Exploration of bioactive compounds and antibacterial activity of marine blue-green microalgae (*Oscillatoria* sp.) isolated from coastal region of west Malaysia

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Abstract

Cyanobacteria is blue–green microalgae that produces a variety of secondary metabolites such as antibiotic, cytotoxic and antimicrobial agents. Present investigation proposed to study an extraction of active agents from Cyanobacteria and its antibacterial activity. The Cyanobacteria were collected, and isolated colony was found to be *Oscillatoria* sp. and it was grown in BG 11 medium for mass cultivation. Then the centrifuged biomass was weighed and used for extraction of bioactive compounds. GCMS analysis from *Oscillatoria* sp. determines fatty acid, triazine derivatives, pyridine derivatives, acridine derivatives. The result revealed that *Oscillatoria* sp. can play crucial roles as antibacterial agents because the methanolic extract are inhibiting the growth of *S. aureus* for gram-positive whereas *P. aeruginosa* for gram-negative bacteria. The crude extract was effective against *S. aureus* with the highest concentration, 100 mg/ml where area of the zone was 14.10 ± 0.8 mm. The crude extract actively inhibited *P. aeruginosa* at 75 and 100 mg/ml with highest area of inhibition were in the average of 13.23 ± 0.1 and 15.43 ± 0.2 mm. Finally, the antimicrobial activity was carried out by disc diffusion method with different concentrations of active compounds shows the MIC value in gram-positive bacteria was 30 µg/ml whereas for gram-negative bacteria was 25 µg/ml. Cyanobacteria extract should be preferred and considered to be new natural drug or antibiotics in pharmaceutical and health care industry. This coastal region should be studied further by researchers to study more information and benefits in the future.

Keywords *Oscillatoria* sp. · GCMS · MIC · Pathogen

1 Introduction

Microalgae are the photosynthetic organisms found in littoral habitats and throughout the ocean waters known as phytoplankton. Phytoplankton include such as diatoms (Bacillariophyta), Dinoflagellates (Dinophyta), green, yellow brown flagellates, and cyanobacteria (blue–green algae) [1–3]. Cyanobacteria is an incredible old group of prokaryotic organisms that produces a variety of secondary metabolites such as antibiotic, cytotoxic,

immunosuppressive and enzyme inhibiting agents [4–6]. Bioactive compounds are defined as the secondary metabolites that can be isolated and purified from the microalgae, plant, microbes and other organisms that has ability in biological activities [7]. The production of bioactive compounds from microalgae can be beneficial in pharmaceutical industry [8, 9]. Algae also contain several bioactive compounds that are beneficial as antimicrobial agent like terpenoids, steroid, phenolic compounds, alkenes and phlorotannins [10, 11].

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Microalgae contain variety compounds that can react to inhibit the microbes like bacteria and function as anti-bacterial agent. This antibacterial agent can be found in variety of natural sources like in animal, plant and microalgae. Marine algae contribute to the biomass production in marine environment and they can also produce a variety of chemically active metabolite compounds to protect themselves against other microorganisms in their surrounding environment [10, 12]. The marine microalgae are one of the main focuses to new function as the anti-microbial agents.

In this research, the marine microalgae were cultured in BG 11 medium. The antibiotics like *Streptomycin*, *Penicillin* and *Vancomycin* were supplemented in culture media to avoid or reduce the growth of the undesired microbes that will disturb the growth of the marine microalgae [6, 13, 14]. The aim of the study is to focus on the isolation of extract form of *Oscillatoria* sp. that possesses various biological active compounds which may play crucial roles as antibacterial agents against various type of gram-positive (*S. aureus* and *B. subtilis*) and gram-negative (*P. aeruginosa* and *E. coli*) pathogenic bacteria. Moreover, the statistical data was determined by applying one-way ANOVA using Statistical Program for Social Sciences (SPSS) 16.0 software.

