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Drenching aqueous extracts of seaweeds for enhancing growth, biochemical constituents and yield of *Solanum melongena*

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Original Article

Abstract

Seaweed liquid fertilizer prepared from aqueous extracts of red and brown seaweeds were tried on Brinjal, *Solanum melongena* L. under lab conditions. Basal application of aqueous extract of *Sargassum wightii* to Brinjal was found beneficial to boost the height of the plant, number of flowers/ plant considerably and the levels of chlorophyll pigments 27% more than the control. This increased the fruit yield exhibited by number and weight of fruits over control as well as the extract of red seaweed *Grateloupia lithophila*. However, extracts of *G. lithophila* promoted early flowering, delayed senescence of leaves of eggplant. Biochemical constituents in the treated plants such as total sugars, proteins and lipids too showed considerable increase (mg/g dry weight, $P<0.001$) due to application of *Sargassum* extract. These results enabled us to confirm that use of extracts of *Sargassum* as manure to crops can improve yield, save chemical fertilizers and protect soil fertility.

Keywords*: Seaweed manure, basal application, crop yield, green farming, soil fertility, Solanum melongena*

Introduction

Algae are morphologically simple, chlorophyll bearing, autotrophic aquatic organisms that range from microscopic and unicellular (single-celled) to very large and multicellular nature. The macroscopic marine algae are called seaweeds. Besides their immediate use as raw material for colloidal polysaccharides and many nutraceuticals, seaweeds have a major role to play in the concept of blue carbon through carbon sequestration (Zacharia *et al.*, 2015; Duarte *et al.*, 2017). In India, there is tremendous demand for food with increasing population. To meet this challenging demand, farmers are forced to use chemical fertilizers and pesticides in agricultural lands to enhance the crop yield. Chemical fertilizers have degraded the fertility of the soil and also accumulated toxic chemicals present in the inorganic fertilizers in plant products cause serious health problems in humans by biomagnifications. The undesirable effect of inorganic fertilizers on soil and environment is the foremost science to examine alternative biofertilizers (Metting *et al.,* 1990).

In recent years, seaweed extracts are produced and marketed as liquid fertilizer because they contain many growth promoting substances like auxins, gibberllins, trace elements, vitamins and amino acids. The use of seaweeds in modern agriculture has been investigated by many workers (Manimala and Rengasamy, 1993; Whapham *et al.*, 1993; Lopez-Musquera and Pazas, 1997). In this study we made an attempt to compare the efficiency of seaweed extracts obtained from seaweeds such as *Sargassum wightii* (Phaeophyceae) and *Grateloupia lithophyla* (Rhodophyceae) and their combined effect on the growth and yield of vegetable crop *Solanum melongena L.* plants.

Material and methods

Preparation of seaweed extract

Fresh seaweeds used in the present study were collected from Mullur (Vizhinjam, 08° 22′ 044″N & 77°00′ 201″E) and Manaserry (Ernakulam, 09°55′ 278″ N & 76° 15′ 011″ E) coasts during low tide (Indian Tide table 2017). The seaweed extracts were prepared separately from two species *viz. S. wightii* and *G. lithophila*. Weighed 400 g of *S. wightii,* were cut into small pieces and boiled in a pressure cooker for 30 minutes in 800 ml of distilled water. The hot extract was then filtered and allowed to cool at room temperature. Benzyl benzoate (2% wt/v.) was used as preservative. Similarly, extract was prepared from *G. lithophila.* The extracts were diluted with distilled water just before field application to achieve suitable dilutions.

Details of Treatments

The following experimental treatments were designed for the present investigation

- A- extract of *S. wightii* (5 ml extract diluted in 15 ml of water/day)
- B- extract of *G. lithophila* (5 ml extract diluted in 15 ml of water/day)
- AB-mixture of extracts of *S. wightii & G. lithophila* (2.5 ml each diluted in 15 ml of water/day)
- C-control (20 ml of water with which seaweed extracts were diluted)

Basal application of seaweed extract was done daily in the morning, three hours before watering.

