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Phytochemical response of sweet basil (*Ocimum basilicum* L.) variety CIM Shishir to foliar application of botanical biostimulants

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Abstract

The aromatic herb basil is rich in minerals, micronutrients, and therapeutic properties. The present pot culture study was conducted to investigate the impact of foliar application of botanical extracts on the physiology of crop along with its biomass, essential oil yield and composition in *Ocimum basilicum* L. cv. CIM-Shisir. The experiment consists of thirteen treatments of foliar application of botanical biostimulants having three consecutive concentrations (0.5%, 1.0% and 1.5%), arranged in completely randomized with four replications. Three foliar applications of all the treatments were done at pre branching stage (25 DAP), branching stage (35 DAP) and pre-flowering stage (55 DAP). The study revealed that 1.0% *Echinochloa colona* L. extract produced higher fresh biomass (266 gram pot^{-1}), dry biomass (89.7 gram pot^{-1}), fresh root (48.5 gram pot^{-1}), dry root (20 gram pot^{-1}), oil yield (2.4 ml pot^{-1}) and relative water content (59.13%) of basil compared to all the other biostimulants and control treatments. Although 1.0% seaweed extract gave higher total chlorophyll content (0.71 mg g^{-1} fresh weight). Major volatile compounds of oil were Linalool (63-68%), Camphor (9.5-10.3%), and 1,8 Cineole (7.1-8.5%). Conclusively these weed leaf extracts may be used effectively to improve the growth and yield of basil.

Keywords biostimulants, essential oil, herb yield, sweet basil, weed leaf extract

Introduction

The basil plant (*Ocimum basilicum* L.) belongs to the mint family (*Lamiaceae*). For millennia, it has been used in a variety of cooking traditions and practices [1]. There are about 50 to 150 species and varieties of Ocimum native to Asia, Central Africa, and South America [2]. The great variation among the constituent species largely contributes to uncertainty in estimating the number of species within the genus. Svecova and Neugebauerova [3] described the differences in the morphology, growth habit, and color of flowers, leaves, and stems as well as the chemical composition of the plants. Some authors have reclassified sections of the genus due to the ease with which basil cross-pollinates [4]. Basil is also known as thyme, a species that lives outside the Ocimum genus, and wild basil, a species that lives in the genus Clinopodium. This can sometimes cause confusion between the two, especially for the inexperienced. Traditional medical use of basil includes treating headaches, coughing, diarrhea, warts, worms, constipation and kidney dysfunction.

Basil oil can be applied to the skin to treat acne, and it is also used externally as a lotion for insect bites. There has been a great deal of effort put forward to improve soil quality, and agricultural products, and eliminate polluters. One of the techniques that are currently being researched extensively is the application of bio-stimulators which is the changes made to specific oligopeptides and purification of proteins and amino acid compounds from biological sources. Metabolic processes are stimulated by bio-stimulators to increase plant yield.

Seaweed extracts have a lot of potential to make agriculture sustainable due to their ability to provide nutrition to the plants without harming the soil and the environment. Many Plant Growth Regulators in Seaweed, including IAA, gibberellins, cytokinins, and glycine betaine, are the reason it is producing many physiological reactions in plants, along with needed macro- and micronutrients. As a result, crops are more productive and quality is enhanced [5-6]. As one of the most useful trees in the world, Moringa (*Moringa oleifera*) offers many beneficial properties that can be used for different purposes [7-8]. In addition to its plant biostimulatory properties, Moringa leaf extract (MLE) is also regarded as a valuable alternative source of nutrition coming from different minerals [9]. In addition to its high content of stimulants, MLE also contains a balanced cocktail of phytohormones, nutrients, and antioxidants, making it one of the most effective plant biostimulants available [10]. Researchers are studying low-cost organic inputs that will improve yield and quality to meet current industrial demands. The higher nutrient concentrations in weeds' respective biomass could also act as a biostimulant because they accumulate nutrients from cropped soil. Their tissue tends to accumulate macronutrients and micronutrients at a quicker rate than other crops as reported by Rao and Matsumoto [11]. It has been reported that weed leaf extracts (WLE) can increase chlorophyll content, pod number plant-1, and yield of beans [12].

Hence, the present experiment was conducted to study the potential of two selected weed plants (*Cyperus rotundus* L. and *Echinochloa colona* L.), Moringa leaf extract and seaweed extract as biostimulants in enhancing the growth, yield, and essential oil quality of sweet basil.

Methodology

Experimental Site/location

The pot experiment was conducted from June to September 2020 at the experimental farm of CSIR-Central Institute of Medicinal and Aromatic Plants, Lucknow, located at 26°5´N latitude 80°5´E longitude with an elevation of about 120 m above mean sea level under the sub-tropical plains of north- India. Metrological data of experimental duration has been presented in Figure 1.