2 Materials and methods

2.1 Sample collection followed by identification

The sample collection was done at the Kuantan (3.812601–103.372003.3°48'45.36" N, 103°22'19.21" E) costal region of east coast of west Malaysia. The sampling was done in the month of august in 2019. The marine water samples were collected with microalgae by plankton net and brought to

laboratory. Serial dilution was carried out to get isolated colonies. The pH value of the medium was adjusted to pH 7.5 and the temperature was maintained at room temperature $25\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$ [15]. The sample was observed and identified by using fluorescent microscope. The morphological observations and referred to Algae manual it confirmed that algae belong to blue–green algae group as shown in Fig. 1. The cell diameter is $3.6\text{--}4.8\text{ }\mu\text{m}$ and the cell length is $1.2\text{--}1.8\text{ }\mu\text{m}$. Algae cell trichome aggregation was solitary with the granulated of cell [16]. According to algae manual the sample was confirmed that belong to *Oscillatoria* sp.

2.2 Microalgae mass cultivation

The single colony of cyanobacteria was cultured into 10 ml BG 11 medium. After that, it was transferred into 250 ml medium and adjusted to 500 ml for mass culture. Cyanobacteria was cultivated in 2-L Erlenmeyer's flask contained BG-11 medium and incubated in the racks sourced with fluorescent lamps as a light source. The photo-incubation was carried out at $30\text{ }^{\circ}\text{C}$ and with illuminated at light intensities 12 h dark period measured by LI-250 Light meter [17].

2.3 Culturing of pathogenic bacteria

The sterile inoculation loop was dipped in the bacterial suspension and the loopful of suspension streaked over the surface of the nutrient agar to obtain uniform growth. A final sweep was made around the rim of agar. The culture was grown at $37\text{ }^{\circ}\text{C}$ for 18 h [15]. The both gram positive and gram-negative pathogenic bacteria were used for the study. The gram-negative bacteria used were *S. aureus* (CP019117) and *B. subtilis* (AL009126); and gram-positive

Fig. 1 Species of microalgae cyanobacteria *Oscillatoria* sp. **a** fluorescent microscopic images and **b** scanning electron microscope photograph

bacteria used for the antimicrobial study were *E. coli* (U00096) and *P. aeruginosa* (PAO1).

2.4 Preparation of crude extract of microalgae

Algal mass culturing using BG II medium were separated from centrifugation at 5000 rpm for 15 min and their pellets were dried at 60 °C for 24 h. The dry algal mass was dissolved in methanol (ratio 1:15 g/ml) and extracted throughout 24 h [18]. The supernatant was collected after being centrifuged at 10,000 rpm for 10 min. The solvent extracts were concentrated under reduced pressure at 40 °C. The dried extracts were resuspended in 3 ml of solvent with different concentration (50, 75 and 100 mg/ml) and preserved at 4 °C till further use in antimicrobial assays [6, 19].

2.5 Antibacterial activity

The antimicrobial activity was measured and determined using a well diffusion test [15]. The antibiotic 50 µl of *Streptomycin* was used in this test as the control. The disks (6 mm) containing 30 µg/ml the extract form of the *Oscillatoria* sp. were placed on the surface of agar. The flame sterile forceps were used to dispense the antibiotic and extract disk one at a time. The distribution of the disks should be apart from each other and not close to the edge of plate. The bacteria inoculum was swabbed and inoculated on the media plates. The inoculated plates were then incubated for 24 h at 37 °C and zone of inhibition was measured in millimeter (mm) [20].

2.6 Minimum inhibitory concentration (MIC)

The 96 well microtiter plates were swept with ethanol before used. After that, the first row of well was pipetted with 50 µl methanol as positive control, second row of well was pipetted with 50 µl of nutrient broth and 50 µl bacteria culture as negative control. Third row until seventh rows were filled with different concentration of sample in the range of 10, 15, 20, 25 and 30 µg/ml mixed with 50 µl bacteria culture each. After leaving for 24 h, the sample was analyzed with micro plate reader (TECAN, INFINITE M200PRO). The minimum inhibition concentration value was determined as the lowest concentration of the extract in the broth medium that inhibit the visible growth of the test microorganism [21].

