L. microlobulatum and the ethyl acetate extract of S. auritum exhibited strong inhibition on the cytotoxic effect induced by the Chikungunya virus (a re-emerging mosquito-borne virus) with 50% effective concentrations of 14.3 ± 0.2, 124.3 ± 1.9 and 176.6 ± 9.7 µg/mL, respectively. Extracts from the two soft corals, L. microlobulatum and S. auritum possessed stronger antibacterial, antifungal and antiviral activities compared to the seaweeds and the sponge.

I. INTRODUCTION

Despite advances in medical technology and scientific knowledge, infectious diseases remain as a leading cause of morbidity and mortality (Tekwu et al., 2012). Bacteria have been recognized as one of the major etiological agents for lower respiratory tract infections and diarrheal diseases, which are among the top 10 killers according to the World Health Organization (WHO) estimates (WHO, 2015a). The incidence of fungal infections, such as candidiasis, cryptococcosis and aspergillosis has significantly increased over the past few decades (Debourgogne et al., 2016; Vallabhaneni et al., 2016). Candidiasis is a major health threat for human immunodeficiency virus/acquired immunodeficiency syndrome (HIV/AIDS) patients.

Antibiotics or antimicrobial drugs used against many bacterial or fungal diseases are rapidly losing their effectiveness as microorganisms develop resistance towards them. Due to the advent of new resistance mechanisms, this has resulted in prolonged illness and increased risk of death (Tanwar et al., 2014). In the European Union alone, an estimated 25,000 deaths were caused by drug-resistant microorganisms and costing more than US 1.5 billion dollars every year in healthcare expenses and productivity losses (WHO, 2015b). One of the measures to minimize the increasing rate of microbial resistance is to have a continuous investigation for new, safe and effective alternative agents.

The Chikungunya virus is a re-emerging mosquito-borne virus that causes Chikungunya fever, typically with acute and chronic musculoskeletal pain in humans. Since 2004, the virus...
has caused millions of cases of disease in the Indian Ocean region and emerged to Europe, Middle East and Pacific regions (Morrison, 2014). The latest outbreaks of Chikungunya fever occurred in Mandra East sub-county of Kenya (WHO, 2016) and in Karachi, Pakistan (Aamir et al., 2017). Since many patients are not reporting to healthcare facilities, it is likely that the incidence of cases is underestimated (WHO, 2016). Patients with Chikungunya fever are only treated symptomatically with anti-inflammatory and analgesic drugs as there are no specific drugs against the virus (Caglioti et al., 2013).

The marine environment is a remarkable pool of a wide variety of organisms which are producers of novel and bioactive secondary metabolites with diverse chemical structures (Devi et al., 2011). However, when compared with terrestrial plants, the use of marine organisms in folk medicine is very restricted because of their relative inaccessibility (Blunden, 2001). Many marine organisms produce secondary metabolites with great pharmaceutical potential. There are four marine natural product-derived drugs approved for clinical use: cytarabine which has antancer properties, vidarabine which acts against herpes simplex and varicella zoster viruses, ziconotide which has analgesic activity and trabectedin which is used for the treatment of ovarian carcinoma and soft tissue sarcoma (Cavalcante-Silva et al., 2013).

Seaweeds are classified into three main divisions: green (Chlorophyta), brown (Phaeophyta) and red (Rhodophyta) (Wang et al., 2014). Caulerpa racemosa and Caulerpa sertularioides, belonging to Chlorophyta are commonly known as sea grapes and green feather alga, respectively. Kappaphycus alvarezii belongs to the Rhodophyta and is commonly known as Elkhorn sea moss. It is a main source of kappa carrageenan (Hayashi et al., 2010). Recent reviews highlight that marine algae possess anticoagulant, antihyperlipidemic, antihyperglycemic, anti-inflammatory, antioxidative, antitumor, antiviral, immunomodulating and hepatoprotective activities (Wang et al., 2014; Wells et al., 2017).