Experimental details

The experiment consisted of thirteen treatments of foliar application of different plant based bio-stimulants or extracts in three consecutive concentrations (0.5%, 1.0% and 1.5%) of each extract. Details of treatment are as follows; T1- only water spray as control, T2- 0.5% seaweed extract, T3- 1.0% seaweed extract, T4-1.5% seaweed extract, T5-0.5% *Cyperus rotundus* extract, T6-1.0% *Cyperus rotundus* extract, T7-1.5% *Cyperus rotundus* extract, T8-0.5% *Echinocloa colona* extract, T9-1.0% *Echinocloa colona* extract, T10-1.5% *Echinocloa colona* extract, T11-0.5% Moringa leaf extract, T12-1.0% Moringa leaf extract, T13-1.5% Moringa leaf extract. The pot experiment was arranged in a Completely Randomized Design (CRB) with four replications. CIM-Shisir cultivar of Ocimum was selected for the experiment as a test crop.

Pot filling and raising of crop

For pot filling, well moist soil collected from the research farm of CSIR-CIMAP and mixed with vermicompost was taken up from the composting unit of CSIR-CIMAP. 8 kg soil mixture was filled in each pot. Thirty days old plants from the nursery of *Ocimum basilicum* L. cv. CIM-Shisir were transplanted from the experimental farm of CSIR-CIMAP, Lucknow to pots during the last week of June 2020. Light irrigation with the help of a sprinkler was provided to all the treatments simultaneously just after transplanting was completed and thereafter timely irrigation was provided to the crop as and when required. Three foliar applications of all the treatments were done at pre branching stage (25 DAP),

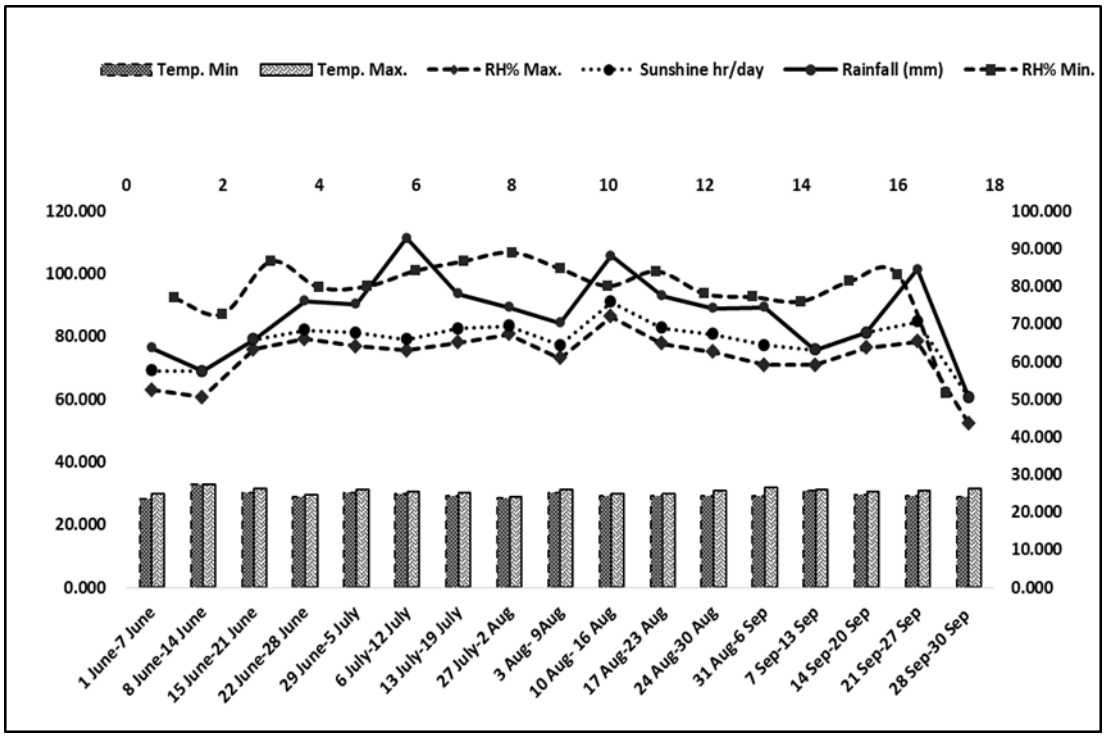

Figure 1. Meteorological data on mean weekly weather parameters and total rainfall during the cropping season

branching stage (35 DAP) and pre-flowering stage (55 DAP). Other important cultural practices including weeding, irrigation, hoeing and foliar application were done as per the schedule made relating to the crop growth.

