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### Antimicrobial activity of macro algae against *Colletotrichum capsici* and its management

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#### Abstract

Marine macroalgae are rich sources of structurally novel and biologically active metabolites that could be used in biological control in the agricultural field. Many bioactive compounds have been identified from marine algae with antimicrobial activity, some of these compounds are sterols, terpenoids, polysaccharides, peptides, proteins, vitamins, acrylic acid, terpenes, chlorophyllides, phenols, heterocyclic compounds, halogenated ketones and alkanes and cyclic polysulphides. The present study is conducted to estimate the antimicrobial potential of extracts of fifteen marine macroalgae. Among 15 seaweed *Padina gymnospora* at 30% highest concentration was found to be the best in the reduction of mycelial growth and increased the germination percentage under *in vitro* condition. Application of *P. gymnospora*, in *in vivo* in the chilli pot experiment significantly decreased the percent of fruit rot in fruit in 43.43, 60.52 and 71.01% on 80,100 and 120 DAT respectively. Moreover, *P. gymnospora* enhanced growth performance of chilli plant in terms of shoot length and root length. Interestingly, the seed and air free pathogen treated with *P. gymnospora* showed significant increase flower compared to the control treatment besides increasing of fruit fresh weights. A considerable antimicrobial activity and the presence of phyto-chemical naphthalene indicated that *P. gymnospora* can be considered as a source of biocide and a biostimulant against anthracnose plant disease.

Keywords: Macro algae, Padina gymnospora, antimicrobial activity, phytochemical analysis

### Introduction

Chilli (Capsicum annum L.) is a spice crop which is extensively grown for its pungency. The capsaicin the alkaloid present in the placenta and pericarp is responsible for its pungency (Geetha and Selvarani 2017)<sup>[7]</sup>. India accounts for a major share in the total area under this crop. The production of chilli is affected by many prone fungal, bacterial and viral diseases. Among the fungal diseases, anthracnose or die back which is caused by crucial fungal pathogen Colletotrichum capsici (Sydow) Butler and Bisby, is a terrible constraint in successful cultivation of chilli throughout the world causing loss upto 80 percentage (Goswami et al. 2013)<sup>[8]</sup>. Management of fruit rot (anthracnose) by adapting the integrated strategies like mechanical, chemical, biological would be suitable to minimize yield loss (Agrios 2005)<sup>[2]</sup> as the use of chemicals lead to the development of soil pollution and food poisoning. The use of biopesticides/ biocides are the foremost approach to get the better of these nasty effects (Priva Reddy et al. 2019)<sup>[15]</sup>. Application of the natural algal extracts are nowadays more applicable instead of synthetic fungicides in controlling plant pathogen due to their higher safety and relatively negligible impacts on the environment (Gatal et al. 2011)<sup>[6]</sup>. Seaweed provide a rich source of antifungal compounds and biologically active secondary metabolites. Application of seaweed extracts is proved to be better to reduce the fungal diseases which ultimately increase its yield and help the growth of plants (Suthin raj et al. 2018) [21]. Seaweed extracts usage is a highly viable strategy, as these formulations contain many bio-elicitors (Phycoelicitors) which can significantly boost natural plant immunity. Certain bioactive elicitors present in a multitude of extracts of seaweeds (both commercially available and bench-scale laboratory formulations) activate pathogen-associated molecular patterns (PAMPs) due to their structural similarity (i.e., analogous structure) with pathogenderived molecules (Pushp Sheel Shukla 2021) <sup>[16]</sup>. A considerable number of recent studies showed that the crude and purified algal preparations are able to protect the plants against several pathogenic fungi (Paulert et al. 2010)<sup>[13]</sup>. The global production estimate of seaweed biomass for soil and plant applications is well over 550,000 tonnes per annum (Nayar and Bott 2014) [10].

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### Materials and Methods

### Isolation and purification of pathogen

The diseased fruit specimens were washed with clean tap water and the blackened parts were cut into small bits (4 mm), sterilized with 0.1% sodium hypochlorite (NaClO) for two minutes and rinsed in sterilized distilled water for three times and dried between folds of sterilized country filter paper. The sterilized infected fruit pieces were transferred on sterilized Potato Dextrose Agar (PDA) plates and incubated at room temperature for 5 days. Mycelial bits were transferred to sterile petridishes containing potato dextrose agar medium, purified by hyphal tip method and transferred to potato dextrose agar slants and pure cultures of the pathogens were maintained for further studies (Choi *et al.* 1999) <sup>[24]</sup>. The pathogen was identified on the basis of morphological, cultural and molecular characterization and further purified by single spore isolation technique (Nene and Thapliyal 1993) <sup>[11]</sup>.