**Spheciospongia vagabunda** is a marine sponge belonging to the family Chionidae. Marine sponges are sessile organisms that lack of circulatory, nervous and digestive systems. Marine soft corals such as *Sarcophyton auritum* are also sessile organisms that have less rigid calcium carbonate structure and with a soft and leathery appearance (Cooper et al., 2014). Extracts of *S. vagabunda* and *S. auritum* have shown strong activity against liver and breast cancer cell lines (Eltamany et al., 2014). Soft corals of the genus *Lobophytum* produce many cbembranoid diterpenes, such as crassumolide E, an inhibitor of acetylcholinesterase, and lobohedrolide, (7Z)-lobohedrolide and 17-dimethylaminolobohedrolide which have anti-HIV activity (Rashid et al., 2000; Bonnard et al., 2010).

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Malaysia has one of the largest continental shelf areas in the tropical world, which is a region rich in biodiversity (Mazlan et al., 2005). As such, the potential of marine organisms in the coastal areas of Malaysia should be explored, in order to have a better understanding on their antimicrobial and antiviral properties. In this study, three species of seaweeds, i.e., Caulerpa racemosa, Caulerpa sertularioides and Kappaphycus alvarezii, two species of soft corals, *Lobophytum microlobulatum* and *Sarcophyton auritum*, and a marine sponge *Spheciospongia vagabunda* were evaluated for antibacterial activity against Gram-positive and Gram-negative bacteria, antifungal activity against yeasts and filamentous fungi, antiviral activity against the Chikungunya virus and cytotoxicity on the African monkey kidney epithelial (Vero) cells.

**II. MATERIALS AND METHODS**

1. **Collection and Identification of Marine Organisms**

Caulerpa racemosa, Caulerpa sertularioides, *Lobophytum microlobulatum*, *Sarcophyton auritum* and *Spheciospongia vagabunda* were collected from the coastal area of Port Dickson, Negeri Sembilan, West Malaysia. *Kappaphycus alvarezii* was collected from the coastal area of Semporna, Sabah, East Malaysia. The fresh materials were transported in sea water to the laboratory located at Kampar, Perak, West Malaysia for processing except for *K. alvarezii* which was oven-dried and delivered to the laboratory via courier service. Species identification was performed by Dr. Kong Soo Khoo (one of the researchers) for *C. racemosa* (Forssskål) J.Agardh and *C. sertularioides* (S.G.Gmelin) M.Howe; Dr. Suhaimi Md Yasir (Seaweed Research Unit, Universiti Malaysia Sabah) for *K. alvarezii* (Doty) Doty ex P.C.Silva; Dr. van Oweghenleen (Naturalis Biodiversity Center, RA Leiden, The Netherland) for *L. microlobulatum* (Tixier-Durivault, 1970); Dr. Ping-Jyun Sung (National Museum of Marine Biology and Aquarium, Pingtung, Taiwan) for *S. auritum* (Verseveldt & Benayahu, 1978); and Dr. de Voogd Nicole (Naturalis Biodiversity Center, RA Leiden, The Netherland) for *S. vagabunda* (Ridley, 1884).

2. **Preparation of Marine Organism Extracts**

The marine organisms were cleaned thoroughly before freeze drying, except for *K. alvarezii* which was oven-dried at 40°C. The dry samples were blended into powder form and subjected to sequential solvent extraction using hexane, chloroform, ethyl acetate, ethanol, methanol and distilled water in order to extract bioactive compounds. The maceration was carried out at room temperature for three cycles (one day per cycle). The filtrate for each solvent was evaporated using a rotary evaporator while the water extract was lyophilized. All the dried extracts were kept at -20°C prior to bioassay.

3. **Bioassays**

1) **Antibacterial Assay**

Two species of Gram-positive bacteria (*Bacillus cereus* ATCC®11778™ and *Staphylococcus aureus* ATCC®6538™) and four species of Gram-negative bacteria (*Acinetobacter baumannii* ATCC®19606™, *Escherichia coli* ATCC®35218™, *Klebsiella pneumoniae* ATCC®13883™ and *Pseudomonas aeruginosa* ATCC®27853™) were evaluated in the study. Prior to inoculum preparation, all the bacteria were sub-cultured on Mueller-Hinton agar (MHA) at 37°C for 24 h. The bacterial inocula were
prepared according to the guidelines of the Clinical Laboratory Standards Institute (CLSI, 2013).

