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Effects of seaweed (*Sargassum crassifolium*) extract foliar application on seedling performance of *Zea mays* L.

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Abstract

At present-day, Necessity for an ecological pleasing agriculture to nourish the increasing population is in high demand for the production of quality and healthy food. Efforts are underway for the sustainable crop production with organic fertilizers and botanicals from natural resources to enhance the production of commercially important crops. In this regard, a pot experiment was conducted at the Crop Farm of Eastern University, Sri Lanka, to assess the effects of foliar application of seaweed (Sargassum crassifolium) extract on seedling performance of Zea mays L. The experiment was arranged in a Completely Randomized Design (CRD) with three replicates. The seaweed extract at different concentrations (5%, 10%, 15%, 20%, 50% and 100%) and distilled water as control were applied to maize seedlings (up to 21 days old) once a week during experimental period. The seedling performance (Plant height (cm), leaf area (cm^2), fresh and dry weight of shoot and root (g)) were recorded at weekly intervals. Highest performance was recorded in application of 20 % seaweed extract than other treatments ($p<0.05$). The 20% concentration significantly increased the dry matter accumulation, plant height and leaf area of the seedlings. Therefore, it could be concluded that the seaweed extract at 20% concentration can be used to boost the vegetative growth of Zea mays seedlings.

Keywords: Foliar application, *Sargassum crassifolium*, Seaweed liquid extract, *Zea mays.*

Introduction

Detrimental effects of excessive usage of inorganic fertilizers to satisfy the food requirement for increasing world population than that of recommended usage have lead farmers towards the use of organic fertilizers. Use of seaweed extract as bio-stimulant is one of such viable options. Seaweeds are marine macro algae, which act as a significant element of the aquatic living resources of the world. They exist in depthless coastal water of sea, estuaries and backwaters. Almost 1700 km of coastline of Sri Lanka consisting of many varieties of seaweeds[1]. About 320 species belonging to different families have been identified by several workers especially in Northern, Western and Southern coastal areas of Sri Lanka[2]. There is no evidence of publication with regards to the availability of seaweeds in the East coast of Sri Lanka, especially in the coastal areas of Batticaloa district.

Essential substances such as trace elements and plant growth hormones required by growing plants are present in seaweeds. Apparently, seaweed manure is rich in potassium compared to farm manure[3]. There are many plant growth hormones, regulators and promoters offered to enhance yield attributes[4]. Seaweed liquid fertilizers consist of such growth stimulating hormones (IAA and IBA), cytokinins, gibberellins, trace elements, vitamins, amino acids, and micronutrients antibiotics through which it enhances the crop production[5]. In recent years, the uses of seaweed extracts have gained popularity due to their potential use in organic and sustainable agriculture[6]. Distinct features of seaweed liquid extract are biodegradable, nontoxic, non-polluting and non-hazardous to human, animals and birds which are commonly absent in chemical fertilizers[7].

Maize is the predominant field crop in the Eastern Province of Sri Lanka second only to paddy. Increasing the production of maize on large scale farms is a priority for the Government of Sri Lanka to meet the requirement of livestock feed mills and to safeguard the livestock industry. Bio fuel production requirements in other countries have driven supply shortages in the world market and the supply of maize has become a significant opportunity. There is significant unmet demand for maize to meet the country's growing requirements especially in the poultry feed industry and for export. The livestock feed industry needs about 200,000 Mt of maize annually whereas only about 65,000 Mt is produced locally. Sri Lanka imports maize annually at a cost of LKR 2.5 billion[8]. Therefore, this research was intended to reveal the potential of using the effects of seaweed of *Sargassum crassifolium* as a foliar application of the seedling performance of *Zea mays* L.

Materials and methods

The selected seaweed: Availability of seaweeds in Batticaloa was confirmed by direct observations and visiting coastal areas of Kallady, Savukkadi and Pasikudah. The species which are highly abundant in the area were collected and identified with the help of taxonomic classification key and confirmed by the experts attached with the Botany Department, Faculty of Science, Eastern University, Sri Lanka.

Collection of Seaweeds: The seaweed *Sargassum crassifolium* which are plenty in quantity were collected from Pasikudah. Foreign particles, sand and ephyphytes which adhered with seaweeds were removed by washing seawater followed by transported to the research laboratory to be used as the raw material for the preparation of seaweed liquid extract.

