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Evaluation of Biochemical composition and bioactivities of two *Ulva* species (*Ulva lactuca* and *Ulva fasciata*); a comparative study

Jelan Mofeed[1]*, Abd El Naser Sabry[2] and Mohamed Ali Deyab[2]

[1]Department of Aquatic Environment, Faculty of Fish Resources, Suez University, El-Salam city, Suez, **Egypt**.
[2]Department of Botany and Microbiology, Faculty of Science, Damietta University, New Damietta City, **Egypt.**
***Correspondence:** Jelanmofeed@hotmail.com **Received: 23-06-2019, Revised: 28-10-2019, Accepted: 29-10-2019 e-Published: 15-12-2019**

Seaweeds represent one of the most important reservoirs of new therapeutic compounds with valuable biological activities. In this study, the extracts of two *Ulva* species (*Ulva lactuca* and *Ulva fasciata*) were investigated to compare between their biochemical and antiviral activities against Coxsackie virus B3 (CVB3) and Rotavirus (RV), beside its biological activities against pathogenic bacteria: *Salmonella typhimurium*, *Staphylococcus aureus* and *Escherichia coli*. Both *Ulva* species showed high antiviral activity against Rotavirus more than Coxsackie virus B3, whereas crud extract of *Ulva fasciata* recorded high therapeutic index (35) than that of *Ulva lactuca* (17). Moreover, extracts of both *Ulva* species inhibited the bacterial growth but at different concentration. Where *Ulva lactuca* extract showed stronger antibacterial activity compared to that of *Ulva fasciata*, especially against *Staphylococcus aureus*, where 100% inhibition recorded with maximum concentration (100 $\mu.ml^{-1}$), while *Ulva fasciata* extracts showed only 70 % inhibition at the same concentration, reflecting differences in biological activities between the two *Ulva* species. The recorded results revealed that, *Ulva lactuca* and *Ulva fasciata* have valuable bioactive compounds, which should be applied in medicinal and pharmaceutical fields.

Keywords: *Ulva fasciata* -*Ulva lactuca* - biochemical - antiviral - antibacterial.

INTRODUCTION

Macro-algae considered as the dominant feature in most aquatic ecosystems. According to its pigmentation, there are three different phyla: Chlorophyta (green seaweeds), Phaeophyta (brown seaweeds) and Rhodophyta (red seaweeds), among them green seaweeds considered as the most diverse and widespread group (FAO, 2018). Since ancient times, it is well known that, traditional medicine of maritime nation have been used green seaweeds in treatment of hypertension, venereal diseases, cough, gout, goiter and even cancer, beside using as antibiotics in therapy of many infectious diseases (Nagarajan and Mathaiyan, 2015). Which indicate that, marine seaweeds are generous source with natural bioactive materials; therefore, more focus must be directed to use it as an available pharmaceutical agent (Pereira, 2016). During recent years, different marine organisms have been conventionally used as rich source of antimicrobial compounds (Abdel-Khaliq et al., 2014). Previously, Lipton and Jose (2006) reported that, broad range of biomedical activities such as antibacterial, anticancer, antioxidant and antiviral had obtained from green macro-algae; hence, it obtained significant importance as a new promising store for innovative bioactive materials that can be used in pharmaceutical production (Mofeed et al., 2018). *Ulva* is the common genus among green seaweeds, known as sea lettuce, and considered as a good source for functional

food beside using in pharmaceutical and agricultural applications (Isuru et al., 2011) due to the ability to produce various bioactive compounds (Satoru et al., 2003), which may inhibit the growth of many pathogenic gram positive and negative bacterial (Kolanjinathan et al., 2009).

Staphylococcus aureus is, the gram positive, high resistance bacteria that infects surface skin and deeper tissues, considered as major pathogen (Lowy, 1998). While, the gram-negative bacilli *Escherichia coli* usually colonize the large intestine and urinary tract cousin more than ninety percent of simple urinary tract infections (Madappa, 2014) especially in women (Gould, 2010). *Salmonella*, the facultative anaerobe bacterium, considered to be the causative agent of salmonellosis. The human pathogen, *Salmonella typhimurium* causes systemic infections and typhoid fever, which cause death to about 200,000 persons worldwide every year (Crump et al., 2004).

Coxsackie viruses B (CVB), a member in family Picornaviridae, which can cause both acute and chronic human diseases, fluctuated from nonspecific febrile, meningitis fever, pancreatitis, neurological disorders and even to cardiomyopathy severe neonatal diseases (Wang et al., 2018). CVB group comprises six serotypes (CVB1– CVB6). The hazardous human pathogen CVB3 can induces chronic and acute viral myocarditis in both adults and children, where it was responsible for 30 – 50 % of all myocarditis cases worldwide. In addition, it was reported that CVB3 cause other diseases such as aseptic meningitis and type “1” diabetes mellitus (Lee et al., 2014).

