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Research Article

Antibacterial Potential of Gulf of Mannar Seaweeds Extracts Against Two Plant Pathogenic Bacteria *Xanthomonas axonopodis* pv. *citri* (Hasse) Vauterin et al. and *Xanthomonas campestris* pv.*malvacearum* (Smith 1901) Dye 1978b

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ABSTRACT

Antibacterial potential of 23 red, 9 brown and 15 green seaweeds collected along the coast of Pampan (Rameswaram), Gulf of Mannar, Tamilnadu, India was evaluated against two plant pathogenic bacteria such as *Xanthomonas axonopodis* pv. *citri* and *X. campestris* pv. *malvacearum* causing canker in citrus and angular spot in cotton, respectively *in vitro*. Among the solvents and methods used for the extraction of antibacterial substances from shade dried specimens of seaweeds, a maximum antibacterial activity was found in the crude extracts obtained in 1:1(v/v) chloroform: methanol followed by 2:1 (v/v) chloroform: methanol, chloroform, methanol, petroleum ether, ethanol and diethyl ether. Efficacy of crude extracts was higher against *X. axonopodies* pv. *citri* than against *X.campestris* pv. *malvacearum*. This investigation shows that seaweeds occurring along the coast of Gulf of Mannar, Tamilnadu, India could be a promising source of bioactive compounds for the inhibition of *Xanthomonas axonopodis* pv. *citri* and *X. campestris* pv. *malvacearum in vitro*.

Keywords: Antibacterial activity; Seaweeds; Pampan; Tamil Nadu.

INTRODUCTION

Since the marine environment is the abode of many groups of microorganisms¹, seaweeds are constantly in contact with these potentially dangerous microbes and they have apparently evolved with chemical defense strategies by synthesizing an array of secondary metabolites to defend against the microbial thread². Thus, seaweeds contain a rich variety of bioactive natural products that exhibit biomedical and antimicrobial properties^{2,3}. Harder⁴ was the first to observe antimicrobial substances in algae. However, it was not until the 1970s that large-scale screening of antimicrobial activity was carried out⁵⁻⁷. Therefore, in the last few decades, numerous reports of macroalgae derived compounds showing various biological activities such as antiviral, antibiotic, anti-neoplastic, antifouling, anti-inflammatory, cytotoxic and anti-mitotic have been published⁸⁻¹¹. Intensive application of synthetic pesticides in agriculture caused damage to the ecological state of the agricultural system¹². Pesticides of biological origin are generally less toxic affect only the target pest and closely related organisms and are,

effective in very small quantities which decompose quickly, therefore resulting in lower exposures and largely avoiding the pollution problems caused by conventional pesticides. Published literature reports on the diverse bioactivities of seaweeds, but the antibacterial efficacy of seaweeds against plant pathogens are comparatively a new concept and a few attempts have been made in this regard^{3,13,14} Therefore in the present study, seaweeds occurring along the coast of Pampan(Rameswaram), Gulf of Mannar, Tamilnadu, India were evaluated against two plant pathogenic bacteria such as Xanthomonas axonopodis pv. citri and X. campestris pv. malvacearum causing canker in citrus and angular spot in cotton, respectively. Up to 60 % yield loss was estimated in these crops as result of infections¹⁵.

MATERIALS AND METHODS

Seaweeds collection site: The Gulf of Mannar comprises of 20 islands located in a chain between Tuticorin (8°48`N; 78°9`E) and Rameswaram (9°14`N; 79°14`E) on the South East coast of India. Species of economically important seaweeds found

abundantly along the coast of Pampan (Rameswaram), Gulf of Mannar were collected during the year 2009 and screened for antibacterial potential.

Collection of seaweeds: A portion of live, healthy, matured and disease free seaweeds (23-red, 15green and 9-brown) occurring along the coast of Pamban (Rameswaram), Gulf of Mannar, Tamilnadu, India collected during the postmonsoon season (November 2009) in spring tide was washed thoroughly in seawater followed by tap water to remove the extraneous materials and sand particles .The algae were immediately air-dried under shade at room temperature for 3 days. Seaweeds were collected during post monsoon season because this season maximum antibacterial activity are found in seaweeds followed by those collected during monsoon, pre-monsoon and the summer season¹⁶. The plant pathogenic bacteria Xanthomnas axonopodis pv. citri and X. campestris pv.malvacearum causing canker in citrus and angular spot in cotton, respectively used in the present study were obtained from the cultures available in our botany department.

