Se separaron diferentes clases sulfolípidos (SL) a partir de los lípidos totales de cinco especies de algas marinas: una especie de Chlorophyta (Ulva fasciata), dos especies de Phaeophyta (Dilophys fasciata, Taonia atomaria) y dos especies de Rhodophyta (Laurencia popillose, Galaxoura cylindriea) que se purificaron mediante cromatografía en columna de DEAE-celulosa. Los componentes de SLs fueron identificados por IR, cromatografía de gases MS/MS y cromatografía líquida MS/ MS. Los contenidos de SL en relación al total de lípidos varió de 1,25% (en L. papilosa) al 11,82% (en D. fasciata). Sin embargo, no hay diferencias significativas en el contenido de sulfato observado entre todas estas especies de algas (desde 0,13 hasta 0,21%). Todos los SL se caracterizaron por un alto contenido de ácido palmitico (C 16:0), que osciló entre 30,91% en G. cylindriea a 63,11% en T. atomaria. El sulfato quinovosido-diacilglicerol y acilglicerol sulfatoquinosilo fueron identificados como los principales constituyentes de los sulfolípidos de estas algas. Los sulfolípidos de las diferentes especies de algas estudiadas mostraron una notable actividad antiviral contra el virus del herpes simple tipo 1 (VHS-1) con una IC 50 entre 18,75 y 70,2 µg mL –1 . Por otra parte, los sulfolípidos de estas algas inhibieron el crecimiento de células tumorales de adenocarcinoma de mama humano (MCF7) con una IC 50 que osciló entre 30,91% en G. cylindriea a 63,11% en T. atomaria. La sulfatoquinosil-diacilglicerol y acilglicerol sulfatoquinosilo fueron identificados como los principales constituyentes de los sulfolípidos de estas algas. Los sulfolípidos de diferentes especies de algas antimicrobianas mostraron una notable actividad antimicrobiana contra el virus del herpes simple tipo 1 (VHS-1) con una IC 50 que osciló entre 18,75 y 70,2 µg mL –1 . Por otra parte, los sulfolípidos de estas algas inhibieron el crecimiento de células tumorales de mama y células de cáncer de hígado humano con valores de IC 50 que van desde 0,40 hasta 0,67 µg mL –1 para las células de adenocarcinoma de mama humano (MCF7).

PALABRAS CLAVE: Algas marinas contra el cáncer – Antibacterianas – Antivirales – Células – HepG2 – HSV-1 – Marine algae – MCF7 – Sulfolipids.

1. INTRODUCTION

Glycolipids (GL), sulfolipids (SL) and phospholipids (PL) are usually present in all photosynthetic membranes in plants, algae and various bacteria. However, Sulfolipids (SLs) represent up to 29% of total lipid and have been regarded as predominant lipid components in both prokaryotic and eukaryotic organisms (Norman et al., 1996; Benning and Somerville, 1992). SLs are located in the thylakoid membrane as an important component to preserve the membrane’s structure and function. Recently, lipid constituents have been recognized as potentially important factors in the processes of signal transduction and endomembrane transport (Simons and Toomre 2000). However, algae represent valuable sources of a wide spectrum of complex lipids, and their components have promising potential applications especially in the food, cosmetic, and pharmaceutical industries (Hossain et al., 2005). The various biological actions of SLs include: inhibitory effects on DNA polymerase and viral reverse transcriptase, antitumor, anti-inflammatory, and inhibition and promotion of cell growth (Mizushina et al., 1998; Liptak et al., 2004; Maeda et al., 2008; Naumann, 2009). Several reports have indicated that SLs compounds isolated from...
different algae species inhibited the growth of several viruses (such as HIV, HSV-1 and the AIDS virus) as well as cancer types (Gustafson et al., 1989; Xue et al., 2002; Chirauswan et al., 2009). For instance, the sulfolipids of S. horneri were found to possess a potent anticancer activity against colon cancer cells (Caco-22 cells) (Hossain et al., 2005). In addition, the SLs of Porphyridium cruentum showed inhibitory effects on human colon (DLD-1), breast (MCF-7), prostate adenocarcinoma (PC-3) and malignant melanoma (M4 Meu) cancer cells (Bergé et al., 2002).

