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Full Length Research Paper

Bioactivity of natural compounds isolated from cyanobacteria and green algae against human pathogenic bacteria and yeast

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Analysis of bioactive natural compounds of algae such as cyanobacteria (*Spirulina platensis*; *Nostoc linckia*; *Phormidium autumnale*; *Tolypothrix distorta* and *Microcystis aeruginosa*) and green algae (*Chlorella vulgaris* and *Dunaliella salina*), and their activity against human pathogenic bacteria and yeast such as *Salmonella suis* ATCC 13076; *Pseudomonas aeruginosa* ATCC 27583; *Escherichia coli* ATCC 25922; *Staphyllococcus aureus* ATCC 25923; *Bacillus subtilis* ATCC 6633; *Shigella sonnei* ATCC 11060 and *Candida albicans* ATCC 10231 have been studied in Saudi Arabia. Extraction of algal metabolites was performed by using the mixture of three organic reagents, methanol:acetone:diethyl ether as 5:2:1 v/v, respectively. All metabolites of algae isolates had shown weak to strong antimicrobial activity toward one or more human pathogenic microorganisms. Almost all the algal extract showed strong activity against *S. sonnei* in agar well diffusion technique. Crude extract of cyanobacteria, *T. distorta* showed moderate to strong activity against *S. aureus, B. subtilis* and *S. sonnei*. Further, crude extract of all the algal metabolites have been analyzed using gas chromatography-mass spectrometry (GC-MS). Results indicated that the main component in the crude extracts of *P. autumnale* is 1-Hexyl-2-Nitrocyclohexane (91.7%); *C. vulgaris* is 2-Butanol, 3-methyl-, (S)- (90.8%); *S. platensis* is Nitrocyclohexane-2-Hexyl-1 (92.1%); *N. nostoc* is Octadecanal (aldehyde) (86.8%); *D. salina* is 3-Methyl-2-(2-Oxopropyl) Furan (90.%); *T. distorta* is Boronic acid, Ethyl-, Dimethyl ester (83.9%) and lastly *M. aeruginosa* is (S)-(+)-1-Cyclohexylethylamine representing 91.9%, respectively. Further, in this study, the extracts of all the algal species especially *T. distorta*, *P. automonate*, *C. vulgaris* and *D. salina* have been found potential for the production of several compounds including biomedically important organic metabolites such as ethane,1,1-diethoxy-; butanal; heptanal and octanal. Further study for the purification of the potent compound will explain their usefulness in the pharmaceutical and biotechnological industry.

Key words: Cyanobacteria, green algae, algal extract, gas chromatography-mass spectrometry (GC-MS), antimicrobial activity, human pathogenic bacteria, biomedical properties.

INTRODUCTION

Cyanobacteria or blue-green algae are a fascinating group of primitive phototrophic prokaryotic organisms whose long evolutionary history dates back to the Proterozoic era. These organisms, endowed with tremendous genome plasticity, are distributed in all possible biotypes of the world. These organisms have tremendous potential in environmental, management as soil conditioners.

Due to their occurrence in diverse habitats, these organisms are the excellent material for investigation by the ecologists, physiologists, biochemists, pharmacists and molecular biologists. Accordingly, looking for cyanobacteria with antimicrobial activity has gained importance in recent years. Biologically active substances were proved to be extracted by cyanobacteria

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(Borowitzka, 1995; Kreitlow et al., 1999; Mundt et al., 2001; Volk and Furkert, 2006). Various strains of cyanobacteria are known to produce intracellular and extracellular metabolites with diverse biological activities such as antialgal (John et al., 2003), antibacterial and antifungal (Ghasemi et al., 2003, 2007; Isnansetyo et al., 2003; Jaki et al., 1999; Kundim et al., 2003; Soltani et al., 2005) and antiviral activity (Moore et al., 1989).