The colorimetric broth microdilution method, with some modifications, of Eloff (1998) was used. A stock solution of each extract at 10 mg/mL was prepared in a methanol-water mixture (2:1, v/v). The stock solution was serially diluted two-fold with Mueller-Hinton broth (MHB) in 96-well microplates to obtain eight concentration values ranging from 0.02 to 2.50 mg/mL. Chloramphenicol/tetracycline with a final concentration range of 1 to 128 µg/mL was used as the positive control. Prepared bacterial inoculum (50 µL) was then added to the wells, making the total volume of each well 100 µL. Sterility control (medium only), growth control (bacterium only) and negative control (extract only) were also included in each 96-well microplate. The microplates were incubated at 37°C for 24 h. Twenty µL of iodonitrotetrazolium chloride (INT) at 0.4 mg/mL was added into each well after incubation. The color change of the indicator was observed and minimum inhibitory concentration (MIC) was recorded. Twenty µL of the wells which showed inhibitory effect (no color change) was then inoculated onto SDA/PDA and incubated accordingly. Formation of fungal colony on the agar surface was observed and minimum fungicidal concentration (MFC) was determined. The assay was performed in three independent experiments.

3) Cytotoxicity Assay

African monkey kidney epithelial (Vero) cell line (ATCC®CCL-81™) was used for cytotoxicity testing of the extracts (Chan et al., 2015). The cell line was cultured in Dulbecco’s modified Eagle medium (DMEM) supplemented with 5% of fetal bovine serum (FBS), 1% of penicillin-streptomycin solution and 3.7 g/L of sodium bicarbonate, and grown at 37°C in a humidified atmosphere with 5% of carbon dioxide.

The cytotoxicity assay was carried out to determine non-cytotoxic concentrations of the extracts to be used in the antiviral assay (Section 3(4)). Vero cells were seeded in each well of 96-well microplates (4 × 10^4 cells/well) and incubated for 24 h at 37°C in a humidified incubator with 5% CO₂. Extracts (100 µL each) of eight different concentrations ranging from 5 to 640 µg/mL, obtained by two-fold serial dilution in DMEM supplemented with 1% of FBS, were added after 24 h of cell seeding. The control wells contained Vero cells without any extract treatment. The microplates were incubated at 37°C in a humidified incubator with 5% CO₂ for 72 h. The cell viability was determined by the Neutral Red uptake assay (Repetto et al., 2008) after incubation. The assay was conducted in three independent experiments with duplicates for each experiment.

4) Antiviral Assay

The antiviral assay was carried out in the Virology & Immunology Laboratory with restricted access and equipped with Biosafety Level 2 facilities. The Chikungunya virus used belonged to the Bagan Panchor strain (accession number EU703761; Asian genotype) and provided by Professor Shamala Devi of Faculty of Medicine, University of Malaya, Malaysia. The virus was propagated in the Vero cells and harvested after cytopathic effect had developed and stored at -80°C prior to bioassay.

Non-cytotoxic concentrations of the extracts with cell viability ≥ 90%, as determined from the cytotoxicity assay, were deployed in the cytopathic inhibition effect assay (Chan et al., 2016). Vero cells (4 × 10^4 cells/well) were seeded in 96-well microplates and incubated at 37°C and 5% CO₂ for 24 h. Extracts of six different concentrations, obtained by two-fold serial dilution in DMEM supplemented with 1% of FBS, were then added together with Chikungunya virus at a multiplicity of infection of one. The virus titre was determined according to the method of Reed and Muench (1938). Medium (DMEM only), virus (cells with virus only) and cell (cells with medium only) controls were incorporated in each microplate. Chloroquine with a final concentration range of 0.39 to 12.4 µM was used as the positive control. The microplates were then incubated at 37°C and 5% CO₂ for 72 h. Determination of cell viability was carried out by the Neutral Red uptake assay (Repetto...
et al., 2008). The assay was performed in three independent experiments with duplicates for each experiment. Upon completion of the experiments, all the microplates were autoclaved prior to disposal.