Sl. No	Scientific name	Common name	Collected from
1.	Padina gymnospora	Brown seaweed	Pamban
2.	Hydroclathrus clathratus	Brown seaweed	Pamban
3.	Sargassum wightii	Brown seaweed	Kanyakumari
4.	Chnoospora implexa	Brown seaweed	Pondicherry
5.	Dictyota bartayresiana	Brown seaweed	Pamban
6.	Halimeda gracilis	Green seaweed	Pondicherry
7.	Caulerpa peltate	Green seaweed	Kanyakumari
8.	Caulerpa racemosa	Green seaweed	Rameshwaram
9.	Ulva lactuca	Green seaweed	Rameshwaram
10.	Ulva reticulate	Green seaweed	Rameshwaram
11.	Acanthophora spicifera	Red seaweed	Pondicherry
12.	Gracilaria corticata	Red seaweed	Pamban
13.	Liagora ceranoides	Red seaweed	Pamban
14.	Jania rubens	Red seaweed	Pondicherry
15.	Hypnea panosa	Red seaweed	Pamban

### List of seaweeds

### Poison food technique (Nishanthi et al. 2020)<sup>[12]</sup>

PDA medium was prepared in a 250 ml conical flask that was sterilized before the experiment. Seaweed extracts at quantities of 10, 20 and 30 ml were added to 70, 80 and 90 ml aliquots respectively in flasks so as to get final concentrations of 10, 20 and 30 percent. PDA medium without extract served as the control. Each plate was inoculated at the centre with a ten days old culture disc (9 mm) of pathogen and incubated at room temperature ( $28\pm2$  °C). Three replications were maintained for each treatment. The diameter of the mycelial growth of the pathogen was measured after 10 days. The inhibition over control of seaweed extracts was measured.

Percent Inhibition =  $(x_1 - x_2) / x_1 \times 100$ 

Where,  $x_1$  is colony diameter of the fungus in control plates (mm) and  $x_2$  is colony diameter of the fungus in treated plates (mm).

### Paper disc assay (Saha et al. 1995)<sup>[17]</sup>

Various concentrations like 10, 20 and 30 percent of seaweed extracts were made. Fifteen ml of PDA medium was seeded with 9 mm of *C. capsici* at the center of the petri dish and solidified. Sterile filter paper discs (10 mm) were dipped separately in known concentration of treatments and placed near the seeded medium. Three replications were maintained. The paper disc dipped in sterile distilled water served as a control. The plates were incubated at  $28\pm2$  °C for 10 days. The inhibition zone of fungal growth around the treated paper discs was measured and the percent inhibition over control was calculated.

### Agar well method (Thongson et al. 2004)<sup>[22]</sup>

Various concentrations of seaweed extract like 10, 20 and 30 percent were prepared individually. Fifteen ml of PDA medium were immediately poured into sterilized petri dishes

and were allowed to solidify. A 9 mm of PDA disc was removed using a cork borer to form wells; 9 mm of 10 days old cultures of *C. capsici* was placed on to the centre of the seeded medium and was incubated at  $28\pm2$  °C for 10 days. Potato dextrose agar medium with sterilized water served as the control. Three replications were maintained. The radial growth of the colony was measured and the percent inhibition of the growth was calculated.

### Effect of different seaweed extract on growth promotion of chilli by rolled towel method (Schroth 1990)<sup>[19]</sup>

Biostimulants of different seaweed extracts were assessed based on the germination percentage, shoot length, root length and seedling vigour index in the standard roll towel method given by the ISTA (1993). Thirty chillies were soaked for half an hour in different seaweed extracts. The standard roll towel was pre-soaked in sterilized water and placed in clean working table which was wipped out with ethanol 70%. The seeds were held in position by placing another one pre-soaked germination paper strip and were gently pressed. The polythene sheet were rolled along with seeds and incubated in a room temperature for 15 days. The chilli seeds soaked in sterilized water without seaweed extracts served as a control. Three replications were maintained for each treatment. The number of seeds germinated were counted and their root lengths, shoot length of individual seedlings were measured and the germination percentage was calculated. The vigour index was calculated using the formula described by Abdul Baki and Anderson (1973)<sup>[1]</sup>.

### Efficacy of seaweeds against anthracnose disease and yield attributes of chilli under greenhouse condition

The pot culture study was conducted with 5 treatments and three replications in the Department of Plant Pathology, Annamalai University, Annamalainagar from December 2019 to April 2020 (Trial-I). Thirty kilograms of topsoil collected from a field was steam pasteurized and filled in a cement pot. Thirty days old chilli seedlings of cv. K2 (K1 x sattur samba) were transplanted to cement pots (3 per pot). The five effective seaweed extracts (10%) were tested against anthracnose disease as detailed below Table 3 and 4. *C. capsici* was inoculated thoroughly over the fruit by spraying and on the 20<sup>th</sup> day after transplanting. The inoculated plants were incubated in a humid condition for 48h and subsequently moved to a greenhouse maintained at 22-28 °C, 98% relative humidity, under a light intensity of 81µmol m<sup>-1</sup>S<sup>-1</sup> and 12 h photo period and subsequently transferred to the pot culture yard.