Antibiotic resistance in bacteria is one of the emerging health related problem in the world nowadays. Plants and among them algae are valuable natural sources effective against infectious agents. Extensive efforts for the identification of bioactive compounds derived from natural resources have been made worldwide, in order to develop safe, nontoxic and efficient anti-microbial agents of valuable practice in pharmacology. Algae of marine and terrestrial origins have been the best choice among natural resources within aquaculture and agriculture fields. Screening bioactivity of algal crude extracts is mandatory in biomedical practice, where antibacterial (Tuney et al., 2006), antifungal (Moreau et al., 1998; Tang et al., 2002), antiviral (Serkedjieva, 2004) and even more antialgal (Hellio et al., 2002) activity have been assessed to these metabolites. Emergence concerns have been raised to establish structural and functional properties of the bioactive compounds described in algal crude extracts, up to date, over 2,400 bioactive metabolites have been isolated and identified from a diverse group of algal communities (Faulkner, 2001).

Due to the pivotal role played by these organisms, it was considered worthwhile to examine growth parameters, physiological attributes and antimicrobial activity for possible biotechnological applications. Our goal in the current study is to analyze and identify the chemical components and structure of algal crude extracts which was previously determined to have a strong antibacterial activity. All the subjected extracts were originated from algae isolates obtained from different desert soil sources in Saudi Arabia.

MATERIALS AND METHODS

Sample preparation

Isolation and cultivation of algal species

Seven algal strains were selected for screening of their antimicrobial activity belonging to cyanobacteria and green algae including *C. vulgaris*; *S. platensis*; *N. linckia*; *P. autumnale*; *D. salina; T. distorta* and *M. aeruginosa*. Algal species were isolated from different desert soils in Saudi Arabia according to standardized algae isolation procedure (Rippka et al., 1979; Vaara et al., 1979), and further identification was performed by scanning electron microscopy (SEM, JEOL, JSM, 6460), in addition to light microscopic morphology. Each isolate was sub cultured in suitable nutrition media (BG11; Rippka et al., 1979) for algae cultivation and allowed to flourish at 20-30°C under constant light for 2-4 weeks. Algal cells were collected in the exponential growth phase by filtration to be applied for extraction. All the algal strains were preserved in the Phycology Laboratory, Botany Department, Faculty of Science, King Saud University, Saudi Arabia.

Extraction of algal biomass

For the extraction of metabolites, dried algae biomass was mixed in a glass flask with methanol:acetone:diethyl ether as 5:2:1 volumes, respectively, and shaken for 3 days at about 20°C. The mixture was separated by filtration. Then, the combined solvents were evaporated to dryness and the residue re-dissolved in 2 ml distilled water to form stock solution as 50 mg/ml.

Bacterial bioassay

Microbial indicators and growth conditions

Seven microorganisms including Gram +ve, Gram -ve bacteria and yeast were used in this study. These are: *S. suis* ATCC 13076, *P. aeruginosa* ATCC 27583, *E. coli* ATCC 25922, *S. aureus* ATCC 25923, *B. subtilis* ATCC 6633, *S. sonnei* ATCC 11060 and *C. albicans* ATCC 10231. Bacterial strains and yeast were kindly provided from Microbiology Laboratory, Botany Department, Faculty of Science, King Saud University, Saudi Arabia. Bacterial bioassay was performed using agar well diffusion method with the extract of algae species. In each assay, fresh cultures were obtained by inoculating the strains on nutrient agar plates and incubated at 37°C for 18-24 h. About 2-3 colonies of bacteria were inoculated into nutrient broth and incubated overnight at 37°C. Bacterial turbidity of all the test organisms as 0.5 Mc Farland standard was obtained and then swabbed with sterile cotton swab onto Muller Hinton agar plates. The swabbed plates were allowed to dry, and then distant wells were made within agar to be loaded with the organic extracts.