Selection of suitable solvents /methods for extraction of antibacterial substances: Five hundred grams of shade dried samples of each alga were chopped and pulverized. The pulverized sample of 50 g of red alga Gracillaria edulis, green alga Ulva compressa and brown alga Sargassum wightii was separately extracted at least thrice for 7 days in 100 ml of different solvents such as diethyl ether, petroleum ether, acetone, chloroform, methanol, ethanol, chloroform: methanol(2:1 v/v) and chloroform: methanol(1:1 v/v) using 250 ml Erlenmeyer conical flasks under dark conditions until the extract became colourless in order to select a suitable solvent/and method to extract the antibacterial substances. Then the extracts were combined keeping each extraction type separate and concentrated using Rotavapor under reduced pressure at 45° C and weighed stored at 0° C until testing was conducted.

Evaluation of antibacterial potential in seaweeds: Based on the preliminary investigation, crude extracts obtained in chloroform: methanol (2:1 v/v) and chloroform: methanol (1:1 v/v) of shade dried algae which showed maximum antibacterial activities were used for further evaluation. Powder weighing 50 g of each shade dried specimen of 23 red seaweeds(Table 2), 15 green seaweeds (Table 3) and 9 brown seaweeds (Table 4) collected along the coast of Pampan (Rameswaram) was soaked in 250 ml Erlenmeyer conical flasks containing 100 ml of chloroform: methanol (2:1 v/v) and chloroform: methanol(1:1 v/v) separately under dark condition. The extraction was repeated thrice for all the samples. The extracts were combined and concentrated using Rotavapour under reduced pressure at 45° C. The concentrated extracts were weighed stored at 0° C till antibacterial assay was conducted.

Antibacterial assay through agar diffusion technique¹⁶: The antibacterial assay was carried out using the agar diffusion technique with 5.0 mm diameter Whatman #1 paper discs. The assay was carried out on 1.5 % nutrient agar medium. The nutrient agar medium consisted of Peptone - 10 g, Beef extract -10 g, Na Cl -5 g, Distilled H₂O -1000 ml, Agar powder - 15 g, pH -7.0. Sterile paper discs were loaded with 50 µl (100 µg of the in mass) of different crude extracts using micropipettes and were allowed to dry thoroughly under aseptic conditions. The discs were impregnated onto Petri plates of 100 mm diameter containing ca. 20 ml of nutrient agar medium smeared with 0.05 ml of bacterial culture in exponential phase of 1.0 OD at 590 nm and incubated at 28°C for 48 hours. The diameters of the agar clear zones of bacterial inhibition around the discs as a result of diffusion of active substances were measured millimeters as a measure of antibacterial activity. Three replicates were maintained for each experiment and the mean values expressed. The solvents used for reconstituting the crude extracts were loaded on paper discs were treated as a solvent control and did not show any inhibition zones. Efficacy of antibacterial activity of algal extracts were compared with inhibition zones obtained from 10 µg disc-1 of streptomycin sulphate as standard control against both test bacteria. Experiments were conducted in triplicates and the mean and standard deviation were analyzed using SPSS-14.

RESULTS

Efficacy of suitable solvents/methods for extraction of antibacterial property: In the present study, solvents/methods such as diethyl ether, petroleum ether, chloroform, methanol, chloroform: methanol(2:1v/v) and chloroform :methanol (1:1v/v) were used for the extraction of antibacterial substances in the red alga Gracilaria edulis, green alga Ulva (Enteromorpha) compressa and brown alga Sargassum wightii in order to select suitable solvents or methods to extract the maximum antibacterial substances from the seaweeds to test against bacteria Xanthomonas axonopodies pv citri and X. campestris pv malvacearum (Table 1). Overall, the antibacterial activity in the crude extracts of seaweeds was more against X. axonopodies pv citri than against X. campestris pv malvacearum. Among the solvents used for the extraction of antibacterial activity, maximum antibacterial activity was found in the crude extracts obtained in 1:1(v/v) chloroform:

methanol followed by 2:1(v/v) chloroform: methanol, chloroform, methanol, petroleum ether, ethanol and diethyl ether. Of the solvents and methods used for obtaining the crude extracts in three seaweeds, maximum antibacterial activity was recorded from the brown alga Sargassum wightii followed by green Ulva compressa and red alga Gracilaria edulis in 1:1(v/v) chloroform : methanol extract against the test bacterium Xanthomonas axonopodies pv citri. Furthermore against X. campestris pv malvacearum, the same trend was followed with moderate activity. The crude extracts obtained in diethyl ether of Gracilaria edulis and Sargassum wightii, extracts obtained in petroleum ether of Gracilaria edulis and extracts obtained in chloroform, methanol and ethanol of Sargassum wightii and Gracilaria edulis showed only trace activity against X. campestris pv malvacearum (Table 1).