### Analysis of the antifungal compound through Gas chromatography mass spectroscopy (GC-MS) (NIST Version. 2.0 2005)

20 gm powdered seaweed was soaked in 50 ml of absolute alcohol overnight and then filtered through a whatman filter paper 41 along with 2 gm sodium sulphate to remove the sediments and traces of water in the filtrate. Before filtering, the filter paper along with the sodium sulphate was wetted with absolute alcohol. The filtrate was concentrated by bubbling nitrogen gas into the solution and the volume was reduced to 1ml. The extract contained both polar and nonpolar phytochemicals of the plant material which was used. Based on the growth inhibition studies, seaweed extract was selected and chemical constituents were determined with a GC Clarus 500 Perkin Elmer Gas chromatography equipped with a mass detector. Turbo mass gold containing an Elite-1 (100% Dimethyl Poly Siloxane), 30 m × 0.25 mm ID employed were the following: Carrier gas, helium (1 mL/min); oven temperature program 110 °C (2 min) to 280 °C (9 min); injector temperature (250 °C); total GC time (36 min). The water extract was injected into the chromatograph in 2.0 ml aliquots. The major constituents were identified with the aid of a computer-driven algorithm and then by matching the mass spectrum of the analysis with that of a library (NIST Version. 2.0 year 2005). The software used for Gas Chromatography Mass Spectroscopy (GC-MS) was Turbo mass-5.1. This work was carried out in the IIFPT (Indian Institute of Food Processing Technology), Tanjavur (Ivanova et al. 2002) [9]

## Assessing the effect of seaweed against *C. capsici* under *in vitro* condition

### Poison food technique

Screening of 15 different seaweed (brown, red, green) against *C. capsici* on PDA plates is presented in Table 1. Among the different seaweeds extracts, the extract of *Padina gymnospora* at a high concentration (30%) was found to be the best in the reduction of mycelial growth (87.60 percent) which was followed by *Caulerpa racemosa* (74.83 percent). A maximum mycelial growth was observed in *Acanthophora spicifera* (54.55 percent).

### Paper disc method and agar well method

Various seaweed were selected and tested for their antifungal activity by paper disc and agar well methods (Table 1). *Padina gymnospora* 30% concentration recorded 85.31 and 86.54 percent inhibition zone which was followed by *Caulerpa racemosa* 70.72 and 71.96 percent. A least inhibition was observed in *Acanthophora spicifera* with 50.52 and 51.76 percent. The experiments revealed the superiority

of Padina gymnospora, hence, was used for further studies.

### Effect of different seaweed extract on growth promotion of chilli by rolled towel method

The chilli seeds were tested with a supernant of different brown, green, red extracts and were tested for their growth promoting activity under *in vitro* condition (Table 2). In general, the extract of different seaweed extracts increase plant growth when compared with the untreated check. The result showed that *Padina gymnospora* could increase the vigour index of chilli seedlings. It recorded a highest germination of 99.23 percent, mean shoot length (9.95 cm), root length (7.73 cm) and vigour index (1754.39) which was followed by *Caulerpa racemosa* which recorded a mean shoot length of 9.76 cm, root length (7.54 cm), vigour index (1684.16) with a germination percentage of 97.35.

The present *in vitro* experimental investigation exposed that out of fifteen different seaweed extracts, *Padina gymnospora* (Brown seaweed) was highly effective in inducing seedling germination. Hence, this seaweed was used for further studies.

### Efficacy of seaweed against anthracnose disease and yield attributes of chilli under greenhouse condition

The seaweed were tested against chilli fruit rot incidence in chilli under greenhouse condition. All the five seaweed significantly reduced fruit rot incidence than the control. Among the treatments, the application of *P. gymnospora* (Seedling root dip (SD) + foliar spraying (FS) on 80 and 120 DAT) recorded significantly a less fruit rot incidence of 43.43, 60.52 and 71.01 percent increase over the control on 100, 120 and 140 days after transplanting than the other treatments Table 3.

### Effect of seaweed extracts on growth and yield attributes under greenhouse condition

Chilli plants were treated with different seaweed extracts effective in promoting the growth of the plant, especially, the application of *P. gymnospora* (Seedling root dip (SD) + foliar spraying (FS) on 80 and 120 DAT) which significantly increased the mean plant height (112.12 cm), number of flowers/plant (179 nos), mean number of fruits/plant (102 nos), mean fruit length (8.8 cm) and fruit yield (350.01 g/plant) compared to all other treatments. In control, a minimum yield was recorded with a mean plant height of 69.13 cm, number of flowers/plant (70 nos), mean fruit yield (75.27 g/plant) Table 4.