Rotavirus (RV), which belongs to family Reoviridae, is a major pathogen causing diarrhea in babies and young children (Parashar et al., 2003 and Gray et al., 2008), which consequently infect and even cause death for more than 450,000 children every year (WHO, 2013).Until now, there are no approved vaccines to control CVB or RV induced infections (Kim et al., 2012; Ge et al., 2014; Wang et al., 2018 and Soukaina et al., 2018). On the other hand, natural compounds derived from herbal medicine and seaweeds are safe, cheap and effective alternatives used to prevent and treat the infections diseases. Nevertheless, exploitation of macro algae as a source for novel bioactive substances is still in an early stage and needs further studies.

In the present investigation, biochemical component, of two green marine algae (*Ulva fasciata* and *Ulva lactuca*) were studied and crude extracts of both the selected *Ulva* species were tested for antimicrobial activities against three pathogenic bacteria (*Escherichia coli, Salmonella typhimurium* and *Staphylococcus aureus*) and also against two enteric viruses (Coxsackie viruses B and Rotavirus).

MATERIALS AND METHODS

2.1 Collection of algal samples:

Ulva fasciata Delile was collected from the Mediterranean Sea at Alexandria Governorate (Abu-Qir N 31° 19` E 030° 03`) Egypt, while *Ulva lactuca* Linnaeus collected from Damietta Governorate (Ras-Elbar City N 31°30′45″ E 31°49′32″) Egypt, during October 2016 (Fig. 1). The algal samples were washed with distilled water after several times of washing by tap water, then dried in shade at room temperature. One hundred gram of grounded samples were stored in brown glass containers at room temperature for further analysis.

2.2 Extraction methods:

Only, 25g of the dried coarsely seaweeds were powdered and packed in soxhlet extractor. The solvent mixture (1methanol:1 hexan) was added into the flask and heated to extract active constituents from the algal biomass, the process was repeated until complete extraction. By using rotary evaporator (at 30 – 45 ° C), the extract was concentrated after cooling, then stored at 4°C until use.

2.3 Biochemical analysis:

Biochemical analysis for the two tested algal species were evaluated. Moisture content had been determined gravimetric until obtained constant weight. To estimate ash content, the dried samples were muffled overnight at 525°C. Protein (% of dry weight) was determined according to AOAC (1995). Total carbohydrate was obtained by following the phenol sulfuric acid method of Dubois et al. (1956). However, lipid contents were evaluated according to AOAC (2000) by using chloroform-methanol mixture (2:1 v/v) for extraction.

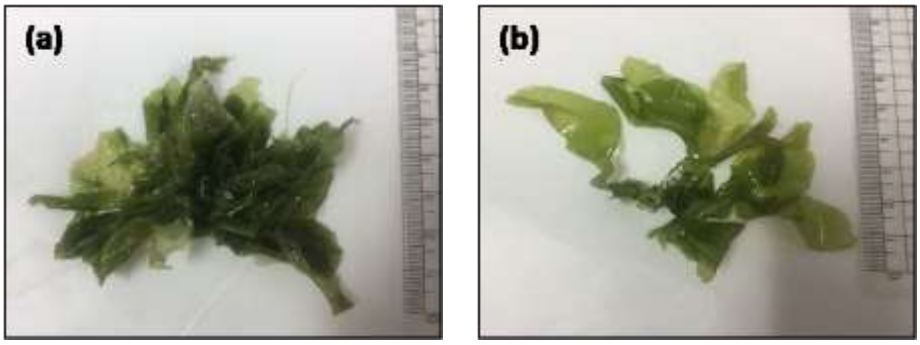


Figure1: The two *Ulva* species: (a) *Ulva lactuca*and (b) *Ulva fasciata*.

2.4 Antibacterial activity:

The antibacterial activities of the two *Ulva* extracts against three different human pathogenic bacterial strains namely; *E. coli* ATCC 8739, *Salmonella typhimurium* ATCC 14028 and *Staphylococcus aureus* ATCC 6538 were tested. Bacterial strains (24 h aged) were inoculated in Tryptic Soya Broth (TSB) media and incubated at 37°C for 24 hours, then 200 µl of each bacterial strain was cultured using pour plate technique in freshly prepared sterile Tryptic Soy agar (TSA) and were treated with algal extracts using 4 different concentration (12.5, 25, 50, 100 µg.ml^{-1}).Then at 37 °C and for 24 hours the samples were incubated (NCCLS, 2003). Bacterial inhibition percent was calculated by following formula:

% of Bacterial Inhibition = $(C_c-C_s/C_c) \times 100$

Where: C_c= control count and C_s= count at different concentration

2.4.1 Scanning Electron microscope:

The preparation of cultured bacterial cells for scanning electron microscope examination was according to Erdos (1986) method. The tested bacteria were inoculated in nutrient agar media containing the algal extract at final concentration (100 µg.ml^{-1}), after 24 hours of incubation, the bacteria were fixed with formalin-glutaraldehyde fixative (4FIG) in 0.1M phosphate buffer (pH 7.4). In the same buffer, post-fixation of the specimens was by 1% Osmium tetroxide, and then dehydrated in a graded series of acetone. Finally, in a polaron E500 sputter coater, the specimens were coated with gold palladium and examined using Scanning Electron Microscope (JEOL JSM 35C).

2.5 Antiviral activities:

2.5.1 Cell lines and Virus titration:

Rhesus monkey kidney cell line (MA 104) and African green monkey kidney (Vero) were used to be infected by simian rotavirus SA-11 strains and coxsackie virus B3, respectively. MA 104 and Vero cells were cultivated in Dulbecco's Modified Eagle Medium (DMEM) and Eagle's minimum essential medium (EMEM), respectively. The used culture media contained only 2% fetal bovine serum (FBS) for cytotoxicity and antiviral assays. Activated simian rotavirus (SA-11) with trypsin (10 mg.ml^{-1} trypsin for 30 min at 37^{o}C) and coxsackie virus B3 stocks were titrated in 96-well plates using MA 104 and Vero cells, as illustrated previously by Shaheen et al., (2015).The viral titers were calculated as $TCID_{50}$. 0.1 ml^{-1} (50% tissue culture infectious doses. 0.1 ml^{-1}) according to Spearman Kärber formula (Finney, 1978). Each virus stock was stored separately at -80°C until used.

2.5.2 Cytotoxicity assay:

Different concentrations (7.8, 15.6, 31.25, 62.5, 125, 250, 500, and 1000 µg.ml^{-1}) from extracts of both the tested *Ulva* species were added to EMEM and DMEM containing 2% FBS and 2% antibiotics. The algal extracts' cytotoxic activity was determined onto MA 104 and Vero by using MTT method (Nabil et al., 2012). The cytotoxic percentage was calculated as:

% of Cytotoxic = (X - Y / Z) x 100

Where, both X and Y refer to the optical densities of the control and treated cells, respectively. The 50% cytotoxic concentration (CC_{50}) is the concentration of the algal extracts (µg.ml^{-1}) that can reduce fifty percent of viability, compared with control.

2.5.3 Antiviral activity of algal extracts on CVB3 and RV SA-11 by MTT method:

In 96-well microtiter plates, MA 104 and Vero (5 x 104 & 5 x 103 cells/well, respectively) cell lines were grown at 37 ^{o}C for 24 hours in CO_2 incubator. Then after removing the culture media, three non-toxic concentrations from both *Ulva* species extracts were selected to be tested against viral infections. The percentage of protection was estimated as:

% of Protection = [(X-Y)/ (Z-Y) x 100]

Where X, Y and Z were the values of absorbance (OD_{540}) of the two *Ulva* extracts with virus infected cells, virus and controls. Fifty percent inhibitory concentration (IC_{50}) is the concentration that protects fifty percent of treated infected cells, compared to control. To determine the antiviral activity, therapeutic index (TI) of the *Ulva* extracts was determined by calculating the ratio CC_{50} over IC_{50}.

RESULTS AND DISCUSSION

3.1 Biochemical analysis:

Biochemical composition (Crude fiber, ash, protein, carbohydrate and lipid) of the two *Ulva* species illustrated in Table 1. It is noticeable that; the results showed significant differences in ash content between the two *Ulva* species (34.15 ±0.98 and 21.08 ±0.75 % for *U. fasciata* and *U. lactuca* respectively), the result which agrees with that observed by Ismail and Mohamed (2017). Wong and Cheung (2001) indicated that, the ash content of *U. lactuca* ranges between 21.3–24.6 %/ DW while, Mc-Dermid and Stuercke (2003) demonstrated that, the ash content in case of *U. fasciata* was 25.4 % per dry weight.

Anent the results of total protein reveals that; the two *Ulva* species had low significant difference, where the protein content of *U. lactuca* was slightly more than that of *U. fasciata*, (21.38 ± 0.76 and 17.35 ±0.91 %, respectively). The same trend of results were demonstrated by Dhargalkar et al., (1980) who mentioned that, among different genera and even different species of the same genus, protein contents were distinctly varied. In this connection, Kokilam and Vasuki (2014) recorded that, *Ulva* species. had a protein content within a range of 10–20 % per dry weight (DW).