### Table 1. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of marine organism extracts against medically-important bacteria.

<table>
<thead>
<tr>
<th>Extracts</th>
<th>Gram-positive bacteria</th>
<th>Gram-negative bacteria</th>
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<tr>
<td></td>
<td>Bacillus cereus</td>
<td>Staphylococcus aureus</td>
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<td></td>
<td>MIC (mg/mL)</td>
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<td>Chl</td>
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<td>Hex</td>
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<td>Chl</td>
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<td>EtOH</td>
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<td>Water</td>
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<td>Antibio</td>
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The MIC and MBC values are expressed as mean of three consistent independent replicates in mg/mL. Abbreviations: Hex, hexane extract; Chl, chloroform extract; EA, ethyl acetate extract; EtOH, ethanol extract; MeOH, methanol extract; Antibio, chloramphenicol (positive control) was used for B. cereus, S. aureus and K. pneumoniae whereas tetracycline (positive control) was used for A. baumannii, E. coli and P. aeruginosa. NA denotes no activity while "-" denotes not performed.

### 4. Data Analysis

The percentages of cell viability at different extract concentrations were analyzed by one-way analysis of variance (ANOVA) using IBM® SPSS® Statistics (Version 20) software. The two-tailed significance level was set at $P \leq 0.05$. Post hoc test, either with Tukey’s (equal variance assumed) or Dunnett’s (equal variance not assumed) test was further conducted to determine which concentration of extract was effective.

### III. RESULTS
### 1. Antibacterial Activity

The bacteriostatic and bactericidal activities of an extract are denoted by the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC), respectively. As shown in Table 1, not all the extracts possess both activities. By considering one extract against one bacterial species as one bioassay, 37.0% (80/216) of the bioassays showed bacteriostatic activity while only 17.1% (37/216) showed bactericidal activity. At the concentrations used for this study, none of the extracts were able to kill *A. baumannii* and *E. coli*.

### Table 2. Minimum inhibitory concentration (MIC) and minimum fungicidal concentration (MFC) of marine organism extracts against medically-important fungi.

<table>
<thead>
<tr>
<th>Extracts</th>
<th>Candida albicans</th>
<th>Candida parapsilosis</th>
<th>Candida krusei</th>
<th>Cryptococcus neoformans</th>
<th>Aspergillus fumigatus</th>
<th>Trichophyton interdigitale</th>
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<td>MIC&lt;sup&gt;a&lt;/sup&gt;</td>
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**Note:** The MIC and MFC values are expressed as mean of three consistent independent replicates in mg/mL. Abbreviations: Hex, hexane extract; Chl, chloroform extract; EA, ethyl acetate extract; EtOH, ethanol extract; MeOH, methanol extract; Antibio, Amphotericin B (positive control); NA denotes no activity while ‘-’ denotes not performed.

Extracts from the soft corals were found to be more active towards the bacteria compared to the seaweeds and marine sponge. The chloroform and ethyl acetate extracts of *L. microlobulatum*, and the hexane, chloroform and ethyl acetate extracts of *S. auritum* exhibited broad spectrum antibacterial activity as they inhibited the growth of five out of six bacteria tested with their MIC ranges of 0.16-1.25, 0.63-1.25, 0.16-2.50, 0.31-1.25 and 0.63-2.50 mg/mL, respectively (Table 1). The chloro-
form extract of *L. microlobulatum* was the only extract that showed bactericidal effect against *S. aureus*. The three extracts derived from *S. auritum* were the only extracts that killed *P. aeruginosa*. In addition, the strongest bacteriostatic and bactericidal activities, as evident in the lowest MIC and MBC values, were observed in the hexane extract of *L. microlobulatum* against *S. aureus* (0.04 mg/mL) and *B. cereus* (0.08 mg/mL), respectively.