### Analysis of antifungal compound through Gas Chromatography Mass Spectroscopy (GC-MS)

On the basis of performance of marine macro algae in the preceding *in vitro* studies, *P. gymnospora* was tested to determine the nature of chemical compound (s) present in its extract. The results revealed the presence of 9 compounds, *viz.*, Naphthalene, Docosane, 1,2-Benzenedicarboxylic acid-diethyl ester, Docosanoic acid- methyl ester, 6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a-tetrahydrobenzofuran-2(4H)-one,

11,14-Eicosadienoic acid, methyl ester, Ethanol 1,2-(9,12octadecadienyloxy), (z,z)-, Hexadecanoic acid and methyl ester which were present in *P. gymnospora*. The molecular weights, name of the compound, chemical formula, retention time and peak area percentage were given in figure 18 and 19, among these, Napthalene may be responsible for the inhibition of the growth of *C. capsici* (Fig 1).

#### Discussion

Various seaweed were tested against the test pathogen. Among the different seaweed extracts of *Padina gymnospora*, a high concentration (30%) was found to be the best in the reduction of mycelial growth in Poison food technique, Agar well method and Paper disc assay method and also reduce the fruit rot infection in green house condition. The brown seaweeds show high antifungal activity as compared to red and green alga. The brown seaweed contain high amount of flavonoid and phenolic compounds which could be the reason for antifungal activity (Cowan *et al.* 1999) <sup>[5]</sup>. Peres *et al.* (2012) <sup>[14]</sup> was the first to observe antifungal substances in seaweeds. They contain all major and minor plant nutrients as well as biocontrol properties and many organic compounds

such as auxins, gibberellins and precursors of ethylene and betaine which affect plant growth (Wu *et al.* 1997) <sup>[23]</sup>. The antimicrobial activity was due to the synthesis of sulphate galectins synthesized by red seaweeds, which are the major components of the extracellular matrix (Aruna *et al.* 2010; Souza *et al.* 2012) <sup>[4, 20]</sup>.

Most of the secondary metabolites are the bactericidal or the antimicrobial compounds derived from seaweed consisting of diverse groups of bacteriostatic properties such as brominates phenols, oxygen heterocyclic; Terpenols, Sterols, Polysaccharides, dibutenolides peptides and proteins. Although most of the antibiotics found from terrestrial sources are used as therapeutic agents to treat various diseases, the ocean has enormous an biodiversity and potential to provide novel compounds with commercial value (Anderson *et al.* 2006) <sup>[3]</sup>.

Table 1: Assessing the effect of	seaweed against C	C. capsici under in vitro condition