Total carbohydrate showed low significant differences between the two *Ulva* species (35.48 ± 0.98 %, 30.82 ± 0.55 %, for *U. lactuca* and *U. fasciata,* respectively). The results which inconsistence with Ismail and Mohamed (2017) who recorded that, the ratio of carbohydrate in *Ulva* species fluctuated from 29.5 to 46.6 % per dry weight, while Khairy and El-Shafay (2013) mentioned that, per dry weight, the content of carbohydrate of *U. lactuca* ranged from 42.1 to 46.5%. A glance on Table 1 reveled that, the total crude fat had significant differences between the two *Ulva* species, where it gave 11.6 ±0.34 % in case of *U. lactuca* and 2.4 ±0.25 % in case of *U. fasciata*. These results were nearly in consistence with that reported by Kaliaperumal et al., (1995) who found that, *U. lactuca* contains 7.4 % lipid per dry weight. Regarding the crude fiber content, the results showed no significant differences between the two tested species.

3.2 Antibacterial activities:

Antibacterial activities of the two *Ulva* extracts against *Escherichia coli* ATCC 8739, *Staphylococcus aurous* ATCC 6538 and *Salmonella typhimurium* ATCC 14028 were determined by using pour plate technique method and the results summarized in Table 2 and Fig. 2.

Ulva lactuca extract showed more inhibition activities than *Ulva fasciata* against the gram negative bacteria; *Escherichia coli* ATCC 8739 at different concentrations. The inhibition activities of *Ulva lactuca* reached 91.4 and 85.7 % within concentrations of 100 and 50 µg.ml^{-1} respectively, while it reached 62.9 and 40 % in case of *Ulva fasciata* at the same concentrations.

Table 1: The mean percentage (± SD) contents of moisture, ash, protein, carbohydrate and lipid per dry weight in both *Ulva lactuca* and *Ulva faciata*.

Seaweeds	Content % Dry weight (±SD*)				
	Total lipids	Total protein	Crude fiber	Carbohydrates	Ash content
Ulva lactuca	**11.6 (± 0.34)**	**21.38 (± 0.76)**	**3.4 (± 0.36)**	**35.48 (± 0.98)**	**21.08 (± 0.75)**
Ulva fasciata	**2.4 (± 0.25)**	**17.35 (± 0.91)**	**3.27 (± 0.44)**	**30.82 (± 0.55)**	**34.15 (± 0.98)**

Anent the inhibition activity against the gram positive bacteria *Staphylococcus aurous* ATCC 6538,*Ulva lactuca* showed 100 % inhibition at concentration 100 µg.ml^{-1}, while it gave 95 % inhibition at 50 µg.ml^{-1}and 72.5 % at concentration 12.5 µg.ml^{-1}. On the other side, *Ulva fasciata* extract showed 70 % inhibition at concentration 100 µg.ml^{-1}, whereas it recorded 47.5 and 25% inhibition at concentrations 50 and 12.5 µg.ml^{-1}respectively.

Again, *Ulva lactuca* extract showed 90% inhibition to the gram negative bacteria *Salmonella typhimurium* ATCC 14028 at concentration 100 µg.ml^{-1}, while it gave 60% inhibition at the lowest concentration (12.5 µg.ml^{-1}). Meanwhile, *Ulva fasciata* extract gave its maximum inhibition (70 %) at concentration 100 µg.ml^{-1}.

It is worth mentioning that, an obvious differences between the antibacterial activities of *Ulva lactuca* and *Ulva fasciata* extracts were observed, where *Ulva lactuca* extract have more antibacterial activities than *Ulva fasciata* extract against the three bacterial species (*Escherichia coli*, *Staphylococcus aurous* and *Salmonella typhimurium*).

The above results were consistent with that recorded by Abdel-Khaliq et al., (2014), who studied the antimicrobial activities of some Egyptian seaweeds collected also from the Mediterranean Sea, and his results indicated that the species of seaweeds showed variety of antimicrobial activities, giving seaweeds its importance as natural products. Earlier studies made by Saritha et al., (2013) showed that, *Ulva lactuca* extract has a broad spectrum of antibacterial activity. Also Pramitha and Lipton (2014) investigate that, *U. fasciata* extract can inhibit 66% of pathogenic *Staphylococcus aurous*. In this context, both Prasanna et al., (2016) and Liela (2017) reported that, the methanolic extract of *U. lactuca* had the ability to inhibit a wide range of clinically relevant *Staphylococcus* strains. On other study for the methanolic extracts of both *U. intestinalis* and *U. rigida* collected from Western Algeria clarify their antimicrobial activities against: *Escherichia coli*, *Salmonella*, *Shigelladysentriae*, *Pseudomonas aeruginosa* (Sahnouni et al., 2016). However, the ethanolic extract of *U. rigida* collected from Vona Bay Perşembe, Ordu (Turkey), exhibited inhibition activities against *Staphylococcus aureus, Salmonella typhimurium, Pseudomonas aeruginosa, Escherichia coli,* and *Bacillus cereus* (Erturk and Tas, 2011).