Collection of plant materials and seaweed extract

The fresh plant material of selected weeds or plants *viz*. *Cyperus rotundus* L., *Echinochloa colona* L. and *Moringa oliefera* were collected from the different fields of the research farm of CSIR-CIMAP. Collected plant material was washed with double distilled water till the material was completely clean from soil particles or any type of dust particles. Seaweed extract was commercialized by IFFCO with the trade name "SAGARIKA" which was procured from an authorized center of Indian Farmers Fertilizer Cooperative Limited (IFFCO), Lucknow, India.

Preparation of plant extracts

Selected weeds and plants were taken from the CSIR-CIMAP research farm in Lucknow, and their leaves were cleaned with distilled water before being dried. To prevent the loss of active ingredients, one kilogram of leaves from each plant was air-dried at room temperature in the shade for two weeks. After that, the leaves were ground into a fine powder using an electric mixer grinder at the CSIR-CIMAP soil science laboratory. The ground sample was then stored for subsequent use in an airtight amber bottle. The extraction technique used was the Handa described hot-water extraction method [13]. To generate a 10 percent stock extract, 50 grams of powdered material were steeped in 500 milliliters (1:10 ratio) of distilled water in a round-bottom flask. The mixture was then heated for 20 minutes at 70°C on a temperature-controlled laboratory heating medium. After boiling, the produced extract was collected in a conical flask and allowed to cool. It was then double-filtered using cheesecloth. Subsequently, the extract was once more purified using Whatman No. 1 filtering paper. Then, airtight glass bottles of the extract were abridged at 4°C in a refrigerator after cooling and within five hours after extracting, the extracts were utilized.

Biometric observations

The estimation of all the parameters related to vegetative growth viz. height, plant canopy, no. of leaves/plant, leaf area index (LAI), no. of branches/plant, and was recorded at the growth stage and harvesting stage. To estimate LAI used a portable leaf area meter (Model: Li-Cor 3100, USA). At least three readings must be taken above and below the canopy to estimate LAI. All yield attributes *viz*. fresh herb yield, fresh root production per plant, shoot root ratio (S:R), and leaf stem ratio (L:S) were recorded at the time of harvesting. For dry matter estimation, 100 g of shoot sample and complete root sample were collected in a brown paper envelope and kept in a hot air oven for five days at 50°C. The samples' dry weight was determined using a laboratory weighing balance after the samples had fully dried.

Estimation of photosynthetic pigments

The chlorophyll-a and chlorophyll-b, along with carotenoid content in leaf samples of *Ocimum basilicum* were determined by Lichtenthaler and Wellburn [25] method with slight modification. To determine chlorophyll-a content the absorbance of extracts was measured at 645 nm, and the chlorophyll-b absorbance of extract was measured at 663 nm using Perkin Elmer Lambda 35 Uv/Vis spectrophotometer. Chlorophyll content estimation was done by grinding 0.2 g of leaf in 80% acetone and later those extracted samples were centrifuged at 7000 rpm for 5 min. The supernatant liquid was collected and kept. The whole process was repeated until the pellet turned whitish or yellowish. The supernatants were pooled, and their optical density was measured to estimate the chlorophyll content.

Estimation of chlorophyll fluorescence (fv fm^{-1}), proline content and relative water content of leaves

Estimation of Proline content was done by Bates et al., [27] method. The relative water content was estimated using the equation: RWC(%) = (FW - DW)/ (TW - DW) X 100, where FW represents the fresh weight of the leaves, TW represents the weight of the leaves at full turgor, while DW represents the fresh weight of the leaves after they have been dried at 80oC for four hours until a constant weight was attained [26]. Chlorophyll fluorescence (fv fm^{-1}) was recorded by software software-operated instrument designed and created by WALZ Mess-und Regeltechnik named Junior-Pam Chlorophyll Fluorometer. Fresh leaves of the *Ocimum basilicum* were collected from each pot in triplicate for chlorophyll fluorescence estimation at each recording.

Harvesting and essential oil extraction of the crop

All plants were manually harvested by secateurs after harvesting, from each pot, 100 grams of fresh plant material was collected for essential oil extraction by Clevenger apparatus through hydro-distillation method [14]. During the analysis of essential oil constituents, the separated essential oil was dried on anhydrous Sodium Sulphate, packed and then kept in the dark and in the freezer. Fresh herb yield was also recorded from each pot of the experiment and later oil content was multiplied by fresh herb yield to determine the oil yield. When the shoots reached full size in the last week of September 2020, the crop's fresh foliage was harvested manually.