			Poi	son foo	d techn	ique							Inhibiti	on zone	•				
s.	Name of the	Maria	Mycelial growth			ibition o		Paper disc assay					A	gar we	ll metho	od			
ъ. No	seaweed		(mm) <sup>*</sup>	owin		ontrol (%		Myc	elial gr	owth		ibition o		Myc	elial gro	owth		ibition o	
	extract		()	1	(,)		$(\mathbf{mm})^*$		control (%)		(mm)*		control (%)						
		10%	20%	30%	10%	20%	30%	10%	20%	30%	10%	20%	30%	10%	20%	30%	10%	20%	30%
1.	Padina gymnospora	40.23 <sup>a</sup>	37.45 <sup>a</sup>	11.12 <sup>a</sup>	55.30 (48.04)	58.38 (49.82)	87.60 (69.38)	44.23ª	36.11ª	13.22ª	50.86 (45.49)	59.88 (50.69)	85.31 (67.46)	43.12 <sup>a</sup>	35.99ª	12.11 <sup>a</sup>	52.09 (4619)	60.01 (50.77)	86.54 (68.48)
2.	Hydroclathrus clathratus	56.77 <sup>ef</sup>	53.53 <sup>fg</sup>	32.20 <sup>g</sup>	36.92 (37.42)	40.52 (39.54)	64.22 (53.26)	60.73 <sup>ef</sup>	57.21 <sup>f</sup>	37.63 <sup>g</sup>	32.52 (34.77)	36.43 (37.13)	58.19 (49.71)	59.62 <sup>ef</sup>	56.10 <sup>f</sup>	36.52 <sup>h</sup>	33.76 (35.52	37.67 (37.86)	59.42 (50.43)
3.	Sargassum wightii	53.44 <sup>def</sup>	50.24 <sup>fg</sup>	30.91 <sup>fg</sup>	40.62 (39.59)	44.18 (41.66)	65.66 (54.13)	57.21 <sup>def</sup>	54.29 <sup>ef</sup>	34.71 <sup>fg</sup>	36.43 (37.13)	39.68 (39.04)	61.43 (51.61)	56.10 <sup>def</sup>	53.18 <sup>ef</sup>	33.60 <sup>gh</sup>	37.66 (37.86)	40.91 (39.76)	62.67 (52.34)
4.	Chnoospora implexa	57.21 <sup>fg</sup>	55.13 <sup>g</sup>	34.80 <sup>h</sup>	36.43 (37.13)	38.74 (38.49)	61.33 (51.54)	61.34 <sup>fg</sup>	58.21 <sup>f</sup>	38.20 <sup>h</sup>	31.84 (34.35)	35.32 (36.46)	57.56 (49.35)	60.23 <sup>fg</sup>	57.10 <sup>f</sup>	37.19 <sup>i</sup>	33.08 (35.11)	36.56 (37.20)	58.68 (49.99)
5.	Dictyota bartayresiana	61.98 <sup>h</sup>	59.52 <sup>hi</sup>	38.29 <sup>i</sup>	31.13 (33.91)	33.87 (35.59)	57.46 (49.29)	65.97 <sup>hi</sup>	61.29 <sup>gh</sup>	42.06 <sup>i</sup>	26.70 (31.11)	31.90 (34.39)		64.86 <sup>hi</sup>	60.18 <sup>gh</sup>	41.05 <sup>j</sup>	27.93 (31.90)	33.13 (35.14)	54.3 (47.52)
6.	Halimeda gracilis	51.23 <sup>cd</sup>	49.45 <sup>de</sup>	28.12 <sup>ef</sup>	43.07 (41.02)	45.06 (42.16)	68.76 (56.02)	55.47 <sup>cd</sup>	51.32 <sup>cd</sup>	32.02 <sup>de</sup>	38.37 (38.27)	42.98 (40.96)	64.42 (53.38)	54.36 <sup>cd</sup>	50.21 <sup>cd</sup>	31.01 <sup>def</sup>	39.60 (38.99)	44.21 (41.68)	65.54 (54.05)
7.	Caulerpa peltate	57.49 <sup>gh</sup>	54.47 <sup>h</sup>	33.14 <sup>i</sup>	36.12 (36.94)	39.48 (38.93)	63.18 (52.64)	61.75 <sup>gh</sup>	58.92 <sup>g</sup>	38.78 <sup>i</sup>	31.39 (34.07)	34.53 (35.99)	56.91 (48.97)	60.64 <sup>gh</sup>	57.81 <sup>g</sup>	37.67 <sup>j</sup>	32.62 (34.83)	35.76 (36.73)	58.14 (49.68)
8.	Caulerpa racemose	45.11 <sup>b</sup>	43.98 <sup>bc</sup>	22.65 <sup>b</sup>	49.88 (44.93)	51.13 (45.65)	74.83 (59.89)	49.21 <sup>b</sup>	45.24 <sup>bc</sup>	26.35 <sup>b</sup>	45.32 (42.31)	49.73 (44.84)	70.72 (57.24)	48.10 <sup>b</sup>	44.13 <sup>bc</sup>	25.24 <sup>b</sup>	46.56 (43.03)	50.97 (45.55)	71.96 (58.03)
9.	Ulva lactuca	47.23 <sup>bc</sup>	44.43 <sup>cd</sup>	23.10 <sup>cd</sup>	47.52 (43.58)	50.63 (45.36)	74.33 (59.56)	51.42 <sup>bc</sup>	46.35 <sup>bcd</sup>	28.21 <sup>cd</sup>	42.89 (40.91)	48.50 (44.14)	68.65 (55.95)	50.31 <sup>bc</sup>	45.24 <sup>bcd</sup>	27.10 <sup>cd</sup>	44.10 (41.61)	49.73 (44.85)	69.89 (56.72)
10.	Ulva reticulate	42.74 <sup>b</sup>	40.61 <sup>b</sup>	23.32°	52.51 (46.43)	54.87 (47.79)	74.08 (59.39)	46.83 <sup>b</sup>	42.72 <sup>b</sup>	26.92°	47.96 (43.83)	52.53 (46.45)	70.08 (56.83)	45.72 <sup>b</sup>	41.61 <sup>b</sup>	25.92°	49.20 (44.54)	53.77 (47.16)	71.20 (57.54)
11.	Acanthophora spicifera	63.01 <sup>ij</sup>	61.23 <sup>ij</sup>	40.90 <sup>j</sup>	29.99 (33.20)	31.97 (34.43)	54.55 (47.61)	68.13 <sup>ij</sup>	62.44 <sup>h</sup>	44.53 <sup>j</sup>	24.30 (29.53)	30.62 (33.59)	50.52 (45.29)	67.02 <sup>ij</sup>	61.33 <sup>h</sup>	43.42 <sup>k</sup>	25.53 (30.34)	31.86 (34.36)	51.76 (46.01)
12.	Gracilaria corticate	54.01 <sup>cde</sup>	52.13 <sup>ef</sup>	31.91 <sup>ef</sup>	39.99 (39.22)	42.08 (40.44)	64.54 (53.45)	59.04 <sup>de</sup>	54.23 <sup>de</sup>	35.78 <sup>ef</sup>	34.40 (35.91)	39.74 (39.07)	60.24 (50.91)	58.03 <sup>de</sup>	53.12 <sup>de</sup>	34.67 <sup>efg</sup>	35.52 (36.58)	40.98 (39.80)	62.14 (52.03)
13.	Liagora ceranoides	47.74 <sup>cd</sup>	45.54 <sup>de</sup>	24.21 <sup>de</sup>	46.96 (43.26)	49.40 (44.66)	73.10 (58.76)	51.92 <sup>cd</sup>	46.51 <sup>cd</sup>	28.44 <sup>de</sup>	42.31 (40.58)	48.32 (44.04)	68.40 (55.79)	50.81 <sup>cd</sup>	45.40 <sup>cd</sup>	27.33 <sup>de</sup>	43.54 (41.29)	49.56 (44.75)	69.63 (56.56)
14.	Jania rubens	60.13 <sup>gh</sup>	57.49 <sup>h</sup>	36.16 <sup>i</sup>	33.19 (35.18)	36.12 (36.94)	59.82 (50.66)	64.56 <sup>h</sup>	59.32 <sup>g</sup>	40.21 <sup>i</sup>	28.27 (32.12)	34.09 (35.72)	55.32 (48.05)	63.45 <sup>h</sup>	58.21 <sup>g</sup>	39.10 <sup>j</sup>	29.50 (32.89)	35.32 (36.46)	56.56 (48.77)
15.	Hypnea panosa	55.10 <sup>de</sup>	53.21 <sup>ef</sup>	32.98 <sup>fg</sup>	38.78 (38.52)	40.88 (39.74)	63.36 (52.75)	59.32 <sup>def</sup>	52.92 <sup>ef</sup>	36.51 <sup>efg</sup>	34.01 (35.67)	59.43 (50.44)	59.43 (50.44)	58.29 <sup>de</sup>	51.81 <sup>ef</sup>	35.40 <sup>fgh</sup>	35.23 (36.41)	42.43 (40.65)	60.67 (51.16)
	Control	90.00 <sup>j</sup>	90.00 <sup>j</sup>	90.00 <sup>k</sup>	-	-	-	90.00 <sup>j</sup>	$90.00^{i}$	90.00 <sup>k</sup>	-	-	-	90.00 <sup>j</sup>	$90.00^{\mathrm{i}}$	$90.00^{1}$	-	-	-
				. —				-	-										11.00