Evaluation of antibacterial potential of seaweeds: Antibacterial activity of streptomycin sulphate against both test bacteria was more than the crude extracts of algae (Table 2). Based on the preliminary studies, in the present study, extracts obtained in 2:1(y/y) and 1:1(y/y) chloroform : methanol were chosen as promising for evaluation of antibacterial potential in seaweeds against two plant pathogenic bacteria. Crude extracts obtained in 2:1 (v/v) and 1:1 (v/v) chloroform: methanol from 23 red, 15 green and 9 brown seaweeds were tested for antibacterial activity and the zones of inhibition was greater against X. axonopodies pv citri than against Х. campestris pv malvacearum(Table 2,3 and 4).

Red seaweeds: Extracts exhibited more zones of inhibition against X. axonopodies pv citri than against X. campestris pv malvacearum. A high antibacterial activity was recorded from the crude extracts obtained in 1:1 (v/v) chloroform: methanol of Portieria hornemanni against Xanthomonas axonopodis pv citri which was the highest antibacterial activities recorded among the crude extracts of the 23 red seaweeds investigated whereas the same crude exhibited comparatively low zone of inhibition against X. campestris py malvacearum. The 2:1 (v/v) chloroform: methanol extracts of *Portieria hornemanni* showed optimum antibacterial activity against test bacteria. Crude extracts obtained in 1:1 (v/v) and 2:1 (v/v) chloroform: methanol of Gelidiella acerosa, Amphiroa fragilissima, G. corticata var. cylindrica, G.edulis, Hypnea musciformis, G. crassa, H.valentiae, Champia parvula and Acanthophora spicifera exhibited moderate antibacterial activity against both the bacteria tested. The crude extracts obtained in 2:1 (v/v) and 1:1 (v/v) chloroform: methanol of Gelidiopsis variabilis, Jania rubens, Sarconema furcellatum and Laurencia obtusa and crude extracts obtained in 2:1 (v/v) chloroform:

methanol of *Grateloupia lithophila*, *Halymenia floresia*, *Gracilaria corticata* var.*corticata*, *G*. *foliifera* did not show any antibacterial activity against both the test bacteria(Table 2).

Green seaweeds: High antibacterial activity was recorded from the crude extracts obtained in 1:1 (v/v) and 2:1 (v/v) chloroform: methanol of Ulva compressa against Xanthomonas axonopodis pv citri, whereas against X. campestris pv zone of inhibitions malvacearum, were comparatively low. Optimum antibacterial activity was recorded from the crude extracts of Halimeda macroloba. Moderate antibacterial activity was recorded in the crude extracts obtained from the green seaweeds Cladophora fasicularis. Caulerpa racemosa, C. scalpelliformis, C.peltata, Bryopsis plumosa and Valoniopsis pachynema. The crude extracts obtained in 2:1 (v/v) and 1:1 (v/v)chloroform: methanol of Chaetomorpha linum, Cladophora colabens, Caulerpa chemnitzia, C. sertularoides, Halimeda gracibis and Boergsenia forbesii did not show any antibacterial activity against both the test bacteria (Table 3).

Brown seaweeds: Maximum antibacterial activity was recorded from the crude extracts obtained in 1:1(v/v) chloroform: methanol of *Dictyota* bartayresiana against Xanthomonas axonopodis pv citri which was the highest antibacterial activity recorded among the crude extracts of brown algae Against X. investigated. campestris pv malvacearum, the zone of inhibition was comparatively less from the chloroform: methanol 1:1(v/v) extract of Dictyota bartayresiana, in 2:1 (v/v) chloroform: methanol extract exhibited optimum antibacterial activity. Moderate antibacterial activity was recorded from the chloroform: methanol (1:1v/v) extracts of Hydroclathrus clathratus and Saragassum wightii. The crude extracts obtained in 1:1(v/v) and 2:1(v/v) chloroform: methanol of Dictyota dichotoma, Padina boergesenii, Stoechospermum marginatum and Sargassum illicifolium exhibited less antibacterial activity against both the bacteria tested. The crude extracts obtained in 2:1 (v/v) and 1:1 (v/v) chloroform: methanol of Sargassum longifolium and Turbinaria conoides showed only trace activity (Table 4).