3.3 Scanning Electron Microscope (SEM) of Bacterial cells:

Escherichia coli, Staphylococcus aurous and *Salmonella typhimurium* were treated by both of *U. fasciata* and *U. lactuca extract*s (at the concentration 100 µg.ml^{-1}) to examine the cytomorphology by SEM, which provided strong evidence that, the tested extracts were stressful for the bacterial populations. Where the cytomorphology of normal *E. coli* cells (control) were apparently normal rods with smooth surface and normal division (Figure 3) with normal length 1.03 to 1.19µm and width ranged 0.42 to 0.53 µm. While, the cytomorphology of treated *E. coli* cells as examined by SEM revealed that the cells altered their morphology with respect to 100 µg.ml^{-1}concentrations of both *Ulva* extracts. It was enlarged (up to 0.56 µm in width and 1.80 µm in length), malformed and appeared with pressed shape cell. A more or less the same scenario was obtained in case of *Staphylococcus aurous*, where the cytomorphology of normal cell (control) were apparently normal round shape bacterium with smooth surface and normal diameter ranged from 0.32 to 0.36 µm, while, the cells reflected malformation and altering in their morphology beside the enlargement in cell diameter (up to 0.43 µm). Similarly *Salmonella typhimurium* cells were affected by treating with both *Ulva* extract, where its cells have been deformed. Audit in Figure 3 reflected that, the abnormalities in size and all forms of malformations which observed with the treatment by *U. lactuca* extract were more tragically than that obtained in case of treatment by *U. fasciata*.

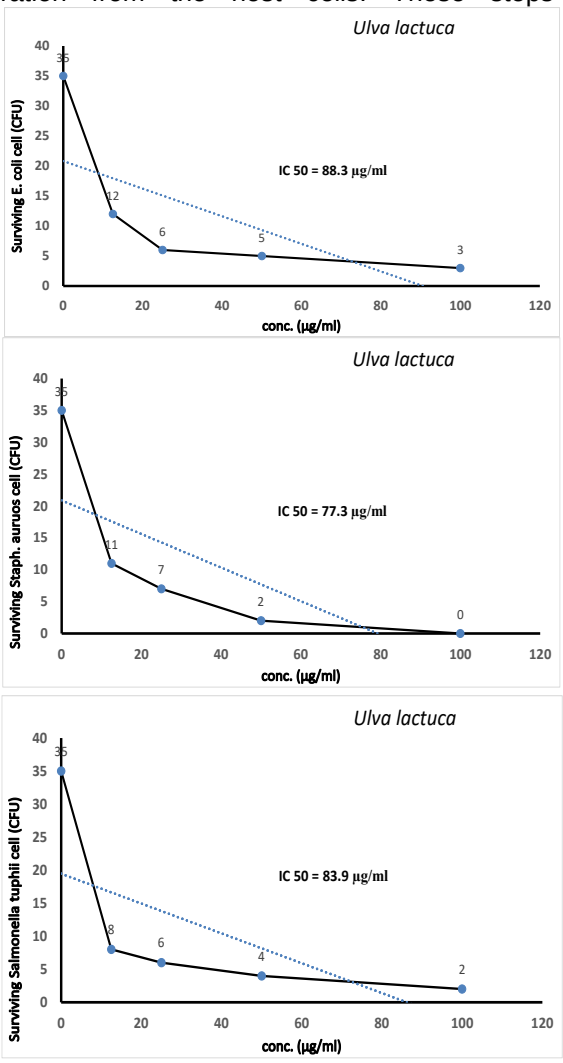
3.4 Antiviral activities:

3.4.1 Cytotoxicity of *Ulva* extracts on Vero and MA 104 cells.

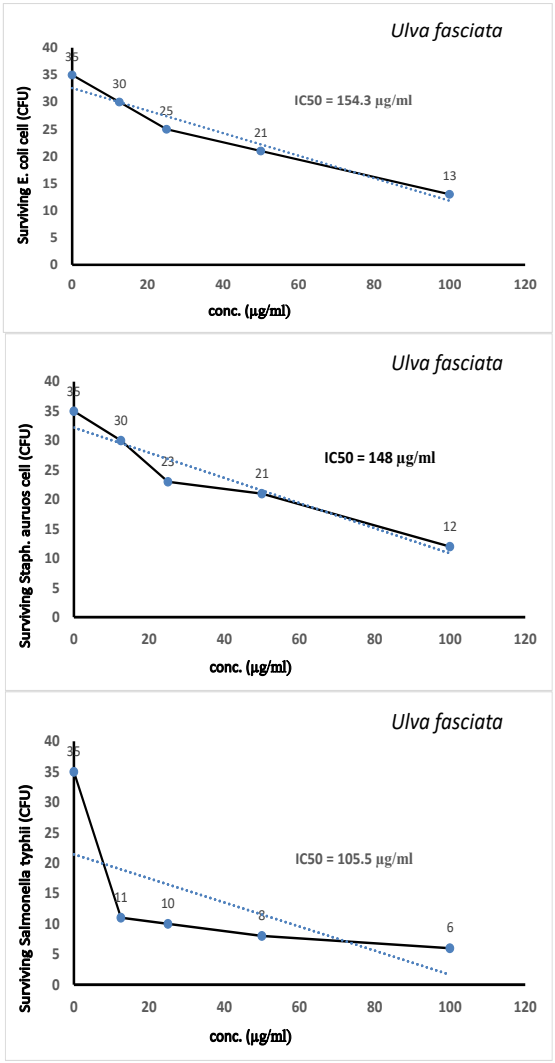
The cytotoxic effects of the two *Ulva* extracts were examined on MA 104 and Vero cells using colorimetric MTT assay. CC_{50} value for the tested extracts after 48 h of incubation with MA 104 and Vero cells were calculated. Inspection of Table 3 reveled that, both extracts exhibited cytotoxicity against Vero cells with CC_{50} 1200 ±10.4 µg.ml^{-1} in case of *U. fasciata* and 1500 ±9.9 µg.ml^{-1} with *U. lactuca*. Furthermore, the two *Ulva* extracts had less CC_{50} on MA 104 cells for *U. lactuca* and *U. fasciata*, (980 and 820 µg.ml^{-1} respectively). In accordance with the cited results, Santibanez et al., (2018) find that, CC_{50} of *Ulva intestinalis* was more than 1500 µg.ml^{-1}.

3.4.2 The Antiviral activity of the algal extracts

Viral infection cycle includes different steps including; viral attachment and penetration followed by replication of viral genetic materials and proteins, assembly and completed by viral liberation from the host cells. These steps represented the targets point for anti-coxsackie virus B3 and anti-rotavirus SA-11 agents.



U. lactuca extracts.



U. fasciata extracts.

Figure 2: Number of surviving cells of Escherichia coli, Staphylococcus aurous and Salmonella typhimurium after being treated with *Ulva lactuca* and *Ulva* fasciata extracts.

Table 2: Inhibition percentage of *U. lactuca* and *U. fasciata* against *Escherichia coli* ATCC 8739, *Staphylococcus aurous* ATCC 6538 and *Salmonella typhimurium* ATCC 14028.

Conc. µg.ml^{-1}	*Escherichia. coli*		*Staphylococcus aurous*		*Salmonella typhimurium*	
	U. lactuca	*U. fasciata*	*U. lactuca*	*U. fasciata*	*U. lactuca*	*U. fasciata*
12.5	65.7	14.3	72.5	25	60.0	45.0
25	82.9	28.5	82.5	42.5	70.0	50.0
50	85.7	40.0	95	47.5	80.0	60.0
100	91.4	62.9	100	70	90.0	70.0

Table 3: The cytotoxicity of algal extracts on Vero and MA104 cells.

Algae extracts	CC_{50} (µg.ml^{-1})[a]	
	Vero Cells	MA 104 Cells
Ulva lactuca	1150 ± 9.9	980 ± 10.7
Ulva fasciata	1200 ± 10.4	820 ± 11.2