The aim of the present work is to characterize the chemical structure and evaluate the biological activity of the sulfolipids of some marine algae collected from the Red and Mediterranean seas in Egypt.

2. MATERIALS AND METHODS

2.1. Collection of marine algal samples

Samples of Laurencia popillose and Galaxoura cylindrea were collected from the Red Sea (Faied and Ein Al-Sokhna at Suez gulf canal). Ulva fasciata and Taonia atomaria were collected from the Mediterranean Sea at (Abu-Qir city) Alexandria Governmate and Dilophy fusciola from (Matroh city) Marsa Matrouh Governament. All algal samples were washed several times with tap water and then left to air dry. Samples of 500 g were ground and stored in brown glass containers at room temperature for further analysis.

2.2. Identification of marine algae species

After preparation of herbarium specimens of the algae species, they were identified by Dr. Rauhaiya Abdul-Latif, Professor of Botany, Department of Botany, Faculty of Science, Al-Azhar University.

2.3. Extraction and determination of total lipid

The total lipids of marine algae (10 g) were extracted with 100 mL of the mixture: methanol: chloroform (2:1, v/v) (Roughan and Bratt, 1968). After filtration, the mixture was evaporated at 40 °C to a minimum volume (15 mL) and dried under N₂. Then, the total lipid contents were determined by weight.

2.4. Isolation and determination of algal sulfolipids

The sulfolipids were separated from the total lipid extracts using diethylaminoethyl-cellulose (DEAE-cellulose) column chromatography (0.6 x 6 cm, i.d.), then eluted with a 21.5 mL mixture of chloroform/ methanol (20 mL: 6:4, v/v) and concentrated NH₄ (1.5 mL) (Roughan and Bratt 1968). The mixture, which contained purified sulfolipids, was dried at 40 °C, under N₂ then the total sulfolipids were calculated by weight.

2.5. Identification of algal sulfolipids

2.5.1. Determination of total sulfate of algal sulfolipids

The sulfate content of algal sulfolipids was determined using a sodium rhodizionate reagent, which in the presence of barium forms a red compound. The reduction of color intensity indicates the quantity of sulfate present in the sample. A standard calibration curve was prepared using sodium sulfate (Na₂SO₄), scaled for 1.0 – 12.0 µg sulfate (Terho and Hartiala, 1971).

2.5.2. Identification of algal sulfolipid fatty acids

The algal sulfolipids were subjected to direct transmethylation in 1.5% sulfuric acid:methanol at 95 °C for 2 h (Luddy et al., 1960). Fatty acid methyl esters were analyzed by gas chromatography (Perkin Elmer Autosystem XL) equipped with a flame ionization detector and fused silica capillary column (DB-5 (American) 60 m x 0.32 mm, i.d.) with a film thickness of 0.25/25µm. The column temperature was initially 150 °C and was then gradually increased at rate of 3 °C min⁻¹ up to 250 °C. The injector and detector temperatures were 230 °C and 250 °C, respectively. Helium was used as the carrier gas (at 1 mL min⁻¹). The split ratio was 1/100. The fatty acids were identified by comparison between the retention times of the samples and those of methyl fatty acid standard mixtures ( Sigma, > 99% purity by GLC).

2.5.3. Identification of function group of marine algal sulfolipids using IR

The IR spectra of algal sulfolipid samples were recorded on a JASCO FT/IR 6100A spectrometer in the wave number range 4000-400 cm⁻¹, according to the KBr disk method.

2.5.4. LC-MS-MS analysis of algal sulfolipids

An aliquot of sulfolipid fraction was analyzed by LC-MS-MS (LCQ Advantage Max, Thermo Finnegan, USA) using a triple mass spectrometer operating in positive electro spray ionization (ESI). The heated capillary and voltage were maintained at 255 °C and 4.5 KeV, respectively. The full scans of mass spectra of the sulfolipids were carried out from m/z 500 to 2000 using 500 ms for the collection of ion in the trap. MS/MS was used to break down the most abundant [M+H]⁺ ion from MS with depended collection induced dissociation (CID) (Pons et al., 2002).