Effect of seaweed extracts on seed germination

The seeds of Brinjal *S. melongena* L. var. *haritha* were purchased from the Kerala Horticulture sales outlet. Seeds were soaked in 25% seaweed extracts separately as shown above for 24 hours. Control seeds were soaked in distilled water for 24 hours. After a period of 24 hours at room temperature, 10 seeds each were placed on petri plates containing filter paper. Seeds containing petri plates were placed in light (two LED bulbs of 9 W at a height of 1 m for10 hours during day time) and in room temperature. The filter paper was kept moist by regular addition of tap water for control as well as treated seeds. The germination percentage was recorded at 120 hours after sowing.

Transplanting seedlings to pots

The seedlings were raised on plastic tray filled with garden soil. Seeds of eggplant were sown and water was sprinkled just sufficient enough to keep the soil moist twice a day. After germination and the seedlings reaching about 5 cm, the plantlets were carefully uprooted and transplanted into pots kept in the field containing equal quantity of potting mixture. 5 pots were selected for each experimental treatment. The removal of weeds and watering of the plants were carried out regularly.

Growth and Biochemical analysis

Growth parameters like plant height, number of leaves, number of flowers & fruits and leaf senescence were recorded at 5 and 10 day's intervals. Leaves were taken for the analysis of various biochemical constituents like total chlorophyll, Chlorophyll *a*, Chlorophyll *b* (Arnon, 1949 modified by Harborne, 1973), total protein (Lowry *et al.*, 1951), total soluble sugars (Dubois *et al.*, 1956), and total lipid content (Barnes and Blackstock, 1973).

Statistical Analysis

All the experimental data were analysed statistically by One-Way ANOVA using SPSS software, version 13 (SPSS, Bangalore, India).

Results

In the present study seaweed extracts obtained from *S. wightii* and *G. lithophila* were applied on brinjal plants through soil drench to evaluate the growth promoting effect by studying various growth parameters. Pre-soaking brinjal seeds in the extracts of two seaweeds and control (water) for 24 hours registered considerable increase in seed germination percentage when compared to control (Table 1). The treatment (A) with *Sargassum* extract had highest germination percentage of 96.66% while those seeds treated with *Grateloupia* extract (B) had the lowest germination percentage *i.e.* 66.66% which was also lower than that of the control plants. However, the mixture of equal quantities of both the extracts (AB) treated seeds also

Table 1. Effect of pre-soaking Brinjal seeds with seaweed extracts on seed germination (mean of three replicates)

Treatment	Number of seeds germinated	Total seeds	Percentage of seed germination
A	9.66 ± 0.38	10	96.66
B	6.66 ± 0. 27	10	66.66
AB	9.33 ± 0.52	10	93.33
C	8.0 ± 0.0.22	10	80

showed higher germination potential (93.33%). Control seeds (C) recorded germination potential of 80% (Table 1).

Height of shoot system of brinjal plants showed increase and positive response to the treatments compared to the control plants (Fig. 1). During the entire study period the brinjal plants drenched with *Sargassum* extract showed progressive increase in their shoot length. The highest numbers of leaves per plant were found in plants treated with *Grateloupia* extract in all fixed intervals of observation (Fig. 2). *Grateloupia* extract treated plants also showed delayed senescence of leaves (Table 2). *Sargassum* extract treated plants induced early flowering and maximum number of flowers (Table 3). Fruit setting was first observed in *Sargassum* extract treatment followed by *Grateloupia* extract treatment and combined treatment (AB). Highest mean fresh weight of fruit was encountered in *Sargassum* (8.97 g) treated plants than the *Grateloupia* (4.18 g) treated plants and the combined treatment plants (3.31 g). The maximum root length (Fig. 3) was found in plants treated with AB (26.44 cm) *i.e.*, combination of extracts of two seaweeds. Results presented in Table 4 shows *Sargassum*