### 2. Antifungal Activity

Minimum inhibitory concentration (MIC) and minimum fungicidal concentration (MFC) are used to illustrate the potency of an extract with regard to its fungistatic and fungicidal activities, respectively. Fungistatic activity was observed in 43.5% (94/216) of the bioassays whereas only 25.0% (54/216) of the bioassays showed fungicidal activity (Table 2). *Aspergillus fumigatus* was the most resistant fungus tested, as none of the extracts were effective against it. Extracts from the two soft corals also appeared to be more active towards the fungi than the seaweeds and sponge. The lowest MIC and MFC values (0.04 mg/mL) were shown by the hexane extract of *S. auritum* against the dimorphic yeast, *C. neoformans*. All extracts of *S. auritum* showed antifungal activity against all the yeasts with MIC ranges of 0.16–1.25 mg/mL against *C. albicans*, 0.08–1.25 mg/mL against *C. parapsilosis*, 0.16–2.50 mg/mL against *C. krusei* and 0.04–2.50 mg/mL against *C. neoformans*. All extracts of *L. microlobulatum* were active against *C. neoformans* with a MIC range of 0.04–2.50 mg/mL. In contrast, neither fungistatic nor fungicidal activity was exhibited by the water extract of *C. sertularioides*, ethanol, methanol and water extracts of *C. racemosa* and *S. vagabunda*, and all the extracts (except the hexane extract) of *K. alvarezii*. The hexane extract of *K. alvarezii* showed antifungal activity against *C. neoformans* and *T. interdigitale* with a MIC value of 2.50 mg/mL.

### 3. Cytotoxicity and Antiviral Activity

The antiviral activity of the extracts was evaluated based on the inhibition of cytopathic effect caused by the Chikungunya virus on the Vero cells. As the Neutral Red uptake assay measures the viability of cells and does not differentiate the virus-induced cytopathic effect and the cytotoxicity caused by extracts, it is essential to use non-toxic concentrations of the extracts for the antiviral assay. The cytopathic inhibition effect can be classified on an arbitrary basis as strong when cell viability is 70% or higher, moderate when cell viability is between 31% to 69%, and weak when cell viability is 30% or lower. Three of the 36 extracts (8.3%) showed significant strong inhibition effect, while 22.2% of the extracts demonstrated moderate inhibition effect, i.e., the chloroform, ethyl acetate, ethanol and methanol extracts of *C. racemosa*, the chloroform extract of *C. sertularioides*, the chloroform and ethyl acetate extracts of *L. microlobulatum* and the ethanol extract of *S. auritum* (Fig. 1). Other extracts exhibited only weak inhibition effect. Three extracts showed strong antiviral activity. These were derived from the two soft corals, hexane and ethanol extracts of *L. microlobulatum* with cell viabilities (mean ± s.d.) of 73.3% ± 2.6% and 79.2% ± 0.9%, respectively, and ethyl acetate extract.
of S. auritum with cell viability of 81.4% ± 1.6% (Fig. 1). The corresponding 50% effective concentrations (EC50), as determined from the plot of percentage of cell viability against extract concentration, were 14.3 ± 0.2, 124.3 ± 1.9 and 176.6 ± 9.7 µg/mL, respectively. Results from the cytotoxicity assay indicated that the 50% cytotoxic concentrations (CC50) for the hexane extract of L. microlobulatum was 34.5 ± 0.2 µg/mL while the CC50 for the other two extracts exceeded the highest concentration used in the study, 640.0 µg/mL. All extracts from the seaweed K. alvarezii and the marine sponge S. vagabunda showed weak cytopathic inhibition effect with the mean cell viability being less than 20%.

IV. DISCUSSION

This study reports for the first time the antiviral activity of seaweeds (C. racemosa and C. serrulatoideas) and soft corals (L. microlobulatum and S. auritum) against the Chikungunya virus, and the antibacterial and antifungal activities of L. microlobulatum against human pathogens.