2.5.5. Identification of algal sulfolipids using GC-MS

Sulfolipids were identified using GC-MS (Chrompack France, Les Ullis, France) equipped
with a 0.25 µm film phase and fused CP-Sil5 CB Low bleed-MS capillary column (25 m x 0.32 mm). The temperature of the injector was 280°C. Sulfolipids were analyzed using the following temperature program: 90°C for 3 min then gradually increased at the ratio of 5°C min−1 until 260°C. The analyses were performed in the EI mode (ionization energy 70 eV; source temperature 150°C) (Pons et al., 2002).

2.6. Biological evaluation of marine algal sulfolipids

2.6.1. Antiviral activity of algal sulfolipids

Preparation of the algal extract for bioassay. A stock solution of algal SLs was freshly prepared by dissolving 100 mg of sulfolipid fraction in 10 mL of dimethyl sulfoxide (DMSO) in water (9:1, v/v) and kept at 4°C until use; appropriate dilutions of the solution were used in each assay. All the tests were carried out in three independent assays, and the means were applied.

Antiviral screening of algal sulfolipids. Algal SLs were evaluated for antiviral activity against herpes simplex virus type-1 (HSV-1). The virus was obtained from the Virology Laboratory, Water Pollution Research Dept., National Research Center (NRC), Egypt. The virus was propagated in viro cell cultures. The Inhibition % of the virus was calculated as plaque reduction as a result of being subjected to a given extract (Tebas et al., 1995).

2.6.2. Antitumor activity of algal sulfolipids

The potential antitumor activity of algal sulfolipids was tested using the method of Skehan et al. (1990). Human hepatocellular carcinoma cells (Hep G2) and breast adenocarcinoma cells (MCF-7) were plated on 96 multi-well plates for 24 h before treatment with the algal sulfolipid to allow for the attachment of cells to the well of the plate. Sulfolipids and antitumor reference drugs (Novantron) were added at serial concentrations to cell monolayers. After incubation for 48 h at 37°C in an atmosphere of 5% CO2, the cytotoxicity was determined spectrophotometrically by measuring the developed color at 570 nm by the ELISA reader (Tecan Sunrise absorbance reader (No. 3008746), software Magllan V.4 , Germany).

2.6.3. Antimicrobial activity of algal sulfolipids

The antimicrobial activities were determined by the conventional agar diffusion assay (Greenwood, 1983) using one gram positive (Bacillus subtilis NRRL B-94) one gram negative (Escherichia coli NRRL B-3703) bacteria, fungi (Aspergillus niger NRRL 313), and yeast (Candida albicans NRRL 477). The microgrowth inhibition zone was measured after incubation at 30°C by the appearance of a clear, microbial free inhibition zone, beginning within 24 h for yeast, 24-48 h for bacteria and 72-96 h for fungus. The most active sulfolipid fractions were tested for their MIC according to Hammer et al., (1999). MIC was determined as the lowest concentration of sulfolipid fractions inhibiting the visible growth of each organism on the agar plate.

2.7. Statistical analysis

Data were statistically analyzed through the analysis of variance (ANOVA) and Duncan’s test and the P> 0.01 probability level was applied (Gomes and Gomes, 1984).

3. RESULTS AND DISCUSSION

3.1. Marine algal total lipid contents

The total lipid contents (TL) of marine algae ranged from 0.09 to 2.35% (Table 1). U. fasciata (2.55%) had the highest TL content followed by D. fasciola (1.11%), whereas the lowest level was found in G. cylindriea (0.09%) followed by T. atomaria (0.66%) and L. papillosa (0.88%). However, the concentrations of TL in the tested algae species were within the ranges (1.0 – 5.0%) reported in the literature for several algae species (Matanjun et al., 2009; Manivannan et al., 2008). For instance, the TL contents in 12 species of marine algae belong to 3 families from

| Algae species | Total lipid Content % | Total sulfolipid (% of total lipid) | Sulfate % |
|---------------|-----------------------|------------------------------------|-----------|
| U. fasciata   | 2.35d                 | 3.60                               | 0.13a     |
| T. atomaria   | 0.66h                 | 4.50                               | 0.16a     |
| D. fasciola   | 1.11*                 | 11.80                              | 0.21a     |
| L. papillosa  | 0.81*                 | 1.25                               | 0.16a     |
| G. cylindriea | 0.09*                 | 10.00                              | 0.17a     |
| LSD           | 0.09                  |                                    | 0.07      |

The mean (n = 3) difference is significant at P ≤ 0.01.