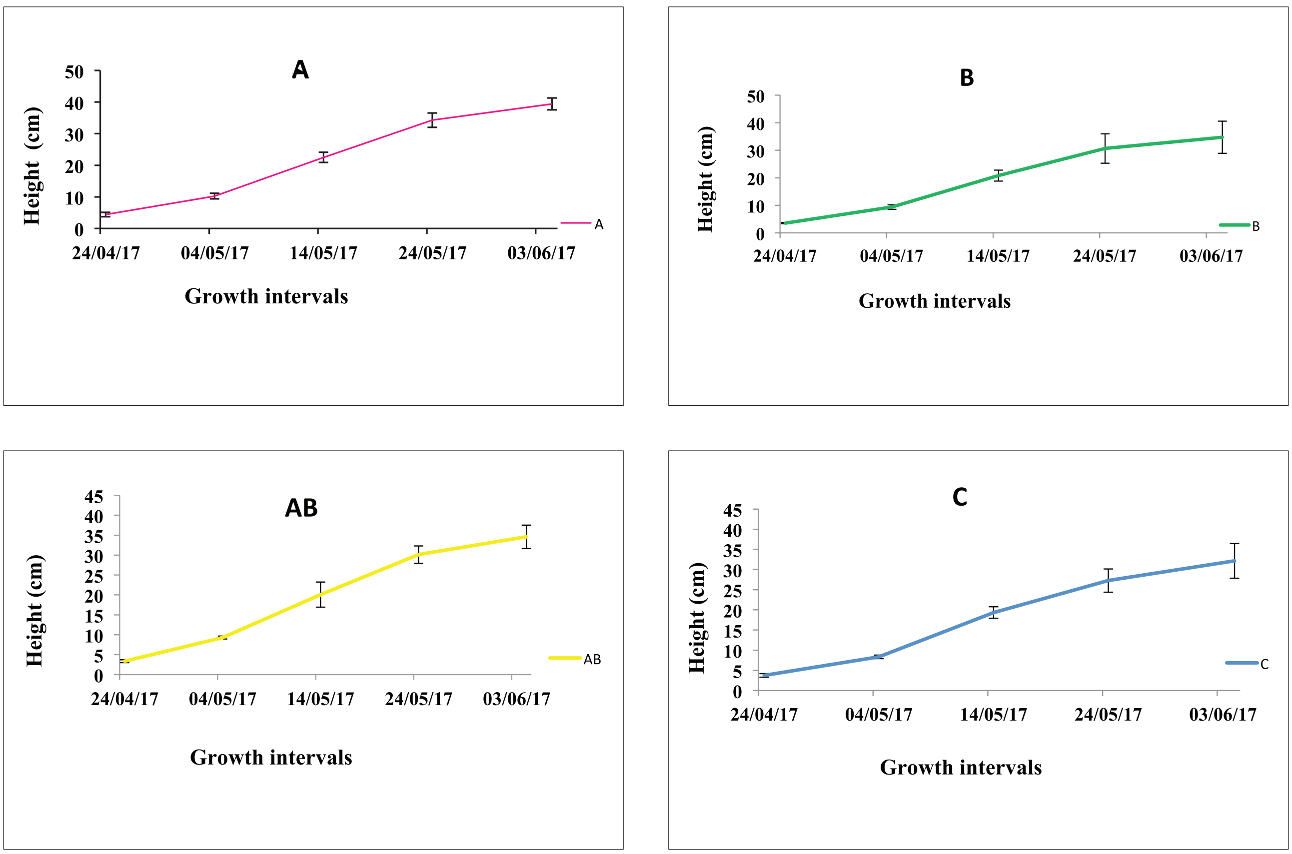

Fig. 1. Height of Brinjal plants treated with seaweed extracts

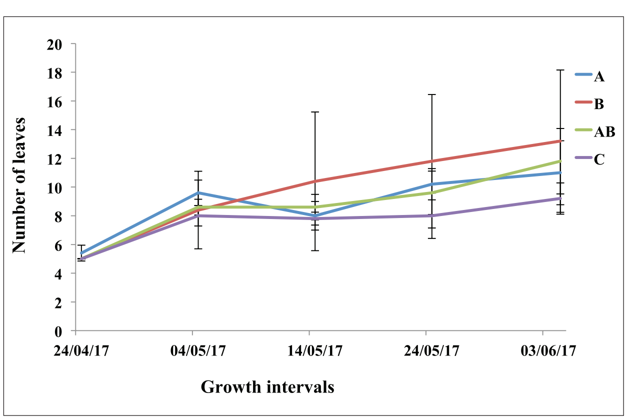

Fig. 2. Number of leaves in Brinjal plants treated with the seaweed extracts

extract treated plants achieved maximum amount of chlorophyll *a* (0.732±0.061mg/g f wt), chlorophyll *b* (0.300±0.028 mg/ g f wt) and total chlorophyll (1.032±0.088 mg/ g f wt). Variation observed in chlorophyll *a*, chlorophyll *b* and total chlorophyll between the different treatments with seaweed extract was found to be statistically significant ($P < 0.05$).