Saraiva et al. (2011) proposed a classification for plant extracts based on the potency of antimicrobial activity according to MIC values, whereby an extract can be regarded as highly active (MIC ≤ 0.1 mg/mL), active (0.1 mg/mL < MIC ≤ 0.5 mg/mL), moderately active (0.5 mg/mL < MIC ≤ 1 mg/mL), weakly active (1 mg/mL < MIC ≤ 2 mg/mL) and inactive (MIC > 2 mg/mL). By using the proposed classification, only the chloroform extract of C. serrulatoideas and hexane extract of L. microlobulatum were classified as extracts having highly active antibacterial activity. The extracts that were classified as having highly effective against fungi were the chloroform, ethyl acetate, and ethanol extracts of L. microlobulatum, and hexane, chloroform, ethyl acetate and ethanol extracts of S. auritum. It is evident that soft coral extracts show promising antibacterial and antifungal properties compared to the seaweeds and sponge.

Despite the paucity of reports on the biological activities of extracts from L. microlobulatum and S. auritum (Eltamany et al., 2014), several chemical constituents have been isolated from these soft corals. Among these compounds, lobane diterpenoids, norsesquiterpene and steroid glycosides have been reported from L. microlobulatum (Anjaneyulu and Kameswara Rao, 1996; Anjaneyulu and Raju, 1996). Four diterpene cembranoids and a ceramide have been isolated from the methanol/dichloromethane (1:1) extract of S. auritum (Eltahawy et al., 2014; Eltahawy et al., 2015). Lobane diterpenoids and cembranoids are unique secondary metabolites found abundantly in many genera of soft corals (Blunt et al., 2013). They exhibit a wide spectrum of biological activities, including antimicrobial activities (Bonnard et al., 2010; Hussein and Mohamed, 2015), and thus may account for the effects observed in this study. The results also suggest that the activity may be attributed to chemical constituents other than lobanes and cembranoids, due to the diversity of solvent polarity used in the extraction. The type of chemicals likely extracted from the marine organisms is determined by the nature of the solvent used (Grosso et al., 2015).

In this study, three extracts which exhibited strong antiviral activity were obtained from two soft corals, i.e., the hexane and ethanol extracts of L. microlobulatum and the ethyl acetate extract of S. auritum. Although the potency (based on EC50 values) for the latter two extracts was lower than the hexane extract, they also possessed lower cytotoxicity (based on CC50 values) compared to the hexane extract. Secondary metabolites of soft corals have attracted much attention from researchers due to their remarkable pharmacological activities. Two norcembranoids, 5-epi-sinuleptolide and sinuleptolide, isolated from a soft coral of the genus Simularia exert moderate inhibition on the Chikungunya virus replicon (Lillsunde et al., 2014). Further studies are needed to isolate and characterize the active compounds from the two soft corals, and to investigate the mechanisms of their action.

The antifungal results of S. vagabunda in this study are corroborated qualitatively by the study of Eltamany et al. (2014) which used percentage of growth inhibition instead of MIC as the end-point measurement. The extract which was obtained using methanol-dichloromethane (1:1) showed 19% and 6% of inhibition against C. krusei and C. neoformans, respectively, which can be considered as non-effective as an antifungal agent. The extract also did not show any inhibitory activity against C. albicans and A. fumigatus (Eltamany et al., 2014). None of the 36 marine organism extracts used (Table 2) exhibited inhibitory activity against A. fumigatus, the causative agent of invasive aspergillosis with a mortality rate of 50%-95% (Kwon-Chung and Sugui, 2013). Such resistance has also been noted in studies using plant extracts (Musciotti et al., 2005; Webster et al., 2008). The resistance might be due to the defence mechanisms of filamentous fungi, such as production of melanin pigment. Melanin is an essential component that maintains the integrity of the conidial walls and mycelia. It protects fungi against damage from ultraviolet irradiation, high temperature, chemical and biochemical stresses (Allam and Abd El-Zaher, 2012). The capacity of the fungus to grow and adapt to stress conditions, its mechanisms for circumventing the effects of immune systems and its ability to cause damage to host cell make A. fumigatus a ubiquitous pathogen (Abad et al., 2010).

V. CONCLUSION

Extracts from the two soft corals, Lobophytum microlobulatum and Sarcophyton auritum possessed stronger antibacterial, antifungal and anti-Chikungunya virus activities compared to the seaweeds and the sponge used in the study. They could serve as potential sources of new anti-infective compounds.

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