Antimicrobial activity by the agar-well diffusion method

For antagonistic activity of algal extracts, agar well diffusion technique was performed. For this reason, surface of Petri dishes was punched with 5 mm cut with sterile straw and bottom of each well was sealed with two drops of sterile water agar. About 100 µl of algal extract were transferred into each well. Wells loaded with the extracting solvents were used as controls. All the plates inoculated with bacteria were incubated at 37°C for 24 h. After incubation, the diameter of the inhibition zone was measured with scale and the results were recorded in mm (data not shown).

Analysis of algal crude extracts using gas chromatography-mass spectrometry (GC-MS)

The gas chromatography coupled with mass spectrometry detection technique allows good qualitative and quantitative analysis of the fractionated extracts with high sensitivity to smaller amounts of components. Accordingly, identification of the chemical constituents of fractionated extracts, for the selected microalgae which showed effective antibacterial activities against the test bacteria were analyzed. This was done by using 1 µl of each sample was injected into an RT x -5 column (30X0.32 nm) of GC-MS model (Perkin Elmer, Clarus 500, USA) and Helium (3 ml/min) was used as a carrier gas. The following temperature gradient program was used; 75°C for 2 min followed by an increase from 75 to 175°C at a rate of 50°C per min and finally 7 min at 175°C. The m/z peaks representing mass to charge ratio characteristics of the antimicrobial fractions were compared with those in the mass spectrum library

Table 1. Antibacterial activities of crude extract from different soil algal species isolated in Saudi Arabia.

Crude extract (Acetone/Methanol/Di-Ethyl-ether, 5:2:1 v/v)	*S. aureus*	*B. subtilis*	*S. sonnei*
Chlorella vulgaris	_	+	+++
Spirulina platensis	_	_	+++
Nostoc linckia	+	_	+++
Phormidium autumnale	_	++	+++
Dunaliella salina	_	_	++
Tolypothrix distorta	++	+++	+++
Microcystis aeruginosa	+	_	++

(-) No activity, (+) low activity, (++) moderate activity, (+++) high activity.

of the corresponding organic compounds (Pandey et al., 2010). The chemical components of the extracts were analyzed in the central laboratory of King Saud University, Riyadh, Saudi Arabia. Identification of the chemical constituents of extracts were made using Perkin Elmer (Clarus 500, USA) gas chromatography coupled with (Clarus 500, USA) mass spectrometer (MS). Neither internal nor external chemical standards were used in this chromatographic analysis. Interpretation of the resultant mass spectra were made using a computerized library-searching program (NIST database) and by studying the fragmentation pattern of such compound resulted from mass spectrometry analysis. Concentration of such compound was calculated by the following formula:

Compound concentration percentage= [P1/P2] x 100

Where, P1 is the peak area of the compound and P2 is whole peak areas in the fractionated extracts.

RESULTS

Soil cyanobacteria isolated from the cultivated fields of different places in Saudi Arabia was evaluated for antimicrobial activity. Mixture of methanol, acetone and diethyl ether extracts from 7 algae were examined for antimicrobial properties against six bacteria and one fungus. Of total microalgae, 100% (5 cyanobacteria and 2 green algae) exhibited antimicrobial effects. Selected cyanobacteria with positive antimicrobial activities were *C. vulgaris*, *S. platensis*, *N. linckia*, *P. autumnale*, *D. salina, T. distorta* and *M. aeruginosa*. Antibacterial activities for all the crude extracts of algal species were examined (Table 1) and significant bioactivities against *S. aureus*, *B. subtilis* and *S. sonnei* were determined. All the algae species used in this study showed strong inhibition against *S. sonnei*. Considering fungi, no antimicrobial activity was observed in all the tested algae.