The accumulation of total sugars, proteins and lipids in the leaves of brinjal were more in the treated plants than in the control plants. Plants received *Sargassum* extract had maximum sugar content (166±12.49 mg/ g d wt). But AB treated plants had lower total sugar content than the control plants, while the *Grateloupia* extract treatment had higher levels than control

Table 2. Senescence of leaves in Brinjal plants treated with seaweed extract

Number of senescent leaves (mean at every 5 days interval, n=5)					
Treatments	22-05-2017	27-05-2017	01-06-2017	06-06-2017	11-06-2017
A	1 ± 0.71	0.4 ± 0.55	0.8 ± 0.84	0.4 ± 0.55	0.4 ± 0.55
B	0.4 ± 0.55	0.4 ± 0.55	0.6 ± 0.55	1 ± 0.71	0.4 ± 0.55
AB	0.8 ± 0.45	0.8 ± 0.45	1 ± 0.71	0.8 ± 0.45	0.4 ± 0.55
C	0.8 ± 0.45	1 ± 1	0.8 ± 0.45	0.8 ± 0.84	0.6 ± 0.55

Table 3. Number of flowers in Brinjal plants treated with seaweed extract

Number of flowers (mean at every 5 days interval, n= 5)						
Treatment	12-05-2017	17-05-2017	22-05-2017	27-05-2017	01-06-2017	06-06-2017
A	1.2 ± 1.09	2 ± 1.41	3 ± 2.83	2.8 ± 2.05	1.8 ± 1.30	1.8 ± 1.09
B	0.8 ± 0.84	1 ± 0	1.8 ± 1.30	1.6 ± 1.52	1.2 ± 0.84	1.8 ± 1.30
AB	0.6 ± 0.55	1 ± 0.71	1.6 ± 0.89	1.4 ± 1.52	1.4 ± 0.55	1.6 ± 1.34
C	0.8 ± 0.83	1 ± 1.22	2.4 ± 1.34	3.2 ± 1.92	2 ± 1	0.8 ± 0.45

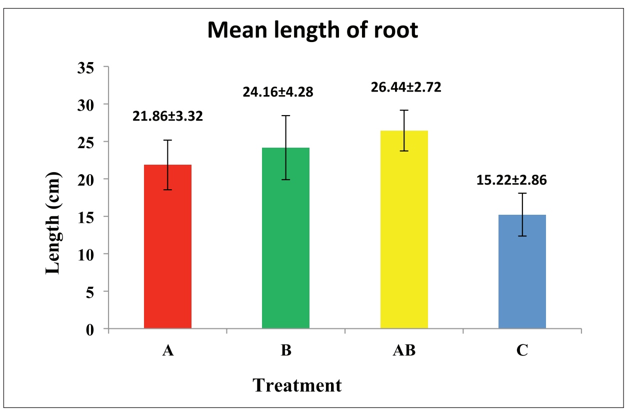

Fig. 3. Root length in Brinjal plants treated with the seaweed extracts

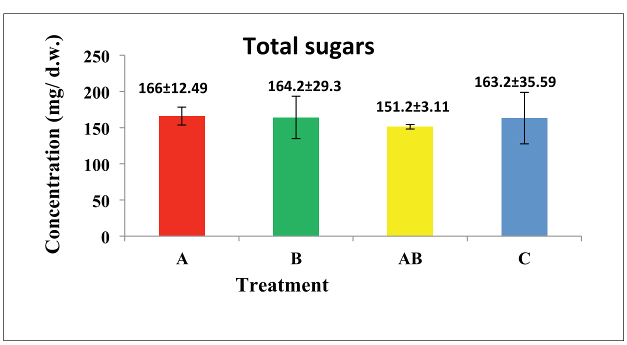

Fig. 4. Concentration of total sugars in Brinjal plants treated with seaweed extract

plants (Fig. 4). Maximum protein content in leaves was found in AB (75.65±11.32mg/ g d wt) treated plants and the set of *Sargassum* extract treated plants had higher protein content than that of *Grateloupia* extract treated plants (Fig. 5). Lipid content showed highest value in *Sargassum* (60.99±8.95 mg/ g d wt) extract treated plants (Fig. 6). Protein concentration in the leaves of the extract treated plants showed statistically significant difference with different seaweed extract application ($P<0.001$). Total sugars and lipid concentration in the leaves