\*Mean of three replications, Data in parameters are arc sin transformed values. Values in the column followed by common letters do not differ significantly by DMRT (P=0.05)

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S. No	Name of the seaweed extract	Germination* (%)	Shoot length* (cm)	Root length* (cm)	Vigour index *
1.	Padina gymospora	99.23(84.97) <sup>a</sup>	9.95ª	7.73ª	1754.39 <sup>a</sup>
2.	Hydroclathrus clathratus	85.11(67.30) <sup>fghi</sup>	8.99 <sup>def</sup>	6.77 <sup>efg</sup>	1341.33 <sup>e</sup>
3.	Sargassum wightii	90.00(71.57) <sup>def</sup>	9.44 <sup>abcd</sup>	7.22 <sup>bcde</sup>	1499.40 <sup>bcd</sup>
4.	Chnoospora implexa	84.98(67.19) <sup>fghi</sup>	8.92 <sup>defg</sup>	6.70 <sup>fg</sup>	1327.39 <sup>ef</sup>
5.	Dictyota bartayresiana	82.21(65.05) <sup>hi</sup>	8.44 <sup>fg</sup>	6.22 <sup>h</sup>	1205.19 <sup>g</sup>
6.	Halimeda gracilis	91.23(72.77) <sup>de</sup>	9.53 <sup>abcd</sup>	7.31 <sup>abcd</sup>	1536.31 <sup>bc</sup>
7.	Caulerpa peltate	88.76(70.41) <sup>defg</sup>	9.22 <sup>bcd</sup>	7.00 <sup>def</sup>	1439.69 <sup>d</sup>
8.	Caulerpa racemosa	97.35(80.63) <sup>abc</sup>	9.76 <sup>ab</sup>	7.54 <sup>abc</sup>	1684.16 <sup>a</sup>
9.	Ulva lactuca	93.23(74.92)bc	9.62 <sup>abc</sup>	7.40 <sup>abcd</sup>	1586.77 <sup>b</sup>
10.	Ulva reticulate	97.29(80.52) <sup>ab</sup>	9.82 <sup>ab</sup>	7.61 <sup>ab</sup>	1395.14 <sup>a</sup>
11.	Acanthophora spicifera	80.01(63.44) <sup>i</sup>	8.32 <sup>g</sup>	6.02 <sup>h</sup>	1147.34 <sup>g</sup>
12.	Gracilaria corticata	86.45(68.40) <sup>efgh</sup>	9.14 <sup>cde</sup>	7.14 <sup>edef</sup>	1407.41 <sup>de</sup>
13.	Liagora ceranoides	92.11(73.69) <sup>cde</sup>	9.69 <sup>abc</sup>	7.47 <sup>abc</sup>	1580.61 <sup>bc</sup>
14.	Jania rubens	83.43(65.98) <sup>ghi</sup>	8.56 <sup>efg</sup>	6.34 <sup>gh</sup>	1243.11 <sup>fg</sup>
15.	Hypnea panosa	89.98(71.55) <sup>def</sup>	9.36 <sup>abcd</sup>	7.14 <sup>cdef</sup>	1484.67 <sup>cd</sup>
	Control	61.58(51.69) <sup>j</sup>	5.02 <sup>h</sup>	3.80 <sup>i</sup>	543.14 <sup>h</sup>