Quality analysis of essential oil

Centurion Scientific Gas Chromatograph was used for the quality analysis of essential oils. Initially, the oven temperature was set at 70°C, then rose to 170°C at a rate of 4°C/min, followed by 240°C at a rate of 5°C/min, and then held for 5 and 15 minutes, respectively. Carrier gas and nitrogen were used at 1.0 ml/min. The detector temperature was 240°C while the injector temperature was 250°C. The sample of 0.2 µL which was diluted in hexane in 1:1 ratio was injected in split mode in 1:70 ratio. Utilizing a homologous series of n-alkanes and correlating data from the literature, the essential oil constituents were identified based on the retention index. With the help of GC peak area (FID response), relative amounts of all the components were calculated (No correction factor applied).

Statistical analysis

Shapiro–Wilk test of the data sets was conducted to check the normality distribution at a 5% level. One-way analysis of variance was applied to study the influence of various treatments on yield and quality of Ocimum basilicum. Duncan's Multiple Range Test was used to find out the significant variations between means of different treatments at p>0.005. SPSS 25, SPSS Inc. Chicago, IL were among the statistical software used.

Results and Discussion

Plant growth attributes

Although there was differential response of plants to these biostimulant applications, plant growth was significantly increased. The data presented in Table 1 revealed that during the harvesting stage, Basil crop growth attributes were significantly influenced by foliar application of different concentrations of plant-based bio-stimulants or extracts. Among different treatments, 1.0% *Moringa oleifera* leaf extract gave significantly highest plant height (67 cm), while 0.5% *Cyperus rotundus* extract and 1.0% seaweed extract gave lowest plant height (60cm, 60.5 cm) compared to all the other treatments. In the case of canopy coverage, 1.0% *Echinochloa colona* extract gave significantly the highest values (120.0 cm), while 1.0% seaweed extract gave the lowest canopy coverage (30.0 cm). 0.5% *Echinochloa colona* extract gave significantly highest number of leaves plant^{-1} (463) and no. of branches plant^{-1} (35.5), while 1.0% *Cyperus rotundus* extract gave the lowest values for no. of leaves plant^{-1} (364.3) and number of branches plant^{-1} (19.0). In the case of leaf area index 0.5% *Echinochloa colona* extract, 1.0% *Echinochloa colona* extract, and 1.5 % *Moringa oleifera* extract gave significantly highest values (1.55, 1.53, 1.53), while 0.5% *Cyperus rotundus* extract treated plants displayed lowest leaf area index value (1.25). The significant increase in plant height was because plant growth regulators (IAA, cytokinins, Zeatins) are naturally present in Moringa leaf extract, stimulating cell growth and resulting in improved plant growth. These findings were similar to those reported by different researchers [10, 15-16]. Similarly increase in the number of branches plant^{-1}, canopy, leaf area index, and number of leaves plant^{-1}, might be due to the reason that through photophosphorylation, weed leaf extracts enhance various metabolic and physiological processes that are essential to vegetative and reproductive growth. These findings were similar to those reported by Anisuzzaman et al., [17], Yadegari and Mosadeghzad [18] and Mazeed et al., [19].

Influence of bio-stimulants on the plant biochemical attributes of basil

Weed extracts gave different beneficial impacts on different biochemicals as there was no trend followed by any of the weed leaf extracts of any concentration in any of the biochemical constituents (Table 2). 1.0% Seaweed extract gave significantly highest total chlorophyll content (0.71 mg/g fresh weight) [Chlorophyll-a + Chlorophyll-b (Figure 2)], while 1.0% *Echinochloa colona* extract gave the lowest total chlorophyll content (0.32 mg/g fresh weight). In the case of chlorophyll fluorescence, 1.5% *Moringa oleifera* extract gave the significantly highest value (0.505 fv/fm), while 0.5% *Moringa oleifera* extract gave the lowest chlorophyll fluorescence value (0.258 fv/fm) 0.5% Echinochloa colona extract gave significantly the highest proline content (0.248 µ mol g-1 fresh weight), while 1.5% Moringa oleifera extract gave the lowest proline content (0.092 µ mol g^{-1} fresh weight). In the case of the Relative water content of leaves, 1.5% Cyperus rotundus extract gave significantly highest relative water content of leaves (59.13%), while control gave lowest relative water content of leaves (29.92%). Chlorophyll and carotenoids in Echinochloa leaf extract likely contribute to the positive effect on photosynthetic pigments of weed leaf extract. In Moringa leaves, magnesium can be found as a chlorophyll component and high levels of cytokinins contribute to chlorophyll synthesis. Because of this, basil leaves possessed a healthy dark green color and provided a boost to photosynthetic pigments. These findings are complemented by Rady et al., [20], Ashraf et al., [21] and Aslam et al., [22].