2.7 Gas-chromatography with mass spectrometer analysis

The bioactive compounds present in the extracts of the *Oscillatoria* sp. which played important roles in the

antimicrobial activity was measured and determined using the Gas Chromatography-Mass Spectroscopy [22]. The chromatographic column used HP-5 column (30 m×0.25 mm ID×0.25 µm film thickness), Agilent Technologies 7890A (GC system) model. The Chromatographic conditions were follows: The one micro liter of algae extract was injected into the GC-MS with Helium carrier gas and was maintained at a flow rate of 1.50 ml/min; injector and column oven temperature of 280× and 80 °C; injection mode 'split' and split ratio 20:1. Oven temperature was held isothermal at 80 °C for 1 min, then increased to 300 °C at a rate of 4 °C /min and held isothermal for 5 min. MS, (Agilent Technologies 5975C model) conditions were followed: ion source temperature of 200 °C; interface temperature of 300 °C; mass range of 40–1000 mass units [17, 23]. The individual peaks formed were identified by comparing their retention time, as well as their mass spectra with the Fienn. Library and NIST 11 (National Institute of Standard and Technology, Gaithersburg, United States) library.

2.8 Statistical analysis

The result obtained from the experiment was statistically analyzed by applying one-way ANOVA with Turkey's Test by using SPSS 16.0 (Statistical Program Social Sciences) software.

3 Results and discussion

3.1 Antibacterial activity

Data and results for the zone of inhibition of crude extract of cyanobacteria against 4 different types of bacteria were illustrated in Table 1. The sample of crude extract of *Oscillatoria* sp. was tested with three different concentrations which were 50, 75 and 100 mg/ml. The streptomycin was used as the control for this inhibition. Each reading was taken as three replicates. The two different gram-positive bacteria (*S. aureus* and *B. subtilis*) and 2 type of gram-negative bacteria (*E. coli* and *P. aeruginosa*) had shown slightly different area of inhibition when reacted with different concentration of extract from *Oscillatoria* sp. The antibacterial activity of microalgae was usually assayed using various organic solvent for effective extraction of biological active compounds. The commonly used organic solvent were methanol, acetone, ether and chloroform-methanol [24]. Most of all indicated that the methanol extraction yielded more compared to hexane and ethyl acetate.[7, 25, 26]. Ethyl acetate and methanol were determined as the most effective organic solvent for the isolation of antibacterial compounds [27, 28].

Table 1 Zone of inhibition of *Oscillatoria* sp. against *S. aureus*, *B. subtilis*, *E. coli*, *P. aeruginosa*

Microbe	Zone of inhibition (mm)			
	Streptomycin control (mm)	Concentration microalgae extract		
		50 mg/ml	75 mg/ml	100 mg/ml
<i>Staphylococcus aureus</i>	7.13±0.9	4.23±0.2	10.77±0.7	14.10±0.8
<i>Bacillus subtilis</i>	9.0±0.3	3.17±0.4	8.40±0.5	9.10±0.7
<i>Escherichia coli</i>	11.0±0.2	3.03±0.8	9.26±0.6	10.17±0.4
<i>Pseudomonas aeruginosa</i>	9.0±0.5	7.50±0.1	13.23±0.1	15.43±0.2

3.1.1 Zone of inhibition of *Staphylococcus aureus*

Table 1 indicated the result for zone of inhibition when crude extract of *Oscillatoria* sp. reacted with gram-positive bacteria *S. aureus*. The crude extract was effective against *S. aureus* with the highest concentration, 100 mg/ml where area of the zone was 14.10 ± 0.8 mm. However, for concentration 50 mg/ml, there was still inhibition, but it was slightly lower, only in the average of 4.23 ± 0.2 mm. The methanolic extract of microalgae show antibacterial activity inhibited in the *S. aureus* with maximum zone of inhibition of 12 mm at the concentration of 0.2 mg/ml [29]. The zone of inhibition produced in reaction of methanol extract of *Oscillatoria* sp. with 2 mg/disk in *S. aureus* was 27 mm [14]. The most sensitive gram-positive bacteria to the methanolic extract of *Oscillatoria* sp. was *S. aureus* with the zone of inhibition of 13.8 ± 0.8 mm [28].