DISCUSSION

Available reports on bioactive substances from seaweeds are mainly confined to human pathogens rather than plant pathogens^{3,14}. Kulik¹³ indicated the significance of evaluating algae for use in the biological control of plant pathogenic bacteria and fungi. In the present investigation, extracts of seaweeds found along the coast of Gulf of Mannar, Tamilnadu,India were evaluated for *in vitro* antibacterial activities against two plant pathogenic bacteria Xanthomonas axonopodis pv. citri and X. campestris pv. malvacearum. The antibacterial potential of shade dried specimens was as equal to fresh sample of seaweeds and extracts prepared from shade dried and fresh specimens of seaweeds possessed higher antibacterial activity than oven dried and sundried samples against the plant pathogenic bacterium Xanthomonas oryzae pv. oryzae in another investigation done¹⁶. They suggested that degradation of antibiotic substances may not occur during shade drying. Therefore in the present study, extracts were prepared from shade dried samples of seaweeds for the evaluation of antibacterial activity. In general, Gram positive bacteria are more susceptible than Gram negative bacteria to seaweeds extracts^{17,18}. However, 80% of algal extracts were active against both Gram negative and positive bacteria¹⁹. Gram negative plant pathogenic bacteria of Xanthomonas species were inhibited by the seaweeds extracts 3,14,16 . In the present study, out of 47 seaweeds belonging to Rhodophyceae(23), Chlorophyceae(15) and Phaeophyceae(9) ,the crude extracts prepared from 33 seaweeds exhibited antibacterial activity against Gram negative plant pathogenic bacteria axonopodis Xanthomonas pv. citri and X. campestris pv. malvacearum. Antibacterial activity was widespread throughout the members of Rhodophyceae, Chlorophyceae and Phaeophyceae⁷. Maximum and minimum antibiotic activities were observed in the members of Phaeophyceae and Rhodophyceae, respectively²⁰. The antibacterial activity in 11 seaweeds was displayed against plant pathogenic Xanthomonas oryzae pv. oryzae causing bacterial blight of rice in vitro¹⁶. The extracts of brown seaweeds showed high degree of antibacterial activity than red and green seaweed extracts. In the present study, among the 33 seaweeds which exhibited antibacterial activity, the red seaweed (Portieria hornemannii) and brown seaweeds (Sargassum longifolium and Turbinaria conoides) exhibited maximum and minimum antibacterial activities, respectively^{21,22}. Suitable solvents/methods determine the strength of the The active principles. most desirable

solvent/method could extract the maximum bioactive substances 16,23 . In the present study, a maximum antibacterial activity was found in the crude extracts obtained in 1:1(v/v) chloroform: methanol followed by 2:1(v/v) chloroform: methanol, chloroform, methanol, petroleum ether, ethanol and diethyl ether against the plant pathogenic bacteria Xanthomonas axonopodis pv. citri and X. campestris pv. malvacearum. Similarly, the lipophilic fraction of brown seaweeds inhibiting the growth of Tobacco Mosaic Virus (TMV) and plant pathogenic fungus Phoma tracheiphila which causes "malsecco" disease in citrus plants in vitro was recorded^{21,24,25}. Sulphoglycerolipids was isolated from the methanolic extracts of brown alga Sargassum wightii inhibiting the growth of *Xanthomonas oryzae* pv. *oryzae in vitro*³. In the present study, extracts obtained in 1:1(v/v) and 2:1(v/v) chloroform: methanol showed good antibacterial activity against plant pathogenic bacteria X. axonopodis pv. citri and X. campestris pv. malvacearum suggested that the antibacterial substances present in the extracts of seaweeds presumably polar and non-polar in nature. Streptomycin sulphate commonly used against Xanthomonas axonopodis pv. citri and X. campestris pv. malvacearum is expensive. Antibacterial activity of seaweeds observed in the present study against test bacteria was lower than streptomycin sulphate. Sulphoglycerolipid isolated from the methanol extract of brown alga Sargassum wightii displayed strong antibacterial activity against Xanthomonas oryzae pv. oryzae causing bacterial blight of rice³. It is evident from the present study that extracts of algae as a source of less expense antibacterial compounds in crude form would provide strong antibacterial activity on purification.