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the Chlorophyceaean (4 species), Phaeophyceae (5 species) and Rhodophyceae (3 species) and ranged from 1.33 ± 0.20% to 4.60 ± 0.17% (Manivannan et al. 2008). In general, the total lipid contents varied among alga species depending on species, genetic origin, climate and the geography of the development of the algae (Araki et al. 1990).

3.2. Algal total sulfolipid contents

The sulfolipid fractions of five macroalgal lipids are illustrated in Table (1). The sulfolipid fraction content ranged from 1.25 to 11.80% of the total lipids. The highest sulfolipid content was found in D. fasciola (11.80%) followed by G. cylindriea (10.00%). The lowest sulfolipid fractions were detected in T. atomaria (4.50%), U. fasciata (3.60%) and L. papillose (1.25%).

3.2.1. Sulfate content of marine algal sulfolipids (SLs)

The sulfate content among the algae sulfolipids ranged between 0.13 and 0.21%. The highest sulfate content was found in D. fasciola (0.21%) followed by G. cylindriea (0.17%), T. atomaria (0.16%), L. papillose (0.16%) and U. fasciata (0.13%). These results agree with those obtained by Sanina et al., (2004) in which the sulfolipid contents of the total lipids in A. tobuchiensis (Rodophyta), L. japonica and S. pallidum (Phaeophyta), U. fenestra (Chlorophyta) and Z. marina (Embriophyta) were 10.0%, 15.0%, 7.1%, 9.7% and 6.2%, respectively. In addition, these results are in accordance with those of Gerasimenko et al., (2010), who stated that the sulfolipid contents of juvenile and adult brown algae Costaria costata ranged from 0.4 to 10.6% of the total lipids.

3.2.2. Fatty acid compositions of marine algal sulfolipids

The fatty acid compositions of the algal sulfolipid fraction are presented in Table 2. All algal SLs have high contents of palmitic acid (C16:0), ranging from 30.9% (G. cylindriea) to 63.1% (T. atomaria); while C20:5 was detected in T. atomaria (13.8%), G. cylindriea (13.8%) and L. papillose (10.80%). Margaric acid (C17:0) was identified as the main fatty acid in the SLs of U. fasciata (21.6%) and L. papillose (12.9%). A remarkable content of arachidonic acid (C20:4) was detected in G. cylindriea (37.4%). Oleic acid (C18:1) and linoleic (C18:2) were also found in considerable amounts at 11.4% in sulfolipid of U. fasciata and 27.1 and 10.7% in the sulfolipid fraction of D. fasciola and L. papillose, respectively. These results reveal that fatty acid composition differed significantly among all the algae species. The FA composition of the sulfolipid fraction is similar to those previously reported for green, brown and red algae (Sanina et al., 2004). Similar results were obtained by Araki et al., (1989), who found that the sulfoquinovosyl diacylglycerols (SQDG) of G. verrucosa were characterized by high levels of C16:0 and C20:4 fatty acids. P. incisa sulfolipid (SQDG) were rich in C16:0, C18:1ω7, C18:2 and C18:3ω3 (Bigogno et al., 2002). Xue et al. (2002) found that the main fatty acids of Spirulina platensis SQDG were C16:0, C18:2ω6, and C18:3ω6. The major fatty acids of sulfoquinovosyl-di-acylglyceride in Phaeophyta, Chlorophyta and Rhodophyta were saturated and monounsaturated.