(a): Concentration of extract that is cytotoxic to 50% of cells

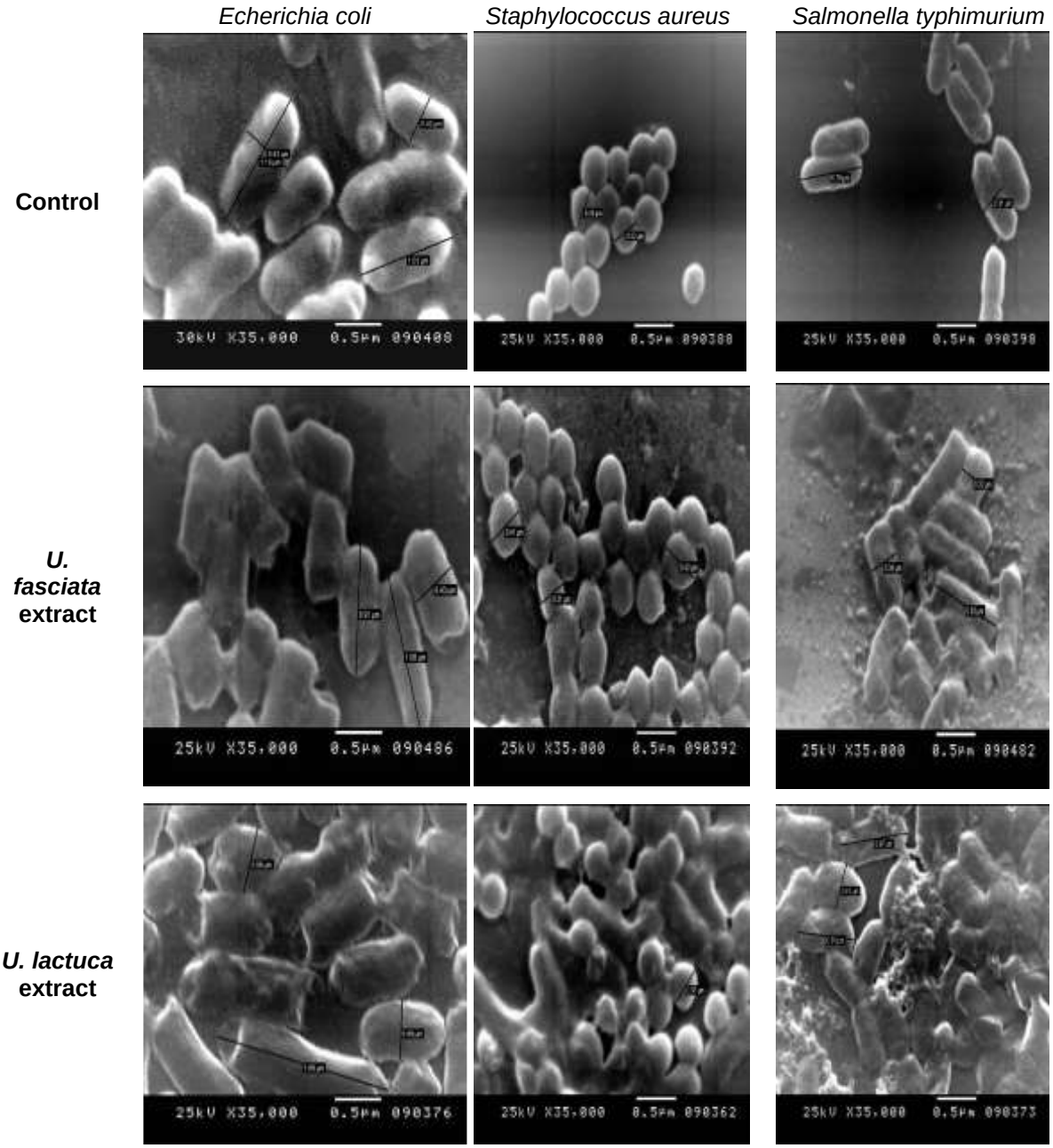


Figure 3; SEM cell morphology observation of the three test microorganisms (*Escherichia coli*, *Staphylococcus aurous* and *Salmonella typhimurium*) before (control) and after treatmentment with both *Ulva fasciata* and *Ulva lactuca* extracts.

Table 4: The antiviral activity of algal extracts against CVB3 and RV SA-11.

Algae extracts	RV			CVB3		
	IC_{50} ($\mu g.ml^{-1}$)[a]	TI	R	IC_{50} ($\mu g.ml^{-1}$)[a]	TI	R
Ulva lactuca	57.6	17	4.5	100	12	1
Ulva faciata	23	35.7	2.5	383.3	3	2.25

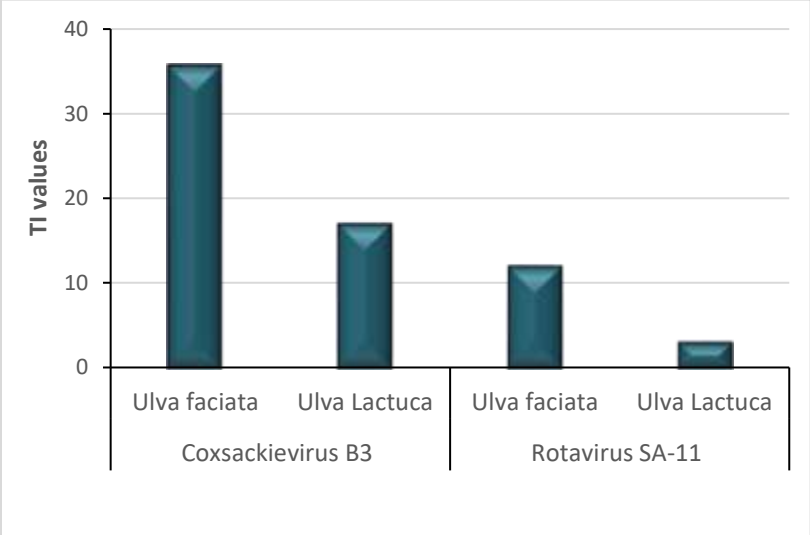


Figure 4; The therapeutic index of algal extracts against CVB3 and RV.