Chemical analysis of the potent algal extract

To determine the active organic components within the described algal species, GC-MS analysis were performed for all of the potent crude extracts. Chemical composition and concentrations of the analyzed fractions are presented in Table 2. In our data, the major peaks in the gas chromatogram were assigned with the highest percentage of the compound concentration in the total extract (REV values). For each algal species, the most 2 intensive fraction was recorded such as *P. automonate*: 1-Hexyl-2-Nitrocyclohexane; Cyclohexane; 1-(1,5-Dimethylhexyl)-4-(4-Methylpentyl)-; *S. platensis*: 2-Butanol; 3-Methyl-(S)-; 2-Hexanol (S)-; *C. vulgaris*: Bromoacetic acid; Pentadecyl ester; 1-Hexyl-2-Nitrocyclohexane; *N. linckia*: Octadecanal; 1,37-Octatriacontadiene; *D. salina*: 3-Methyl-2-(2-Oxopropyl) Furan; 1-Hexyl-2-Nitrocyclohexane; *T. distorta*: Boronic acid; Ethyl-Dimethyl ester; 7,9-Di-Ter-Butyl-1-Oxaspiro(4,5) Deca-6,9-Diene-2,8-Dione; *M. aeruginosa*: (S)-(+)-1-Cyclohexylethylamine; Boronic acid, Ethyl-Dimethyl ester (Table 2). The resultant major peaks of the extract were surveyed by the use of the available data base PubChem, provided by the National Center for Biotechnology Information (NCBI) at http://pubchem.ncbi.nlm.nih.gov. Classification, biomedical features and biological assay activity was obtained for some of the resultant fractions, and data are represented in Table 3.

DISCUSSION

Emergence of microbial diseases in pharmaceutical industries implies serious loss. Usage of commercial antibiotics for human disease treatment produces undesirable side effects. Cell extracts and active constituents of various algae may be potential bioactive compounds of interest in the pharmaceutical industry (Rodrigues et al., 2004). In the current research, combination of methanol, acetone and diethyl ether was the best solvents for extracting the bioactive compounds compared to acetone, methanol and diethyl ether alone (data not shown), meanwhile it gave the highest antimicrobial activities against the selected pathogens. This was in contrast with the study of Tuney et al. (2006). However, Das et al. (2005) examined acetone, ethanol and methanol extracts of other algae and showed from moderate to high activity against strains of virulent

Table 2. The GC-MS analysis of different components in (methanol:acetone:diethyl ether) extracts of soil algal species.

S/N	Compound	Rev*
Phormidium autumnale		
1	Boronic acid, Ethyl-, Dimethyl ester	843
2	3,4-Hexanediol, 2,5-Dimethyl-	772
3	2-Butanol, 3-Methyl-, (S)-	882
4	2-Butanol, 3-Methyl-	862
5	Butanal	874
6	2-Thiophenecarboxylic acid, 5-(1,1-Dimethylethoxy)-	854
7	Cyclopropanepentanoic acid, 2-Undecyl-, Methyl Ester, Trans-	804
8	1,6-Anhydro-3,4-Dideoxy-.Beta.-D-Gluco-Hexopyranose	773
9	1-Hexyl-2-Nitrocyclohexane	917
10	Cyclohexane, 1-(1,5-Dimethylhexyl)-4-(4-Methylpentyl)-	903
11	1-Hexyl-1-Nitrocyclohexane	870
Chlorella vulgaris		
1	N-(3-MethylButyl) Acetamide	776
2	D-Mannoheptadecane-1,2,3,4,5-Pentaol (methyl ester-fatty acid)	769
3	2-Butanol, 3-Methyl-, (S)-	908
4	2-Hexanol, (S)-	905
5	5,9-Dodecadien-2-one,6,10-Dimethyl-, (E,E))-	773
6	Heptanal	772
7	3-Decyn-2-Ol	763
8	3-Nonyn-2-Ol	755
9	2(3H)-Furanone, 3-(15-Hexadecynylidene)Dihydro-4-Hydroxy-5-Methyl-, [	887
10	3-Methyl-2-(3-Methylpentyl)-3-Buten-1-Ol	884
11	(3R,2E)-2-(Hexadec-15-Ynyliedene)-3-Hydroxy-4-Methylenebutanolide	893
12	4-Methyldocosane	879
	Spirulina platensis	
1	1-Propanamine, N1-Methyl-2-Methoxy	682
2	1,2-Benzendicarboxylic acid, Bis(2-Ethoxyethyl) Ester	673
3	1-Dodecanol, 3,7,11-Trimethyl	849
4	Hexadecen-1-Ol, Trans-9-	842
5	3-Decyn-2-Ol	838
6	1,6;2,3-Dianhydro-4-Deoxy-.Beta.-D-Lyxo-Hexopyranose	825
7	3-Chloropropionic acid, Heptadecyl Ester	892
8	Acetic acid, Chloro-, Hexadecyl Ester	887
9	retsE lycedatneP ,dica citecaomorB	909
10	10-Heneicosene (C,T)	901
11	2(3H)-Furanone, 3-(15-Hexadecynylidene)Dihydro-4-Hydroxy-5-Methyl-, [	904
12	(3R,2E)-2-(Hexadec-15-Ynyliedene)-3-Hydroxy-4-Methylenebutanolide	890
13	enaxeholcycortiN-2-lyxeH-1	921
14	1-Hexyl-1-Nitrocyclohexane	893
15	1-Propanamine, N1-Methyl-2-Methoxy	682
16	1,2-Benzendicarboxylic acid, Bis(2-Ethoxyethyl) Ester	673
17	1-Dodecanol, 3,7,11-Trimethyl	849
	Nostoc linckia	
1	Boronic acid, Ethyl-, Dimethyl ester	848
2	3,4-Hexanediol, 2,5-Dimethyl-	772