| Fatty acids | Algae strains |
|-------------|---------------|
|             | Mediterranean sea | Red sea |
|             | U. fasciata | T. atomaria | D. fasciola | L. papillose | G. cylindriea |
| C16:0       | 0.90        | 10.9        | 3.9        | 4.7        | 2.8        |
| C16:0       | 49.9        | 63.1        | 57.7       | 34.5       | 30.9       |
| C16:1       | –           | –           | 1.2        | 2.1        | 1.2        |
| C17:0       | 21.5        | 4.51        | 0.78       | 12.8       | 1.0        |
| C18:0       | 2.8         | 4.0         | 1.9        | 6.4        | 4.0        |
| C18:1       | 11.3        | 3.0         | 3.1        | 6.4        | 3.1        |
| C18:2       | 2.4         | –           | 27.1       | 10.7       | 4.4        |
| C20:3       | 1.7         | 0.5         | 0.1        | 0.75       | 1.2        |
| C20:4       | 1.3         | –           | –          | 1.6        | 37.4       |
| C22:5       | 7.4         | 13.8        | 3.9        | 10.8       | 13.8       |
| SFAs %      | 75.2        | 82.6        | 64.4       | 58.5       | 38.7       |
| MUFAs %     | 11.3        | 3.0         | 4.3        | 8.5        | 4.4        |
| PUFAs %     | 5.5         | 14.3        | 31.1       | 23.6       | 43.0       |

FA, fatty acid; SFAs, saturated fatty acids; MUFAs, monounsaturated fatty acids; PUFAs, Polyunsaturated fatty acids.
fatty acids except for SQDG from Chlorophyta (Z. marina), which contained the highest contents of C<sub>18:2</sub> n-6 and C<sub>18:3</sub> n-3 acids (Khotimchenko, 2003). Hossain et al. (2005) found that the major fatty acids of SQDG from S. horneri (brown algae) were C<sub>16:0</sub>, C<sub>18:1</sub>, C<sub>18:3</sub>, C<sub>20:1</sub>, C<sub>20:4</sub>, and C<sub>22:5</sub>.

3.3. Proposed algal sulfolipid structures of bioactive constituents

3.3.1. IR analysis

As illustrated in Figure 1, the infrared spectra of all algal sulfolipid fractions showed two characteristic absorption bands for sulphur-containing compounds. The first one appeared at 927 cm<sup>-1</sup>, indicating the presence of a strong dehydration of SO<sub>3</sub> and the second was at 771 cm<sup>-1</sup>, indicating a symmetrical C-O-S association with a C-O-SO<sub>3</sub> group (Ermanno et al., 1994 and Ranjaniv and Steven 1995). Regarding other absorption bands, there were large amounts of OH stretching at 3400 cm<sup>-1</sup>, symmetric CH<sub>2</sub> bending at 1380 cm<sup>-1</sup> and C-H stretching at 2930 cm<sup>-1</sup>.

3.3.2. Identification of marine algal sulfolipid compounds by GC-MS and LC-MS/MS

The proposed chemical structure of the active constituents of the algal sulfolipids was determined using GC-MS and LC-MS/MS. Several compounds of sulfolipids were separated from algae and two of them were identified by EI-MS and ESI/MS fragmentations. The EI/MS and EI/MS of the major component (compound 1) of sulfolipids consisted of molecular ion [M + H]<sup>+</sup> at m/z = 564.3 (Fig. 2) and was due to the loss of fatty acyl side chain as corresponding acid (palmitic acid, C<sub>16:0</sub>). The main fragmentations of compound 1 were corresponding to a likely loss of sulfonic acid (SO<sub>3</sub>H) and sodium salt (SO<sub>3</sub>Na), respectively. The product ion observed at m/z 345 appears to have originated by the loss of a fatty acyl side chain as corresponding acid (palmitic acid, C<sub>16:0</sub>). Based on the fragmentation pattern, the sulfolipid with pseudo-molecular ion [M–H + 2Na]<sup>+</sup> at m/z 601 was characterized as 2-O-palmitoyl-3-O(6′-sulfoquinovopyranosyl)-glycerol (Al-Fadhli et al., 2006). SQDG structures gave three molecular ions by ESI/MS at m/z 737.46, 765.77 and 793.77. These ions showed that the glycerol moiety was esterified by the following combinations of fatty acids: (C<sub>14:0</sub> + C<sub>16:0</sub>) giving m/z 737.46; (C<sub>14:0</sub> + C<sub>16:0</sub>) giving m/z 765.77 and (C<sub>14:0</sub> + C<sub>18:0</sub> or C<sub>16:0</sub> + C<sub>16:0</sub>) giving m/z 793.77. Tandem-MS gave fragments corresponding to sulfoquinovosyl (m/z 243.07 and 225.44); and sulfoquinovose linked to glycerol (m/z 283.35). Minor ions at m/z 509.71 and 537.83 are probably due to the respective loss of palmitic (C<sub>16:0</sub>) and myristic (C<sub>14:0</sub>) acids (De-Souza et al., 2007).