This study focused on, effects of the two tested extracts on both attachment and penetration steps. Where, therapeutic index (TI) of the two tested *Ulva* species for the antiviral activities were obtained by calculating the ratio of CC_{50} over IC_{50} and the reduction in virus titer (R) was calculated as the difference between treated and untreated virus and expressed in log10 $TCID_{50}$/0.1 ml. The result illustrated in Table 4 clarify that, *Ulva fasciata* extract showed maximum antiviral activity against RV with therapeutic index (TI) of 35.7 with reduction of virus titer 2.5 log 10 $TCID_{50}$ / 0.1ml, while the minimum therapeutic index (17) was recorded within *Ulva lactuca* extract with 4.5 log 10 $TCID_{50}$ / 0.1ml reduction of virus titer. While in case of CVB3, *Ulva lactuca* extract displayed higher therapeutic index (12) than *Ulva fasciata* (3) with reduction of virus titer fluctuated from 1.0 to 2.25 log 10 $TCID_{50}$ / 0.1ml, respectively (Figure 4).

The cited results revealed that, *Ulva* extracts have the ability to bind with viral capsids, preventing them to attach to cell receptors and thereby prevented their penetrations into host cells. Where, IC_{50} is the concentration which can inhibit 50% of the viral infectivity (Cytopathic Effect). *Ulva lactuca* extract was records 100 and 57.5 $\mu g.ml^{-1}$, meanwhile the *Ulva fasciata* extract was 383.3 and 23 $\mu g.ml^{-1}$ for coxackie virus and Rotavirus, respectively (Table 4).

In a previous study Soares et al., (2012) demonstrated that, seaweeds extract have antiviral activities against the herpes simplex virus (HSV) and reported that, the green algae *Ulva fasciata* showed high antiviral activity (99.9%) against HSV-1 due to the existence of high polysaccharides and fatty acids concentrations. In accordance with that, Shi et al., (2017) described that, the secondary algae metabolites has been tested as antiviral agents for many enveloped viruses of medical and veterinary importance, not only as prophylactic strategy before viral infection but also as a successful treatment after infection to avoiding virus dissemination (Santibanez et al., 2018). Moreover Abd-Elbaky et al., (2014) noted that, the phospholipids content of *U. fasciata* have a maximum inhibition effect against HSV1 (75.25%) at concentration 20 $\mu g.ml^{-1}$.Generally many studies mentioned that, the observed antiviral activities of marine macro algae may be due to the presence of phospholipid content

(Dumay et al., 2006).

It is of interest to mention that, despite the risk of infection with CVB3-induced, there are no approved antiviral agents or vaccines to treat and/or prevent its chronic or even acute infections (Fechner et al., 2011; Ge et al., 2014 and Wang et al., 2018). Similarly, there is no available effective medication to control diarrhea caused by RV infection (Kim et al., 2012). Alternatively, an effective, safe and cheap active metabolites from seaweeds are good candidates as therapeutic agents against CVB3 and RV infections (Shaheen et al., 2015). In this study, the antiviral activities of *U. lactuca* and *U. fasciata* were tested against CVB3 and RV infection. Although there are many previous studies on the application of different seaweeds for antiviral activities, but the studies on coxsackie virus and rotavirus are very rarely and the present investigation considered as one of the earliest studies which interested with using off two Egyptian marine *Ulva* species as anti-coxsackie virus B3 and anti-rotavirus SA-

CONCLUSION

The recorded results clarified the presence of an obvious difference between the two *Ulva* species (*Ulva lactuca* and *Ulva fasciata*) in their biochemical components and bioactivities, in accordance. It was concluded that, the two *Ulva* species possess various antibacterial activity against hazardous pathogenic bacteria; *Staphylococcus aureus, Escherichia coli* and *Salmonella typhimurium*, in addition to an efficient antiviral activity against Coxsackie B and Rotaviruses. Based on the results in this study, *Ulva lactuca* and *Ulva fasciata* have valuable bioactive compounds, which should be applied for production of natural antibiotics, antivirus and novel drugs. Furthermore, detailed studies on purification, quantification and evaluation of such compounds can take this to a wide scale application in the pharmaceutical industries.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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This article does not contain any studies with animals performed by any of the authors.

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