Table 2. Contd.

3	2-Butanone, 3-Hydroxy-	828
4	(S)-Isopropyl Lactate	797
5	(EDYHEDLA) lanacedatcO	868
6	eneidatnocairtatcO-1,37	861
	Dunaliella salina	
1	2-Amininonadecane	837
2	1,6;3,4-Dianhydro-2-Deoxy-.Beta.-D-Lyxo-Hexopyranose	792
3	2-Hexanol, Acetate	811
4	Octanal	785
5	3-Methyl-2-(2-Oxopropyl)Furan	909
6	1-Hexyl-2-Nitrocyclohexane	906
7	2-Octadecyl-Propane-1,3-Diol	888
8	Pentadecanal-	887
	Tolypothrix distorta	
1	Boronic acid, Ethyl-, Dimethyl ester	839
2	3,4-Hexanediol, 2,5-Dimethyl-	778
3	7,9-Di-Ter-Butyl-1-Oxaspiro(4,5)Deca-6,9-Diene-2,8-Dione	791
4	Silane,Trichlorodecyl-	714
5	Tetraethylene Glycol Diethyl Ether	712
6	Ethane, 1,1-Diethoxy-	699
	Microcystis aeruginosa	
1	(S)-(+)-1-Cyclohexylethylamine	919
2	Octodrine	842
3	Boronic acid, Ethyl-, Dimethyl ester	849
4	3,4-Hexanediol, 2,5-Dimethyl-	772
5	2-Butanone, 3-Hydroxy-	831
6	(S)-Isopropyl Lactate	801
7	2-Thiophenecarboxylic acid, 5-(1,1-Dimethylethoxy)-	786
8	Cyclobutanol	790
9	2-Dodecen-1-Yl(-)Succinic Anhydride	793
10	Cyclohexanol, 4-Ethyl-4-Methyl-3-(1-Methylethyl)-, (1.Alpha.,3.Alpha.,4.	785
11	Decane, 5,6-Bis(2,2-Dimethylpropylidene)-, (E,Z)-	782
12	Cyclohexanol, 4-Ethyl-4-Methyl-3-(1-Methylethyl)-, (1.Alpha.,3.Alpha.,4.A	780

*(Rev), % of the compound concentration in the total extract.

Table 3. Biomedical properties of different mass spectra components in crude extracts belonging to soil algal species.