Table 4. Mean fresh weight of fruits and levels of chlorophyll pigments in the leaves of Brinjal plants treated with seaweed extract

Treatments	Chlorophyll a (mg/g fresh wt.)	Chlorophyll b (mg/g fresh wt.)	Fruit weight (g)
A	0.732 ± 0.061	0.300 ± 0.028	8.97 ± 1.32
B	0.594 ± 0.074	0.243 ± 0.026	4.18 ± 1.04
AB	0.551 ± 0.132	0.234 ± 0.043	3.31 ± 0.76
C	0.553 ± 0.129	0.258 ± 0.037	0.0

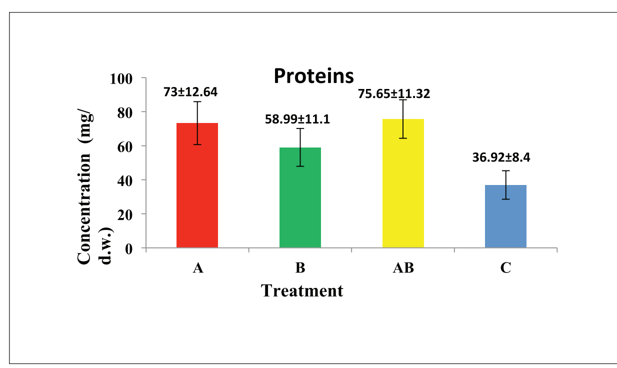

Fig. 5. Concentration of proteins in Brinjal plants treated with seaweed extract

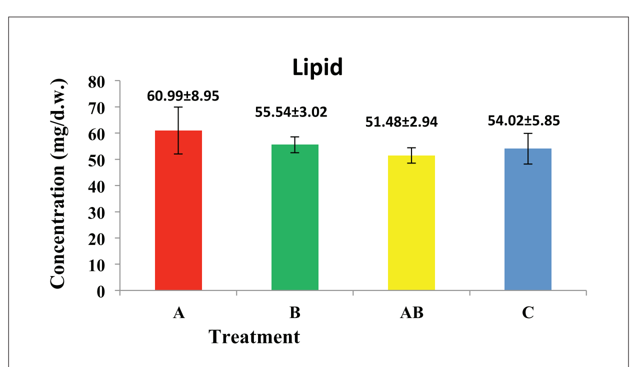

Fig. 6.Concentration of lipids in Brinjal plants treated with seaweed extract

of treated plants had P value greater than the 0.001. Hence total sugars and lipid did not exhibit any statistical significance.

Discussion

Seaweeds are utilised in different parts of the world in diversified fields such as phycocolloids, food, animal feed, fertilizer for crops etc (Dave *et al*., 1977). The high amount of water soluble potash, other minerals and trace elements present in seaweeds are readily absorbed by plants and they check deficiency diseases in crops. The manurial value of these products is not related to their N P K content and they show unusual properties such as enhanced germination of seeds, increased frost resistance and induce resistance to fungal and insect pests (Booth, 1969).

In the present study on the effect of treating seeds of *S. melogena* L. with different seaweed extracts from *S. wightii* and *G. lithophila* elucidated varied germination percentage. Seeds pre-soaked in *S. wightii* induced early germination and highest germination percentage (96.66%). The enhanced germination percentage of brinjal seeds due to pre-soaking in *Sargassum* extract obtained in this study may be due to the organic nature of seaweed extract containing several minerals, vitamins and plant growth hormones (Hong *et al*., 2007 and Kumari *et al*., 2002). Previous studies reveal that higher concentrations of seaweed extracts do play negative role in promoting germination (Jothinayagi and Anbazhagan, 2009; Kumar *et al.,* 2012 and Gayathri *et al*., 2014).