Preparation of Seaweed Liquid Extract: For the preparation of Seaweed Liquid Extract (SLE) according to the method described by Rao[9], seaweeds were shade dried for four days. The coarse powders of selected seaweeds and distilled water with the ratio of 1:20 (w/v) were mixed followed by autoclaved at 121°C, 15 lbs/sq inch for 20 minutes. After that, by using double layered cheese cloth, the hot extract was filtered followed by allowed to cool down. Finally the filtrate was centrifuged at 5000 × g for 15 minutes. The collected supernatant was considered as the 100% of SLE.

Physiochemical Analysis of Seaweed Liquid Extract: The color of SLE was observed visually and the pH and Electrical conductivity were measured using the pH/Conductivity/TDS tester (Model- HANNA HI 98130) followed by, the SLE was analyzed for different macro (nitrogen, phosphorous and Potassium, Magnesium and Calcium) and micro nutrients (Iron, Manganese, Zinc, and Copper). Nitrogen tested using kjelahl digestion, Phosphorous tested using UV visible spectrophotometry and other nutrients were analysed via atomic absorption spectrometry.

The studied Plant: Hybrid maize (*cv*. Passific) was used in this study for homogeneity of seed weight. And Seeds were taken at Seed Sales Unit from Eastern University, Sri Lanka.

Pot Experiment: Plastic pots were used to raise maize seedling. There were forty two pots with the dimension of 15 cm diameter and 20cm height. Firstly, the bottoms of the pots were covered by net in order to prevent the root penetration via drainage holes. Followed by, the potting mixture which consisting top soil, red soil and cow dung at the ratio of 1:1:2 was added. With that, the chemical fertilizers were also added according to the recommendation of department of agriculture.

One week after the basal application, the maize seeds were planted at the rate of two per pot. Later, only one plant was allowed to grow and other one was thinned out.

Experimental Design: The experiment consisted of seven treatments (5% SLE, 10% SLE, 15 SLE, 20% SLE, 50% SLE, 100% SLE (v/v) and distilled water spray (control)) and was designed in a Completely Randomized Design (CRD) with three replicates. The foliar spraying was done three times during the experimental period at one week interval. The recommendation of the Department of Agriculture, Sri Lanka was used for other crop management practices including fertilizer application. The total spray volume was 10 ml per plant at one time. For proper adherence of extracts with leaf surface, it was mixed with commercially available surfactant Hybrid^{SB}. Separate hand sprayers were used for each treatment.

Measurements: Data were collected at 21 days after sowing (DAS). Growth parameters i.e. plant height, leaf area, shoot fresh weight, root fresh weight; root dry weight and shoot dry weight were taken via destructive sampling method.

Data analysis: Data were statistically analyzed using SAS 9.1 and mean comparison was performed using Least Significant Test at 5% significant level.

Results and discussions

Physio-Chemical Properties Analysis of Seaweed *Sargassum crassifolium* Extract: The physiochemical properties of seaweed *Sargassum crassifolium* liquid extract have been analyzed. The extract was brown in colour and the pH recorded was 9.0 at room temperature. The extract contained macro nutrients like Nitrogen, Potassium, Phosphorous, Magnesium and micro nutrients like Iron, Manganese, Zinc and copper and their values are given in Table-1. *Sargassum crassifolium* liquid extract was rich in macronutrient of Potassium followed by Nitrogen, Magnesium and Phosphorous. Further, it also has reasonable quantity of micro nutrient of Zinc and Iron. Findings of this study revealed that *Sargassum crassifolium* liquid extract have the elements which required for plant growth and further activities.

The physio- chemical properties of the seaweeds used in this experiment were in agreement with the study of Sasikumar *et al*.[10].

Effect of Seaweed, *Sargassum crassifolium* Liquid Extract on seedling of *Zea mays* L. seedling: Plant Height: Analysis of data in Table-2 showed that height of Maize seedling was significantly influenced by different treatments compared with the control. Application of 20% concentration of SLE (T5) significantly increased plant height by 25.07% of Maize seedling in comparison to control (T1) plants ($P<0.05$) followed by T6, T4 and T3 This increasing effect might be due to the presence of macro and micro nutrients as well as growth promoting substances like auxin and cytokinin[5] in SLE of *Sargassum crassifolium*.

This was supported by Zodape *et al*.[11] who reported that application of *Kappaphycus alvarezii* (red algae), with lower concentration had significantly increased plant height of tomato. Similar results have been reported in *Vigna sinensis* L. by Sivashankari *et al*.[7] and in tomato by Featonby and Staden[12].