Algal species	Mass spectra compound	Classification	Compound ID in PubChem (CID)	Pharmacological and biomedical effects
T. distorta	Ethane, 1,1-Diethoxy-	Stearyl alcohol	7765	(+) Ezra et al.,1998
P. automonate	Butanal	Aldehyde	261	(+) Gregory et al., 2006
C. vulgaris	Heptanal	Aldehyde	8130	(+) Cueto et al., 1992
D. salina	Octanal	Caprylic Aldehyde	454	(+) Singer, 2000

Reported properties (+), no reported activity (-).

pathogens *Pseudomonas florescence*, *Aeromonas hydrophila*, *Vibrio anguillarum* and *E. coli*.

In this study, 4 cyanobacteria and 2 green microalgae were tested in compliance with the agar well diffusion

method for their antibacterial agent production on various organisms that incite diseases of humans. The antimicrobial activity was maintained by using mixture of methanol, acetone and diethyl ether. It was found that, cyanobacteria *T. distorta* and *P. autumnale* had the highest antibacterial activity towards the tested bacteria. In a previous study by Rania and Taha (2008), 3 cyanobacteria (*Anabaena oryzae*, *Tolypothrix ceytonica* and *Spirulina platensis*) and 2 green microalgae (*Chlorella pyrenoidosa* and *Scenedesmus quadricauda*) were tested in compliance with the agar well diffusion method for their antibacterial and antifungal agent production on various organisms that incite diseases of humans and plants (*Escherichia coli*, *Bacillus subtilis*, Staphyllococcus *aureus*, *Pseudomonas aeruginosa*, *Aspergillus niger*, *Aspergillus flavus*, *Penicillium herquei*, *Fusarium moniliforme*, *Helminthosporium* sp., *Alternaria brassicae*, *Saccharomyces cerevisiae*, *Candida albicans*). In their study, the antimicrobial activity was maintained by using ethanol, acetone, diethyl ether and methanol. It was found that, *Spirulina platensis* and *Anabaena oryzae* had the highest antibacterial and antifungal activity towards the tested bacteria and fungi (Rania and Taha, 2008).

Very recently, in another similar study, methanolic extracts of four marine algae of Algeria coast were investigated for antibacterial activity against six pathogenic bacteria (*Bacillus subtilis, Listeria innocua, S. aureus, E. coli, Klebsiella pneumonia* and *Pseudomonas aeruginosa*). Susceptibility assays using disc diffusion and broth micro dilution test for the determination of minimum inhibitory concentration (MIC) were employed to assess the antibacterial activity of methanolic extracts of algae. All algae extracts showed antibacterial activity against four of the six pathogenic bacteria tested with MIC values ranged between 0.25-3.0 mg/ml. In their study, extract of *Rhodomela confervoïdes* exhibited the highest activity against *Bacillus subtilis* (24.0 mm) and *Cystoseira tamariscifolia* exhibited the highest activity against *Listeria innocua* (19.67 mm) (Bedjou et al., 2011).

On the other hand sufficient data regarding crude extracts of seaweed and terrestrial algae have been obtained in previous studies, revealing the antimicrobial properties of organic compounds derived from natural sources (Robles-Centeno et al., 1996; Manilal et al., 2009b; O'Sullivan et al., 2010; Wijesinghe and Jeon, 2011). Analysis of bioactive metabolites have been studied mostly with marine (Wijesinghe and Jeon, 2011) algal species (Caccamese et al., 1985; Lima-Filho et al., 2002). On the other hand, little is known for the terrestrial or soil originated algae, but some studies were subjected to cyanobacteria (Bloor and England, 1989; Burja et al., 2001).

In the current study, all our isolates belonged to cyanophytae and chlorophytae groups of soil origins, assigned antibacterial effect of these species are presented in Table 1. The most effective activity was recorded for *T. distorta* against *S. aureus, B. subtilis* and *S. sonnei*, followed by *P. autumnale* affecting *B. subtilis* and *S. sonnei*.

In the present work, a simple chemical methodology to carry out the screening for natural functional compounds is presented. To do that, a strategy has been conducted including the use of unexplored natural sources (that is, algae and microalgae) together with environmentally clean extraction techniques and advanced analytical tools.