When the brinjal plants grown in pots were drenched with seaweed extracts, both the shoot and root lengths were found to increase progressively in various seaweed extract treatments. Similar observation was achieved by Zodape *et al.* (2011) when *Kappaphycus alvarezii* sap (50%) was applied as foliar spray to tomato plant.s over control plants. In the present study, the root length was enhanced by 43% by *S. wightii*, 58% by *G. lithophila* and 73% by the combination of both the extracts (AB) over the control. Similar trend is reported by Kumar *et al.* (2012) in *Vigna radiata*. In the present study the shoot length was the highest in those plants treated with *S. wightii.* Foliar spray of *S. wightii* extracts was found to exhibit promising effects on growth and yield characteristics of the brinjal plant (Divya *et al.,* 2015) which was attributed to the availability of macro and micro nutrients (Romero *et al*., 2000). The number of leaves per plant also was enhanced in different seaweed extract treated plants than the control. Drenching of *G. lithophila* extract (B) produced more number of leaves in all their progressive growth stages. Similar trends could be traced from different crops treated with different seaweed extracts like black gram (Renukabai *et al*., 2014), sunflower (Akila and Jeyadoss, 2010), groundnut (Sridhar and Rengasamy, 2010).

In the present study brinjal plants treated with seaweed extract showed early flowering and more number of flowers than the control plants. Seaweed extract is known to trigger early flowering and fruit set in a number of crop plants (Abetz and Young, 1983; Arthur *et al*., 2003; Kumar *et al.,* 2012 and Renukabai *et al.,* 2014). In the present study *Sargassum* extract induced early fruit set in treated plants followed by B treatment and finally in the combined (AB) treatment. According to Dwivedi *et al.* (2014) seaweed extracts not only increase the vegetative growth of the plant but also trigger the early flowering, fruiting in crops and ultimately on seed yields.

There was significant difference in chlorophyll content in seaweed extract treated brinjal plants. The plants that received *S. wightii* extract showed maximum amount of chl *a* (0.732$\pm$0.061mg/g f.wt.) and chl *b* (0.300$\pm$0.028mg/g f.wt.). It has been reported that seaweed liquid fertilizer of *S. wightii* increased the content of photosynthetic pigments in *V. radiata* (Sivasankari *et al.,* 2006). The *G. lithophila* extract treated plants also showed higher chlorophyll content than the control plants. The increase in photosynthetic pigments may be due to the presence of betaines (Blunden *et al.,* 1997). But the combined extract of *S. wightii* and *G. lithophila* treated plants had less chl *a* (0.551$\pm$0.132mg/g f.wt.), chl *b* (0.234$\pm$0.043mg/g f.wt.) and total chlorophyll (0.234$\pm$0.043mg/g f.wt.) than the control. Erulan *et al.* (2009) reported that increase in pigment content was observed in lower concentration of seaweed extract treatment and decreases when its concentration increases.

The biochemical analysis of the leaves of experimental plants revealed that the treated plants of brinjal contained protein, total sugars and total lipids considerably higher than the control plants. The plants treated with *S. wightii* had higher levels of total sugars (166$\pm$12.49 mg/g d.w.), protein (73.29$\pm$12.64 mg/g d.w.) and lipid (60.99$\pm$8.95 mg/g d.w.). Similar findings were reported by Kumar *et al.* (2012) who reported the accumulation of total protein, total sugars and total lipid content which was found maximum in *S. wightii* treated green gram. It has been reported that seaweed liquid fertilizer at 10% extracted from brown alga *S. wightii* increased the content of protein and total sugars in *V. radiata* (Sivasankari *et al.,* 2006). *G. lithophila* treated brinjal plants had total sugars (164.2$\pm$23.9 mg/g d.w.), protein (58.99$\pm$11.1 mg/g d.w.) and lipid (55.54$\pm$3.02 mg/g d.w.). But the combined treatment promoted higher protein (75.65$\pm$11.32 mg/g d.w.) accumulation than the total sugars and lipids. The enhanced biochemical constituents such as protein and sugars might be due to absorption of most of the necessary elements (co factors) present in the seaweeds that can trigger the metabolic pathways (Anantharaj and Venkatesalu, 2001). The present study has proved beyond doubt that aqueous extract (25%) of *S. wightii* can be a better substitute to inorganic fertilizers that are applied to brinjal plants and seaweed extracts prepared from seaweeds cast ashore can be promoted for organic farming of vegetable crops.

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