Table-1: Physio-chemical properties of seaweed liquid extract of *sargassum crassifolium* used for this preliminary study.

Variables	Value
Nitrogen	400.0 ppm
Phosphorous	9.0 ppm
Potassium	1520.0 ppm
Magnesium	176.0 ppm
Calcium	ND
Iron	2.4 ppm
Manganese	0.4 ppm
Zinc	3.2 ppm
Copper	0.2 ppm
Color	Brown
pH	9.0
EC	2.4 mS/ cm

Table-2: Effect of Foliar Application of Seaweed, *Sargassum crassifolium* Liquid Extract on Plant height at 21 Days after Sowing of *Zea mays* L. Seedlings.

Treatments	Plant height (cm)
T1 (control)	60.47±0.73de
T2 (5% SLE)	62.47±1.01 dc
T3 (10% SLE)	64.57±1.11bc
T4 (15% SLE)	66.70 ±1.46b
T5 (20% SLE)	75.63±1.23a
T6 (50% SLE)	75.60±1.21 a
T7 (100% SLE)	58.26±1.71 e

Value represents mean ± standard error of three replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (LSD).

Leaf Area: Leaf area is one of the important parameter which determines the final yield of Maize. Khan *et al.*[13] stated that growth parameters such as germination, leaf area index (LAI), relative growth rate (RGR) and net assimilation rate (NAR) are commonly used for the assessment of any crop growth. Those are the indicators used to express the productivity of the crops especially since cultivar having stable vegetative growth under environmental stresses and this leads to better economic return.

The present investigation showed that foliar application of *Sargassum crassifolium* with 20% concentration (T5) on Maize seedling increased the average leaf area per plant by 37.87% These results were supported by Gurusaravanan *et al.*[14] who reported, lower concentration of seaweed extract of *Turbinaria decurrens* on chickpea increased the leaf area (15.5 cm^2 per plant) It also supported by Erulan *et al.*[15].

Table-3: Effect of Foliar Application of Seaweed, *Sargassum crassifolium* Liquid Extract on on Leaf area at 21 Days After Sowing of Zea mays L. Seedlings

Treatments	Leaf area (cm^2)
T1 (control)	578.89±3.47 e
T2 (5% SLE)	592.30±4.47 ed
T3 (10% SLE)	610.14±5.31 cd
T4 (15% SLE)	673.40±11.70 b
T5 (20% SLE)	797.97±5.13 a
T6 (50% SLE)	667.91±9.47 b
T7 (100% SLE)	625.23±13.3 c

Value represents mean ± standard error of three replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (LSD).

It clearly indicates that foliar application of seaweed (*Sargassum crassifolium*) with 20% concentration favours the Maize seedling growth by increasing the photosynthesis through increased leaf area which is in agreement with Rasheed *et al.*[16]. Further, Crouch and Staden[5] reported that, there are many plant growth hormones, regulators and promoters available to enhance yield attributes.

Shoot Weight: The treatment of 20% concentration of SLE foliar application (T5) increased average fresh weight of shoot and average dry weight of shoot per plant significantly by 39.55% and 43.95% respectively ($p<0.05$) when compare to the control followed by T6, T4 and T3 while treatment T7 (application concentration is 100%) exhibited inhibitory effect on both fresh and dry weight of shoot (Table-3). It clearly showed that, foliar application of seaweed (*Sargassum crassifolium*) with 20% concentration on Maize increased the dry matter accumulation of shoots while 100% concentration exhibits inhibitory effects on dry matter accumulation. This was supported by Stephenson[17] where it was recorded that lower concentration of seaweed extract prepared from *Ascophyllum* (brown algae) and *Laminaria* (brown algae) accelerated the growth in maize. Further, Featon and Staden[18] observed an improvement in the growth of tomato root and shoot when a seaweed extract was used either as foliar spray or mixed with the soil, might be due to macro and micro elements as well as growth promoting substances like cytokinin.