3.4. Biological evaluation of sulfolipid fractions

3.4.1. Antiviral activity

The antiviral activity of algal sulfolipids was evaluated according to the plaque reduction method and the results are summarized in Table 3. Algal sulfolipids showed high antiviral inhibitions against HSV-1, which ranged from 18.75 to 70.12 %. All algal sulfolipids had low virus inhibition at the concentration of 10.0 µg, while the highest inhibition HSV1 was observed at 20 µg mL<sup>-1</sup> for D. fasciola (70.12%). Thus, the sulfolipid fraction had dose-dependant antiviral activity. Among several antiviral available, acyclovir was used as a positive control in this study. The inhibitory effect of algae SLs (IC<sub>50</sub> ranged from 15.0 to 25.0 µg mL<sup>-1</sup>) was shown to be quite a bit more potent than that of acyclovir (IC<sub>50</sub> 5.5 µg mL<sup>-1</sup>). These results are in agreement with the results obtained by Chirasuwan et al., (2009) who found that sulfoquinovosyl-di-acylglycerol compounds extracted from Spiroplasma platensis have antiviral activity (HSV-1) in virus cells. Ohta et al., (1998) found that the new sulfolipid, 6-sulfo-alpha-D-quinovopyranosyl-1′,2′-diacylglycerol (SQDG) isolated from G. tenella (marine red alga) contains a potent inhibitor of eukaryotic DNA.
polymerases and HIV-reverse transcriptase type 1 (Gustafson et al. 1998). The SQDG fraction of *S. hofmanii* contains (C18:2/C16:0) and showed a higher antiviral activity against HCM-virus, with an IC50 of 19.0 µg mL⁻¹ (Naumann, 2009).

### 3.4.2. Antitumor activity

**Antitumor activity against human breast carcinoma (MCF-7).** The cytotoxic activities of five algal sulfolipid fractions were tested against MCF-7 and the results are illustrated in Table 4. All algal sulfolipid fractions showed high inhibition percentages (%) toward the MCF7 cell, which ranged from 66.24 to 94.19%. *L. papilllose* presented the highest antitumoral activity against the MCF-7 at all concentrations followed by *T. atomaria*, *G. cylindriea* and *U. fasciata*. In addition,
Algal sulfolipids of *T. atomaria* showed the highest inhibition activity (85.37%) against HepG2 cells followed by *D. fasciola* (80.29%), *L. papillosa* (79.89%) and *U. fasciata* (72.97%). Therefore, all the algae sulfolipids showed potential antitumor activity against HEPG2 with IC$_{50}$ values ranging from 0.60 to 2.75 µg mL$^{-1}$ (Table 5). These results are in agreement with those obtained by algal sulfolipids of *T. atomaria* showed the highest inhibition activity (85.37%) against HepG2 cells followed by *D. fasciola* (80.29%), *L. papillosa* (79.89%) and *U. fasciata* (72.97%). Therefore, all the algae sulfolipids showed potential antitumor activity against HEPG2 with IC$_{50}$ values ranging from 0.60 to 2.75 µg mL$^{-1}$ (Table 5). These results are in agreement with those obtained by
Bergé et al. (2002), who found that the sulfolipid compounds extracted from red algal Porphyridium possessed an inhibition effect against MCF-7. In another study carried out by Shao et al. (2002), they found that the sulfolipid compounds ((2R)-1-O-myristoyl-2-O-palmitoyl-3-O-(6-sulpho-a-D-quinovo-pyranosyl)-glycerol) from the brown algae Chondria crassicaulis possessed an inhibition effect against HL60 and MCF-7 cell lines. Bhaskar et al. (2004), reported that the total lipid and lipid classes of brown algae S. marginatum have potent inhibition of human pro-melocytic leukemia HL60. Also Hossain et al., (2005) reported that the SQDG of brown algae S. horneri showed significant apoptosis activity towards Caco-2 cells. The SQDG fraction obtained from dried spinach was an inhibitor of mammalian DNA polymerases and a growth inhibitor of NUGC3 human gastric cancer cells (Maeda et al., 2005). Sahara et al. (2002) and (Mizushina et al., 2003) found that the synthetic sulfoquinovosyl-mono-acylglycerols had a highly significant effect in suppressing the growth of solid tumors (human lung adenocarcinoma A-549 cells), and showed a potent inhibition of DNA polymerase and potent antineoplastic agents against the gastric cancer cell line (NUGC3).