The procedure also allowed estimating the functional activities of the different extracts obtained and even more important, to correlate these activities with their particular chemical composition. By applying this methodology, Plaza et al. (2010) reported that it is possible to carry out the screening for bioactive compounds in the algae *Himanthalia elongata* and the microalgae *Synechocystis* sp. Both algae produced active extracts in terms of both antioxidant and antimicrobial activity. In their study, the obtained pressurized liquid extracts were chemically characterized by GC-MS and HPLC-DAD. Different fatty acids and volatile compounds with antimicrobial activity were identified, such as phytol, fucosterol, neophytadiene or palmitic, palmitoleic and oleic acids. Based on the results obtained, ethanol was selected as the most appropriate solvent to extract this kind of compounds from the natural sources studied.

Crude extract analysis of the described species using gas chromatography-mass spectrometry (GC-MS) had revealed several important organic volatile compounds as fatty acids in this study (Table 2). Similar to our findings, it was reported that the soil cyanobacteria isolated from the paddy fields of seven provinces in Iran was evaluated for antimicrobial activity. Aqueous, petroleum ether, and methanol extracts from 76 microalgae were examined for antimicrobial properties against four bacteria and two fungi. Of the total microalgae, 22.4% (17 cyanobacteria) exhibited antimicrobial effects (Soltani et al., 2005).

Little privileges to the biomedical properties for these compounds have been assessed so far, however, the bioactivity of fatty acids has been approved in certain microorganisms and fouling organisms (Russel, 1991). Such compounds have showed antimicrobial properties (Katayama, 1960; Bloor and England, 1989; Khairy and El-Kassas, 2010). Similarly, in another study, the methanol, dichloromethane, petroleum ether, ethyl acetate extracts and volatile components of *Spirulina platensis* were tested *in vitro* for their antimicrobial activity (four Gram-positive, six Gram-negative bacteria and *Candida albicans* ATCC 10239).

GC-MS analysis of the volatile components of S. platensis resulted in the identification of 15 compounds which constituted 96.45% of the total compounds. The volatile components of S. platensis consisted of heptadecane (39.70%) and tetradecane (34.61%) as major components. The methanol extract showed more potent antimicrobial activity than dichloromethane,