Table-4: Effect of Foliar application of Seaweed, *Sargassum crassifolium* Liquid Extract on on Fresh and Dry weight of shoot at 21 Days after Sowing of *Zea mays* L. Seedlings

Treatments	Fresh weight of shoot (g)	Dry weight of shoot (g)
T1 (control)	52.053±1.17ed	7.53±0.43 dc
T2 (5% SLE)	54.077±1.52 d	7.79±0.76 dc
T3 (10% SLE)	58.853±0.91 c	8.74±0.43 bc
T4 (15% SLE)	63.707±1.16 b	9.66±0.34 ab
T5 (20% SLE)	72.643±1.43 a	10.84±0.54 a
T6 (50% SLE)	65.033± 1.09b	7.90±0.09 dc
T7 (100%SLE)	49.107±0.88 e	7.53±0.43 dc

Value represents mean ± standard error of three replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (LSD).

Leaf area is the main factor influence on dry matter production of plants and it is directly associated with fresh and dry weight of plant. As per the results obtained revealed that higher value of leaf area recorded in treatments which received seaweed foliar application than control treatment. It could be the reason for higher shoot fresh and dry weight in treatments of T5, T6, T4, T3 and T2 than control. Soldati *et al.*[19] reported that proportion of dry matter allocated to leaves remain fairly constant hence, an increase in leaf area leads to an increase in dry matter accumulation at the same time an increase in leaf area leads to an increase in rate of dry matter accumulation since light interception is directly related to leaf area during this stage of development.

Root Weight: According to the results (Table-5), T5 was the treatment which obtained the maximum average root fresh and dry weight, followed by T6, T4, and T3 while the least average root fresh and dry weight was witnessed in T7 when parallel to control (T1). In this present investigation, seedlings received with 20% SLE (T5) produced the highest root fresh and dry weight while seedling received with 100% SLE (T7) produced the lowest root fresh and dry weight. The present finding indicated, application of *Sargassum crassifolium* extract has increased both root fresh and dry matter accumulation in treated seedlings with 20% concentration by 58.49% and 55.02% respectively compared to control while higher concentration of 100% has exhibited inhibitory effect on root dry matter accumulation than control.

In this experiment, presence of good amount of P in the SLE which might promote the root development. An improved root system could be influenced by endogenous auxins as well as other compounds in the extracts[5, 20,21]. SLE is the opulent source of secondary nutrients like Mg; hence, it helps in root growth. At the same time, inhibitory effect of Seaweed liquid extract on growth of *Zea mays* seedlings might be due to very high salt index observed in seaweed extracts which in turn affected the growth.

Table-5: Effect of Foliar application of Seaweed, *Sargassum crassifolium* Liquid Extract on Fresh and Dry weight of root at 21 Days after Sowing of *Zea mays* L. Seedlings

Treatments	Fresh weight of root (g)	Dry weight of root (g)
T1 (control)	32.053±1.17 c	5.27±0.39 c
T2 (5% SLE)	34.350±1.71ed	5.19±0.18 c
T3 (10% SLE)	38.833±1.18cd	6.12±0.29 bc
T4 (15% SLE)	42.653±1.49cb	6.56±0.22 b
T5 (20% SLE)	50.803±1.28 a	8.17±0.46a
T6 (50% SLE)	44.000±2.28 b	6.39±0.32 b
T7 (100%SLE)	20.337±0.98 f	3.91±0.20 d

Value represents mean ± standard error of three replicates. Mean values in a column having the dissimilar letter/letters indicate significant differences at 5% level of significance (LSD).

The present findings coincide with earlier studies made in Tomato by Couch and van Staden[5] who reported that dry matter accumulation in root of Tomato enhanced with the foliar application of seaweed extract with lower concentration by stimulating root growth. Further, Finnie and Van Staden[22] reported that which high concentrations inhibited root growth but stimulatory effects were found at a lower concentration in tomato. Similar results were recorded by Gurusaravanan *et al.*[14] in *Turbinaria decurrens* liquid extract which induced maximum root growth in chick pea at lower concentration. All the parameters tested in this experiment revealed, Lower concentration showed better performance while higher concentration exhibited inhibitory effect on growth of *Zea mays* seedlings over the control. The literature says this might be due to the higher salt index of seaweed liquid extract[6], hormonal effect of the seaweed is activated at lower concentration of seaweed extract and when the dilution rate is decrease, the availability of nutrients become reduced[23].

Conclusion

From the results demonstrated, it is concluded that seaweed (*Sargassum crassifolium*) liquid extract enhanced the maximum tested parameters of Maize seedling at the concentration of 20%. It is also obvious that seedling performances of maize increased up to 20% but reduced at higher concentration. However, it's clear that the treatment with *Sargassum crassifolium* liquid extract would be very effective at 20% concentration of the growth performances of *Zea mays* L. seedlings.

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