\* Mean of three replications, Data in parameters are arc sin transformed values,

Values in the column followed by common letters do not differ significantly by DMRT (P=0.05)

Table 3: Efficacy of seaweed against anthracnose disease and yield attributes of chilli under greenhouse condition

Fruit rot incidence on 100 <sup>th</sup> day	Disease reduction over control (%)	Fruit rot incidence on 120 <sup>th</sup> day	Disease reduction over control (%)	Fruit rot incidence on 140 <sup>th</sup> day	Disease reduction over control (%)
27.44(31.58) <sup>d</sup>	30.16	26.23(30.81) <sup>de</sup>	49.58	24.01(29.34) <sup>d</sup>	64.45
23.31(28.87) <sup>c</sup>	40.67	21.24(27.44) <sup>c</sup>	59.17	19.17(25.97) <sup>c</sup>	71.62
20.97(27.25) <sup>a</sup>	43.43	19.25(26.02) <sup>a</sup>	60.52	18.12(25.19) <sup>a</sup>	71.01
25.38(30.25) <sup>d</sup>	35.40	24.51(29.67) <sup>c</sup>	52.88	23.62(29.08) <sup>d</sup>	65.03
22.87(28.57) <sup>b</sup>	41.79	20.05(26.60) <sup>b</sup>	61.46	19.01(25.85) <sup>b</sup>	71.85
39.29(38.23) <sup>g</sup>	-	$52.02(46.16)^{g}$	-	67.54(55.27) <sup>g</sup>	-
	incidence on 100 <sup>th</sup> day 27.44(31.58) <sup>d</sup> 23.31(28.87) <sup>c</sup> 20.97(27.25) <sup>a</sup> 25.38(30.25) <sup>d</sup> 22.87(28.57) <sup>b</sup>	incidence on 100 <sup>th</sup> day reduction over control (%)   27.44(31.58) <sup>d</sup> 30.16   23.31(28.87) <sup>c</sup> 40.67   20.97(27.25) <sup>a</sup> 43.43   25.38(30.25) <sup>d</sup> 35.40   22.87(28.57) <sup>b</sup> 41.79   39.29(38.23) <sup>g</sup> -	incidence on 100 <sup>th</sup> day reduction over control (%) incidence on 120 <sup>th</sup> day   27.44(31.58) <sup>d</sup> 30.16 26.23(30.81) <sup>de</sup> 23.31(28.87) <sup>c</sup> 40.67 21.24(27.44) <sup>c</sup> 20.97(27.25) <sup>a</sup> 43.43 19.25(26.02) <sup>a</sup> 25.38(30.25) <sup>d</sup> 35.40 24.51(29.67) <sup>c</sup> 22.87(28.57) <sup>b</sup> 41.79 20.05(26.60) <sup>b</sup> 39.29(38.23) <sup>g</sup> - 52.02(46.16) <sup>g</sup>	incidence on 100th dayreduction over control (%)incidence on 120th dayreduction over control (%)27.44(31.58)d30.1626.23(30.81)de49.5823.31(28.87)c40.6721.24(27.44)c59.1720.97(27.25)a43.4319.25(26.02)a60.5225.38(30.25)d35.4024.51(29.67)c52.8822.87(28.57)b41.7920.05(26.60)b61.4639.29(38.23)g-52.02(46.16)g-	incidence on 100th dayreduction over control (%)incidence on 120th dayreduction over control (%)incidence on 140th day27.44(31.58)d30.1626.23(30.81)de49.5824.01(29.34)d23.31(28.87)c40.6721.24(27.44)c59.1719.17(25.97)c20.97(27.25)a43.4319.25(26.02)a60.5218.12(25.19)a25.38(30.25)d35.4024.51(29.67)c52.8823.62(29.08)d22.87(28.57)b41.7920.05(26.60)b61.4619.01(25.85)b39.29(38.23)g-52.02(46.16)g-67.54(55.27)g

\* Mean of three replications, Data in parameters are arc sin transformed values,

Values in the column followed by common letters do not differ significantly by DMRT (P=0.05)