The area for zone of inhibition increases as the concentration of cyanobacteria extract increases. The extract of

Oscillatoria sp. was most effectively in the inhibited growth of *S. aureus* with high concentration of 100 mg/ml (Fig. 2).

3.1.2 Zone of inhibition of *Bacillus subtilis*

The zone of inhibition for the reaction of crude extract of *Oscillatoria* sp. against *B. subtilis* was indicated as in Table 1. The crude extract was actively inhibited *B. subtilis* at 75 and 100 mg/ml with the highest area of inhibition, 9.10 ± 0.7 mm. Other concentration of 50 mg/ml zone of inhibition was only in average 3.17 ± 0.4 mm. The maximum zone of inhibition of methanol extract of marine microalgae with 2 mg/disk in against *B. subtilis* was 22 mm [14]. The previous study by [27] proved similar result for the zone of inhibition of microalgae against *B. subtilis*.

In Fig. 2, shows the area for zone of inhibition of crude extract of *Oscillatoria* sp. against *B. subtilis* which was active with the concentration 100 mg/ml which almost close to the zone of inhibition produced by Streptomycin

Fig. 2 Zone inhibition for **a** *Staphylococcus aureus* **b** *Escherichia coli* **c** *P. aeruginosa* **d** *B. subtilis*

(control). Besides 100 mg/ml, the concentration of 75 mg/ml also produced large zone of inhibition whereas the 50 mg/ml concentration showed the lowest activity against *B. subtilis*.

3.1.3 Zone of inhibition of *Escherichia coli*

Based on Table 1, it states that the crude extract of *Oscillatoria* sp. was highly responsible in inhibiting *E. coli* at the concentration of 100 mg/ml with area of inhibition was 10.17 ± 0.4 mm whereas for concentration 50 mg/ml, the area of inhibition was quite low which was only 3.03 ± 0.8 mm. The zone of inhibition from this experiment was similar with the previous study by Salem et al. [28] who reported the zone of inhibition of *E. coli* when treated with methanolic extract of microalgae from area of inhibition in the range of 8 ± 1 – 13.3 ± 1.2 mm. However, in previous study of Anandhan et al. [27] it shows that the zone of inhibition of methanolic extract of microalgae inhibited *E. coli* was 12 mm. The zone of inhibition reported by Rebecca et al. [30] was 20 mm for 100 mg/ml and 15 mm for 50 mg/ml.

In Fig. 2, shows the increasing zone of inhibition against *E. coli* as the concentration of *Oscillatoria* sp. extract increased. The 75 and 100 mg/ml slightly produced the same area for zone of inhibition and effectively inhibited growth of *E. coli* whereas the 50 mg/ml concentration proved the lowest antibacterial activity against *E. coli*.

3.1.4 Zone of inhibition of *Pseudomonas aeruginosa*

The zone of inhibition for reaction of crude extract of *Oscillatoria* sp. in against *P. aeruginosa* was indicated in Table 1. The crude extract actively inhibited *P. aeruginosa* at 75 mg/ml and 100 mg/ml with highest area of inhibition were in the average of 13.23 ± 0.1 mm and 15.43 ± 0.2 mm. For the concentration of 50 mg/ml, the area of zone of inhibition was slightly high in average 7.50 ± 0.1 mm. The most gram-negative bacteria that was sensitive to methanolic extract of microalgae were *P. aeruginosa* with the zone of inhibition was 15 ± 1 mm [28]. The zone of inhibition of methanol extract of microalgae inhibited in *P. aeruginosa* obtained from this experiment was similar to the experiment done by Anandhan et al. [27].