It is concluded that seaweeds occurring along the coast of Gulf of Mannar, Tamilnadu, India showed to be a promising source of bioactive compounds to control the plant pathogenic bacteria such as *Xanthomonas axonopodis* pv. *citri* and *X. campestris* pv. *malvacearum* causing canker in citrus and angular spot in cotton, respectively.

Crude extracts	Zone of inhibition(mm diameter)					
	Gracilaria edulis		Ulva flexuosa		Sargassum wightii	
	X.a.c	X.c.m	X.a.c	X.c.m	X.a.c	X.c.m
Diethyl ether	7.3±0.56	+	6.6±0.74	6.5±0.50	7.0±1.50	+
Petroleum ether	7.6±0.53	+	7.6±0.53	7.0±1.00	7.6±0.53	7.0±1.51
Chloroform	8.0±0.40	+	8.0±1.50	7.0±0.50	7.0±1.05	+
Methanol	8.0±1.00	+	9.0±0.50	8.0±0.70	7.6±0.53	+
Ethanol	7.3±0.56	+	9.0±1.00	8.0±1.50	6.0±0.50	+
Chloroform:methanol(2:1 v/v)	11.6±1.76	7.0±0.81	13.7±0.71	8.7±0.50	16.3±1.17	10.3±0.56
Chloroform: methanol(1:1 v/v)	14.2±2.60	11.5±0.67	15.5±1.30	12.7±2.5	18.2±1.50	14.0±1.00

Table 1: Antibacterial activity(zone of inhibition) of crude extracts obtained in different solvents and methods from three selected seaweeds collected along the coast of Pamban(Rameswaram), Gulf of Mannar, Tamilnadu, India during November 2009

+ Trace activity

X.a.c- Xanthomonas axonopodis pv. citri

X.c.m-X. campestris pv. malvacearum

Table 2: Antibacterial activity (zone of inhibition) of crude extracts obtained in chloroform: methanol 2:1(v/v) and 1:1(v/v) from the red seaweeds collected along the coast of Pamban (Rameswaram), Gulf of Mannar, Tamilnadu, India during November 2009

Red seaweeds	Zone of inhibition(mm diameter)					
	2:1	.(v/v)	1:1(v/v)			
	X.a.c	X.c.m	X.a.c	X.c.m		
Streptomycin sulphate	38.6 ± 0.53	31.7±0.56				
Liagora albicans Lamour.	+	-	8.3±0.56	7.6±0.53		
Gelidiella acerosa (Fors.) J.Feld & Ham.	10.7±0.54	7.0±1.00	12.0±0.76	8.0±1.00		
Portieria hornemannii (Lyngbye) P. Silva	20.0±1.00	18.3±1.80	25.3±2.10	22.4±1.20		
Amphiroa fragilissima(L.) Lamour.	13.44±0.44	9.3±0.56	15.0±0.24	10.7±0.55		
Gelidiopsis variabilis (Greville) Schm.	-	-	-	-		
Jania rubens(L.) Lamour.	-	-	-	-		
Grateloupia lithophila Børgesen	-	-	7.0±0.66	+		
Halymenia floresia(Clemente y Rubio)C. Agardh	-	-	8.3±0.56	+		
Gracilaria corticata var. corticata (J. Agardh)J. Agardh	-	-	7.9±1.70	+		
G. corticata var. cylindrica (J. Agardh) Umamaheswara Rao	16.60±1.30	9.2±0.58	18.30±0.99	10.4±0.35		
G. crassa Harvey ex J. Agardh	15.0±0.35	7.0±1.00	21.8±1.90	14.8±1.90		
G. edulis (S. Gemelin) P. Silva	7.9±0.77	9.6±0.53	8.7±1.80	10.6 ±0.67		
G. foliifera(Forsskal) Børgesen	-	-	7.8±0.33	+		
Sarconema furcellatum Zanardini	-	-	-	-		
Hypnea musciformis(Hulgen)Lamour.	15.3±1.70	7.0±1.00	16.0±2.1	8.3±1.88		
H. valentiae (Turner) Mont.	7.0±1.0	8.3±0.55	8.0±2.0	7.6±0.23		
Champia parvula (C.Ag.)Harvey	16.3±0.50	7.0±1.00	18.5±1.41	8.7±2.10		
Centroceras clavulatum (C.Ag.) Mont.	+	+	10.0±1.0	7.3±0.56		
Spyridia filamentosa(Wulfen) Harvey	10.4±2.50	+	11.2±0.53	+		
Acathophora spicifera (Vahl.) Børgesen	15.0±0.23	7.6±1.30	17.7±1.33	9.40±1.33		
Laurencia obtusa (Huds.) Lamour.	-	-	-	-		
L. papillosa (C.Agardh) Greville	9.6±0.57	+	11.1±1.44	+		
L.poiteii(Lamour.) Howe	7.0±1.50	+	9.3±0.56	+		