Table 4: Effect of seaweed extracts on growth and yield attributes under greenhouse condition

Treatments	Mean plant height (cm)	Mean no. of flowers/ plant	Mean no. of fruits/ plant	Mean fruit length (cm)	Fruit yield (g/ plant)
T <sub>1</sub> - Seedling root dip (SD) + Foliar spraying (FS) of <i>Caulerpa racemosa</i> on 80 and 120 DAT	98.12d	157 <sup>d</sup>	79 <sup>e</sup>	7.5 <sup>d</sup>	313.03 <sup>cd</sup>
T <sub>2</sub> - Seedling root dip (SD) + Foliar spraying (FS) of <i>Ulva</i> reticulate on 80 and 120 DAT	109.16 <sup>ab</sup>	170 <sup>b</sup>	97 <sup>b</sup>	$8.5^{ab}$	346.47 <sup>a</sup>
T <sub>3</sub> - Seedling root dip (SD) + Foliar spraying (FS) of <i>Padina gymnospora</i> on 80 and 120 DAT	112.12ª	179ª	102ª	8.8ª	350.01ª
T <sub>4</sub> - Seedling root dip (SD) + Foliar spraying (FS) of Gracilaria corticate on 80 and 120 DAT	104.04 <sup>c</sup>	160 <sup>cd</sup>	83 <sup>d</sup>	8.2°	330.28 <sup>bc</sup>
T <sub>5</sub> - Seedling root dip (SD) + Foliar spraying (FS) of <i>Hydroclathrus clathratus</i> on 80 and 120 DAT	107.50 <sup>bc</sup>	165 <sup>bc</sup>	90°	8.6 <sup>b</sup>	339.12 <sup>ab</sup>
T <sub>6</sub> - Inoculated control	55.25 <sup>g</sup>	65 <sup>h</sup>	41 <sup>i</sup>	4.0 <sup>g</sup>	151.21 <sup>h</sup>
T <sub>7</sub> - Control	69.13 <sup>f</sup>	70 <sup>h</sup>	50 <sup>h</sup>	5.2 <sup>f</sup>	75.27 <sup>g</sup>

\* Mean of three replications, Data in parameters are arc sin transformed values,

Values in the column followed by common letters do not differ significantly by DMRT (P=0.05)

No RT (min)		Name of the compound	Molecular	Molecular Weight	Peak Area %	
1	5.27	Naphthalene	C10H8	128	35.47	
2	10.04	Docosane	C22H46	310	14.02	
3	11.36	1,2-Benzenedicarboxylicacid, diethyl ester	C12H14O4	222	21.96	
4	12.84	Docosanoic acid, methyl ester	C23H46O2	354	2.18	
5	13.15	6-Hydroxy-4,4,7a-trimethyl-5,6,7,7a- tetrahydrobenzofuran-2(4H)-one	C11H16O3	196	3.57	
6	14.16	Z,E-3,13-Octadecadien-1-ol	C18H34O	266	2.47	
7	14.24	11,14-Eicosadienoic acid,methyl ester	C21H38O2	322	6.66	
8	14.68	Ethanol,2-(9,12-octadecadienyloxy),(Z,Z)-	C20H38O2	310	2.33	
9	15.21	Hexadecanoic acid, methyl ester	C17H34O2	270	11.34	

### GC-MS/MS Chromatogram

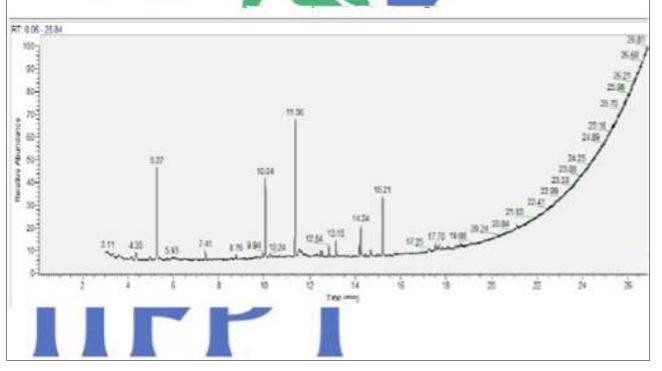


Fig 1: Analysis of antifungal compound through Gas Chromatography Mass Spectroscopy (GC-MS)

### Conclusion

A extensive review was done by Saxena *et al.* (2016) <sup>[18]</sup> on pathogen and disease management approaches in chilli anthracnose. In the absence of any exact method of controlling the disease, the chemical method has been sought as the most effective measure to control the spread of the disease. However, the remant toxic residues of the chemicals in the fruits create hindrance to the expected export of chilli products to other countries, inturn affecting the economy of the country. So, shift into biological strategies for disease management has stood up as a sustainable approach required for restoring the host homeostatis of the environment. So modifications of organic practices suiting to a particular agro climatic region will prove helpful in better management of the disease.

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