Based on Fig. 2, the graph shows that the 100 mg/ml was the most active concentration of *Oscillatoria* sp. extract that can inhibit growth of *P. aeruginosa* with larger zone of inhibition compared to Streptomycin that act as control. However, concentration at 50 mg/ml showed the lowest zone of inhibition compared to other concentration.

For concentration of 50 mg/ml, the crude extract produced more efficiency to inhibit the *P. aeruginosa* with area of zone inhibition is 7.50 ± 0.1 mm and less efficiency

in inhibiting the *B. subtilis* and *E. coli* with zone of inhibition was 3.03 ± 0.8 mm respectively as shown in Fig. 2. The crude extract with 75 mg/ml concentration was also more effective against *P. aeruginosa* and *S. aureus* with the reading of 13.23 ± 0.1 mm and 10.77 ± 0.7 mm respectively and less effective against *B. subtilis* where the area of inhibition was 8.40 ± 0.5 mm as shown in Fig. 2. The crude extract of *Oscillatoria* sp. with concentration 100 mg/ml showed high antibacterial activity against the gram-negative bacteria that was *P. aeruginosa* which area of zone of inhibition was 15.43 ± 0.2 mm as shown in Fig. 2 whereas shows less antibacterial activity against the gram-positive bacteria that was *B. subtilis* which area of inhibition was 9.10 ± 0.7 mm.

The similar study reported by Arnab Pramanik and Joydeep Mukherjee [6] have revealed the *Phormidium* sp. strongly acted as antibacterial agent against the *P. aeruginosa* with 14 mm zone of inhibition. The LPP group B showed the effectiveness in inhibiting the *S. aureus* (18 mm), *B. subtilis* (15 mm) and *E. coli* (15 mm).

The microalgae extract was mostly active in gram negative bacteria compared to gram positive [28]. Previous studies indicated that the methanol extraction yielded more compared to hexane and ethyl acetate [25, 31]. In the previous report of Rao et al. and Rajasulochana et al. [32], they revealed similar investigation as in this experiment where the active compounds in the extract of microalgae was active against *S. aureus* for gram-positive bacteria and *P. aeruginosa* for gram-negative bacteria. *Oscillatoria* sp. proved the antibacterial activity against gram-negative and gram-positive bacteria [33].

3.2 Minimum inhibitory concentration (MIC)

3.2.1 Gram-negative bacteria: *Pseudomonas aeruginosa*

The results from 96-well micro titer plate analyzed by micro titer plate reader. The positive control is the solvent namely methanol that was used to extract the bioactive compound from the crude extract of the *Oscillatoria* sp. The negative control used was the culture of selected bacteria with the nutrient broth. The different concentrations of *Oscillatoria* sp. extract were prepared from 10, 15, 20, 25 and 30 µg/ml (Fig. 3). From the data, the reading is slightly in ascending form of the inhibition level that occurred in the micro plate between reactions of *Oscillatoria* sp. extract against the bacteria culture. The pattern of inhibition level formed can be seen more clearly from the graphical data.

Figure 3 shows the level of inhibition produced from the reaction of crude extract from *Oscillatoria* sp. against the bacteria *Pseudomonas aeruginosa*. From the graph, it shows the increasing level of inhibition. The most minimal concentration that can cause this inhibition to occur

Fig. 3 Graph of level of inhibition (OD) against concentration of sample for minimal inhibitory concentration (MIC) for *P. aeruginosa*

effectively is 25 µg/ml with the optimal density is 0.9391. The *Oscillatoria* sp. extract depends on 25 µg/ml to control the growth of gram-negative bacteria *P. aeruginosa* because the active compounds which function to inhibit growth of bacteria was active in this concentration.