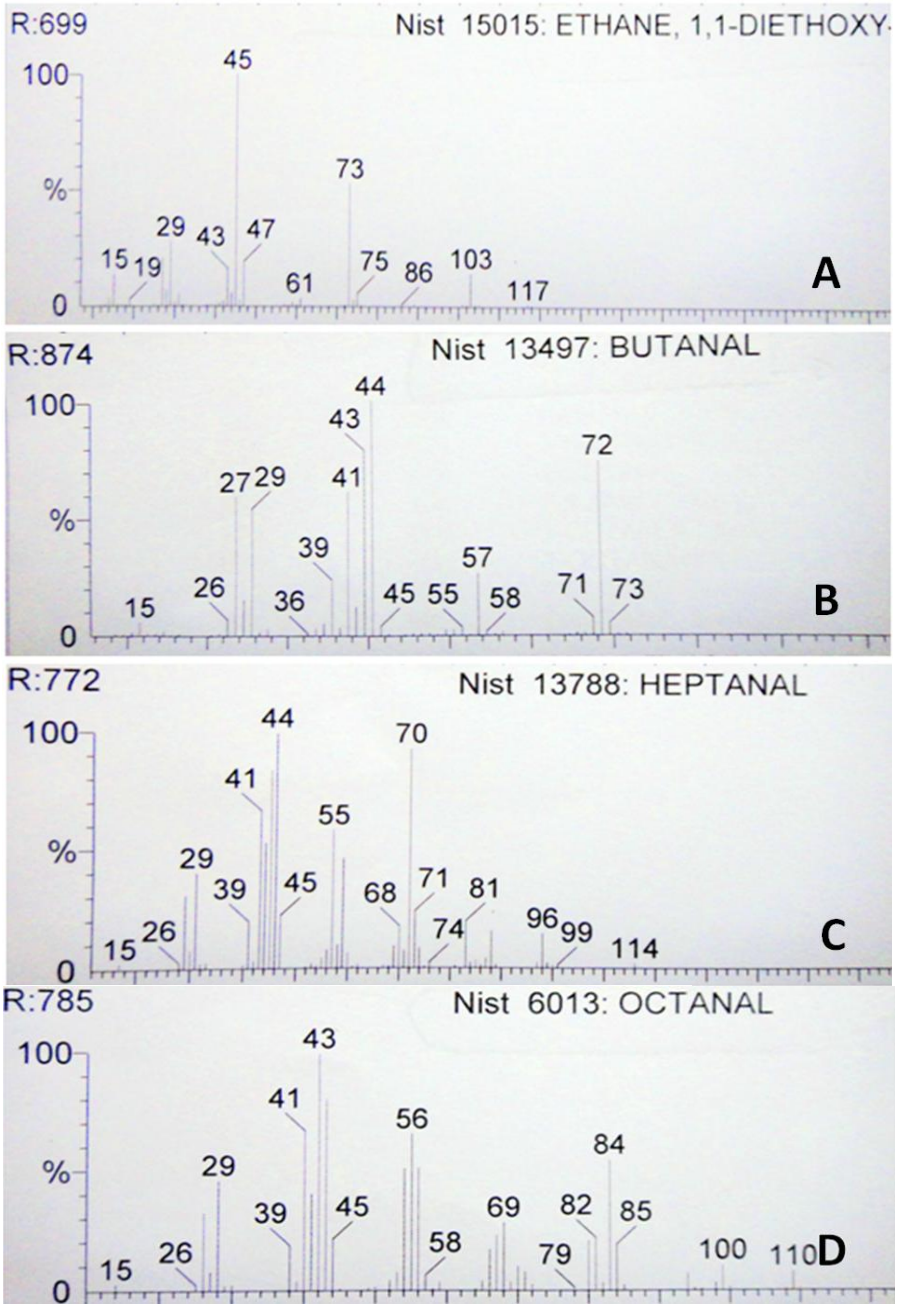

Figure 1. Mass spectrum of ethane,1,1-diethoxy- (A); butanal (B); heptanal (C) and octanal (D) in the methanol:acetone:diethyl ether extract of *T. distorta; P. automonate*; *C. vulgaris* and *D. salina*, respectively.

petroleum ether, ethyl acetate extracts and volatile components (Karabay et al., 2007). These findings were correlated with our present observation, as shown in Table 2. In this study, similar chemical components with different percentage were detected for some species belonging to different groups probably due to the difference in polarity of the used solvents and to habitat environmental factors.

The mass spectra of the compounds were investigated with those similar in the PubChem database and some of our chemical components are reported to have a known biomedical value in the pharmacological fields (Table 3). Fractionated matrices of *T. distorta* crude extract, contained as stearyl alcohol and Ethane, 1,1-Diethoxy- which is known to demonstrate valuable therapeutic uses including anti-inflammatory, antipyretic, antithrombotic and analgesic effects (Figure 1). Interestingly, some of our resultant spectra compounds exhibited important

biomedical features as described in Table 3. Fractionated constituents specified in each algal species were inspected for biomedical characteristics such as butanal was applied in controlling Bovine soles symptoms in cattle (Gregory et al., 2006); heptanal was investigated in vitro as biomarker for ozonation, suggesting an ozonation mechanism in lung cell lines of rats (Cueto et al., 1992). Finally, octanal was studied with olfactory receptor model, suggesting an interaction of octanal with such receptors in rat cell lines (Singer, 2000). In this preliminary study, assessing the biomedical characteristics of such compounds in the assayed species was still under prediction. Further, utilization of naturally occurring bioactive compounds from algae could obtain a wide alternatives of manufactured therapeutics. Interestingly, these findings would be the opening the new trends in biomedical and pharmaceutical industries in the region.

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