+ Trace activity

- Nil activity X.a.c- Xanthomonas axonopodis pv. citri

X.c.m-X. campestris pv. malvacearum

Table 3: Antibacterial activity(zone of inhibition) of crude extracts obtained in chloroform:methanol 2:1(v/v) and 1:1(v/v) from the green seaweeds collected along the coast of Pamban(Rameswaram), Gulf of Mannar, Tamilnadu, India during November 2009

Green seaweeds	Zone of inhibition(mm diameter)				
	2:1(v/v)		1:1(v/v)		
	X.a.c	X.c.m	X.a.c	X.c.m	
Streptomycin sulphate	38.6±0.53	31.7±0.56			
Ulva(Enteromorpha) compressa (L.) Nees	16.0±1.70	11.5±1.50	18.7±2.65	14.7±2.50	
Chaetomorpha linum (O.F.Muller) Kutzing	-	-	-	-	
Cladophora colabens Børgesen	-	-	-	-	
C.fasicularis Kutzing	10.5±0.39	8.5±0.67	14.6±1.50	11.2±0.77	
Caulerpa chemnitzia(Esper)Lamour.	-	-	-	-	
C. racemosa (Forsskal) J. Agardh	12.9±1.50	9.3±0.58	15.7±2.0	13.2±1.60	
C. scalpelliformis (R.Brown ex Turner) C.	12.1±1.50	9.7±0.58	15.6±2.0	13.7±1.60	
Agardh					
C. sertularioides (S. Gmelin) Howe	-	-	-	-	
C.peltata Lamour.	12.7±0.34	11.8±0.57	14.0±0.57	12.0±0.65	
plumosa(Hudson) C. Agardh	9.3±0.56	8.7±0.85	13.3±0.73	9.1±0.75	
Codium tomentosum Stackhouse	+	-	9.3±0.56	7.3±0.56	
Helimeda macroloba Decaisne	14.8±1.2	11.7±0.60	17.9±0.95	14.2±0.74	
Helimeda gracilis Harvey ex J.Agardh	-	-	-	-	
Borgesenia forbesii (Horvey) J. Feldmann	-	-	-	-	
Valoniopsis pachynema (G. Martens) Børgesen	10.6±0.65	7.6±0.51	14.3±0.70	9.8±0.58	

+ Trace activity

- Nil activity

X.a.c- Xanthomonas axonopodis pv. citri X.c.m-X. campestris pv. malvacearum

Table 4: Antibacterial activity (zone of inhibition) of crude extracts obtained in chloroform:methanol 2:1(v/v) and 1:1(v/v) from the brown seaweeds collected along the coast ofPamban(Rameswaram), Gulf of Mannar, Tamilnadu, India during November 2009

Brown seaweeds	Zone of inhibition in mm diameter				
	2:1(v/v)		1:1(v/v)		
	X.a.c	X.c.m	X.a.c	X.c.m	
Streptomycin sulphate	38.6± 0.53	31.7±0.56			
Dictyota bartayresiana Lamour.	15.7±1.87	12.4±2.10	19.8±2.56	17.3±1.50	
Dictyota dichotoma (Hudson) Lamour.	8.6±0.53	7.5±0.57	10.3±0.67	9.7±0.44	
Padina boergesenii Allender & Kraft	8.3±0.46	7.7±0.46	12.2±1.60	8.5±0.97	
Stoechospermum marginatum (C. Agardh) Kutzing	7.0±1.00	8.0±1.0	8.5±1.30	9.3±0.59	
Hydroclathrus clathratus(C.Agardh) Howe	14.7±1.00	10.3±0.51	18.4±1.80	15.5±0.67	
Sargassum longifolium(Turner) C.Agardh	+	+	+	+	
Sargassum ilicifolium(Turner) C.Agardh	9.4±0.56	7.8±1.50	13.2±1.0	9.8±1.60	
Sargassum wightii Greville	14.3±1.60	7.4±0.80	15.3±1.16	10.7±1.03	
Turbinaria conoides (J. Agardh) Kutzing	+	+	+	+	

+ Trace activity

X.a.c- Xanthomonas axonopodis pv. citri

X.c.m-X. campestris pv. malvacearum

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