Besides that, the graph also shows that the inhibition level for each concentration started to increase at the concentration 5 µg/ml. So, it shows that at the concentration of 25 µg/ml the antibacterial activity of crude extract from *Oscillatoria* sp. against the gram-negative bacteria, *P. aeruginosa* is higher. The MIC result obtained from this experiment was almost the same as the previous study by [28] Salem et al. where the MIC for *P. aeruginosa* was 20 µg/ml.

3.2.2 Gram-positive bacteria: *Staphylococcus aureus*

Figure 4 indicates the data analyzed by the micro titer plate reader from the reaction of the gram-positive bacteria that is *S. aureus* with different concentration of crude extract of *Oscillatoria* sp. The different concentration prepared were 10, 15, 20, 25 and 30 µg/ml.

The solvent methanol was used as the positive control whereas the selected bacteria culture within nutrient broth was served as negative control. The reading of level of inhibition produce was ascending for each concentration. The graphical data may present clear pattern produced from that reaction.

Figure 4 above presents the increasing reading pattern for the level of inhibition produced from the reaction of crude extract of *Oscillatoria* sp. in inhibiting the gram-positive bacteria *S. aureus*. The different concentration of

crude extract from *Oscillatoria* sp. studied in this experiment were 10, 15, 20, 25 and 30 µg/ml. Hence, from the graph above the minimal concentration that may inhibit the reaction of bacteria was 30 µg/ml with the level of optimum density was 0.4764. It proved that at concentration of 30 µg/ml, the crude extract of *Oscillatoria* sp. can function as antibacterial agent efficiently against the gram-positive bacteria *S. aureus*. The minimal inhibitory concentration (MIC) for microalgae against *S. aureus* was 25 µg/ml because the di-phenolic metabolites like bromo-phenols were found to be the most effective compound as antimicrobial agents [34, 35].

The minimum inhibitory concentration (MIC) of marine algae against *S. aureus* was 0.5 mg/ml and maximum MIC was 1 mg/ml. (Selim, 2012). The MIC for *S. aureus* obtained in this experiment was almost the same as the previous study of Salem et al. (2011) which was in the range of 20–50 µg/ml.

3.3 Statistical analysis

Figure 5 shows the antibacterial activity of *Oscillatoria* sp. extract with different concentrations (50, 75 and 100 mg/ml) in 2 different types of gram-positive bacteria (*S. aureus* and *B. subtilis*) and 2 different types of gram-negative bacteria (*E. coli* and *P. aeruginosa*). Therefore, the antibiotic Streptomycin was served as control. The result showed a significantly higher antibacterial activity ($p < 0.05$) which was determined by a one-way ANOVA and the error bars were by SEM, $n = 3$.

Fig. 4 Graph of level of inhibition (OD) against concentration of sample for minimal inhibitory concentration (MIC) for against *S. aureus*

Fig. 5 Antibacterial activity of *Oscillatoria* sp. extract with different concentrations (50, 75 and 100 mg/ml)

3.4 Bioactive compounds analysis of cyanobacteria extract

The bioactive compound identified in cyanobacteria were fatty acid, amino acid derivative, indolizine derivative, ether derivative, cinchoninic acid, phenol, pyridine derivative, benzyl alcohol derivative and triazine derivative as shown in Fig. 6 and Table 2. Halogenated compounds present in microalgae and macroalgae were indicated as biological active compounds [36, 37]. The important

compounds that were indicated as antimicrobial are fatty acid, acrylic acid, terpenes, phenol and halogenated compounds [29]. Previous study by Kolanjinathan et al. [20] found the most bioactive compounds that present in microalgae extract was chemically classified as aromatics, sulphate polysaccharide, brominated, nitrogen-heterocyclic, protein, peptides, dibutanoids, nitrosulphuric-heterocyclic and sterols. The *Oscillatoria* sp. was active against pathogenic bacteria with the presence of pyridine and n-butanol [33].