Antimicrobial activity of algal sulfolipids. Marine algal sulfolipids (SLs) presented a high growth inhibition of the bacterial strains (B. subtilis and E. coli) at the concentration of 100 µg/well, while all algal SLs did not show any inhibition effect against fungal or yeast cells (Table 6). The highest bacterial growth inhibition was obtained by T. atomaria sulfolipids (15.0 mm) against E. coli followed by U. fasciata sulfolipids (13 mm) and G. cylindriea sulfolipids (11 mm). L. papillose

Table 4. Antitumor activity of algal sulfolipids against MCF7 and HepG2 cells after 48h incubation

| Algae species   | Sulfolipids Concentrations (µg mL⁻¹) | Growth Inhibition % |
|-----------------|--------------------------------------|---------------------|
|                 |                                      | MCF7 Cell | HepG2 Cell |
| U. fasciata     | 1.0                                   | 65.40a    | 48.15a    |
|                 | 2.5                                   | 69.16a    | 54.39a    |
|                 | 5.0                                   | 76.27ab   | 71.93a    |
|                 | 10.0                                  | 79.48a    | 72.97a    |
|                 | 1.0                                   | 79.46a    | 71.19a    |
|                 | 2.5                                   | 82.25a    | 76.15a    |
| T. atomaria     | 5.0                                   | 85.35a    | 76.70a    |
|                 | 10.0                                  | 85.69a    | 85.67a    |
|                 | 1.0                                   | 71.12a    | 57.36a    |
|                 | 2.5                                   | 73.33a    | 65.50a    |
| D. fasciola     | 5.0                                   | 74.43a    | 72.95a    |
|                 | 10.0                                  | 79.34a    | 80.29a    |
|                 | 1.0                                   | 76.93a    | 37.35a    |
|                 | 2.5                                   | 90.66a    | 44.85a    |
| L. papillose    | 5.0                                   | 92.48a    | 71.56a    |
|                 | 10.0                                  | 94.19a    | 79.89b    |
|                 | 1.0                                   | 66.37a    | 28.31a    |
| G. cylindriea   | 2.5                                   | 76.01b    | 42.21b    |
|                 | 5.0                                   | 77.30b    | 69.06b    |
|                 | 10.0                                  | 82.46b    | 72.27b    |
|                 | 1.0                                   | 42.73     | 26.1      |
| Novantron       | 2.5                                   | 52.81     | 42.27     |
| (reference drug)| 5.0                                   | 52.81     | 47.66     |
|                 | 10.0                                  | 52.81     | 59.50     |
| LSD             |                                      | 6.52      | 20.15     |

The mean (n = 3) difference is significant at P ≤ 0.01.
C_{43}H_{78}O_{12}S and the second compound (SQMG) was the consistent molecular ion [M + H] + at m/z = 556.71 corresponding to the molecular formula of C_{25}H_{48}O_{11}S. The algal sulfolipid fraction has antitumor activity against (HepG2 and MCF-7 cell lines) and antibacterial activity against Bacillus subtilis and Escherichia coli. The antitumor and antibacterial activities of algal sulfolipids are probably owing to the presence of the sulfolipids as sulfoquinovosyl-di-acylglycerol, sulfoquinovosyl acylglycerol, unsaturated fatty acids and sulfate contents.

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