Fig. 6 Total ionic chromatogram biological active compound of methanol extract of *Oscillatoria* sp. with HP-5 column (30 m×0.25 mm) with helium as carrier gas

Table 2 Total ionic chromatogram biological active compound of methanol extract of *Oscillatoria* sp. with HP-5 column (30×0.25 mm) with helium as carrier gas

Compounds name	Compound nature	Molecular formula	Retention time (min)
4,6-bis(diethylamino)-1,3,5-triazine	Triazine derivative	C ₃ H ₃ N ₃	26.544
5-Nitro-3-cyano-2(1H)-pyridone	Pyridone derivative	C ₅ H ₄ NH(O)	32.401
Triphenylphosphine oxide	Phenyl derivative	C ₁₈ H ₁₅ OP	40.946
1,4 benzenedicarboxamide	Phthalic acid derivative	C ₆ H ₄ (COOH) ₂	47.525
Benzenemethanol	Benzyl alcohol derivative	C ₇ H ₈ O	49.460
trimethylsilyl ether	Ether derivative	C ₅ H ₁₂ OSi	49.927
2-Methyl-7-phenyl indole	Indole derivative	C ₈ H ₇ N	57.984
Acetic acid	Fatty acid	C ₂ H ₄ O ₂	58.026
Silicic acid	Carboxylic acid	Si (OH) ₄	58.292
9-Oxo-9,10-dihydro acridine-4-yl	Acridine derivative	C ₁₃ H ₉ N	59.823
2-(p-Fluorophenyl)-6-methylcinchoninic acid	Cinchoninic acid	C ₁₀ H ₇ NO ₂	60.003
5-Methyl-2-phenyl indolizine	Indolizine derivative	C ₁₅ H ₁₃ N	60.832

The 1, 3, 5-triazine was the bioactive compound identified at retention time of 26.544 min. Triazine was included in chemical species of heterocyclic aromatic ring compound where the three nitrogens replaced the carbon-hydrogen units on the benzene ring. 1, 3, 5-triazine represent incredible class of compound with wide spectrum of

biological activities such as antimicrobial, anticancer, antiviral and fungicidal [38, 39]. The earlier report by Sarmah and Patel [40] also had reported that the triazine derivatives have potential against gram-positive bacteria such as *B. subtilis* and *S. aureus* and gram-negative bacteria such as *P. aeruginosa* and *E. coli*.

4 Conclusion

The result obtained from this experiment clearly revealed that *Oscillatoria* sp. can play crucial roles as antibacterial agents because the extract from *Oscillatoria* sp. showed impressive result for zone of inhibition area against gram-positive bacteria (*S. aureus* and *B. subtilis*) and gram-negative bacteria (*E. coli* and *P. aeruginosa*). However, the methanolic extract of *Oscillatoria* sp. from this experiment proved to be more actively in inhibiting the growth of *S. aureus* for gram-positive whereas *P. aeruginosa* for gram-negative bacteria. The antibacterial activity from the same extract of *Oscillatoria* sp. are for several bioactive compounds which has also been reported in many studies such as fatty acid, triazine derivatives, pyridine derivatives, acridine derivatives and etc. which had proven to be very effective in playing their role as antibacterial. The minimal inhibitory concentration (MIC) value in gram-positive bacteria was 30 µg/ml whereas for gram-negative bacteria was 25 µg/ml. *Oscillatoria* sp. extract depends on those concentrations to control the growth of bacteria because of the bioactive compounds which function as antibacterial agent active and high amount in those concentrations. Based on the experiment, marine cyanobacteria (*Oscillatoria* sp.) have proven their potential as antimicrobial, anticancer, antioxidant and antitumor activity. The extract from cyanobacteria can be applied for various treatment for several diseases instead of using chemical drugs or treatments. Cyanobacteria extract should be preferred and considered to be new natural drug or antibiotics in pharmaceutical and health care industry. The authorities and responsibility should come out with new alternative in culturing microalgae in large scale for commercialization and improvement of the health care industry without side effects.

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Compliance with ethical standards

Conflict of interest There are no conflicts to declare.

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