Table 1. Effect of foliar spray of biostimulants on growth attributes of sweet basil

Treatments	Height (cm)		Canopy (cm)		No. of Leaves pl^{-1}		No. of Branches pl^{-1}		Leaf Area Index (LAI)	
	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage
Control	27.3^{a}	61.5^{ab}	62.8^{a}	108.8^{bcd}	165.5^{a}	368.3^{ab}	8.5^{ab}	22.0^{ab}	0.98^{abc}	1.30^{abc}
0.5% Seaweed Extract	31.8^{a}	63.0^{abc}	63.3^{a}	106.3^{abcd}	185.3^{bc}	396.5^{b}	8.5^{ab}	22.3^{ab}	1.18^{de}	1.28^{ab}
1.0% Seaweed Extract	29.5^{a}	61.0^{a}	69.5^{ab}	90.0^{a}	171.8^{ab}	458.5^{d}	7.5^{a}	20.8^{ab}	0.88^{abb}	1.38^{abcde}
1.5 % Seaweed Extract	28.5^{a}	63.0^{abc}	77.5^{bcd}	101.3^{abc}	181.8^{bc}	390.8^{ab}	8.5^{ab}	25.0^{abc}	1.10^{cd}	1.50^{de}
0.5% *Cyperus rotundus* Extract	28.5^{a}	60.5^{a}	79.8^{cd}	96.3^{ab}	192.8^{cd}	407.0^{bc}	8.0^{a}	21.5^{ab}	0.83^{a}	1.25^{a}
1.0% *Cyperus rotundus* Extract	29.3^{a}	65.0^{abc}	72.0^{bcd}	98.8^{ab}	196.5^{cd}	364.3^{a}	7.0^{a}	19.0^{a}	0.98^{abc}	1.45^{bcde}
1.5 % *Cyperus rotundus* Extract	30.3^{a}	62.8^{abc}	74.5^{bcd}	111.3^{bcd}	208.8^{de}	460.3^{d}	9.8^{ab}	26.0^{abcd}	1.00^{bc}	1.38^{abcde}
0.5% *Echinochloa colona* Extract	28.0^{a}	66.3^{bc}	69.8^{ab}	117.5^{cd}	186.8^{bc}	463.0^{d}	7.5^{a}	35.5^{ef}	1.13^{d}	1.55^{e}
1.0% *Echinochloa colona* Extract	30.3^{a}	66.0^{bc}	70.8^{abc}	120.0^{d}	198.5^{cde}	387.8^{ab}	11.5^{b}	32.0^{cdef}	1.30^{e}	1.53^{e}
1.5% *Echinochloa colona* Extract	30.8^{a}	66.0^{bc}	80.8^{d}	112.5^{bcd}	186.0^{bc}	388.8^{ab}	6.5^{a}	28.0^{bcde}	1.00^{bc}	1.33^{abcd}
0.5% *Moringa oleifera* Extract	29.3^{a}	63.8^{abc}	76.3^{bcd}	108.8^{bcd}	205.5^{cde}	428.8^{c}	7.3^{a}	30.3^{cdef}	1.18^{de}	1.50^{de}
1.0% *Moringa oleifera* Extract	27.8^{a}	67.0^{c}	72.5^{bcd}	108.8^{bcd}	213.8^{e}	406.3^{bc}	8.5^{ab}	32.8^{def}	0.98^{bc}	1.48^{cde}
1.5 % *Moringa oleifera* Extract	31.0^{a}	66.3^{bc}	72.8^{bcd}	101.3^{abc}	214.3^{e}	396.5^{b}	8.8^{ab}	33.8^{def}	1.18^{de}	1.53^{e}

Note: No significant difference between the treatments when the same letter follows the mean in the same column (DMRT)

Table 2. Influnce of foliar spray of biostimulants on plant biochemical attributes of basil variety CIM-Shisir

Treatments	Total Chlorophyll (mg g^{-1} fresh weight)		Chlorophyll Fluorescence (fv fm^{-1})		Proline (µ mol g^{-1} fresh weight)		Relative Water Content of Leaves (RWC %)	
	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage	Growth Stage	Harvesting Stage
Control	0.92^{a}	0.50^{cde}	0.528^{b}	0.343^{b}	0.177^{e}	0.180^{c}	46.98^{a}	29.92^{a}
0.5% Seaweed Extract	1.08^{abc}	0.52^{cde}	0.653^{de}	0.461^{de}	0.103^{cd}	0.104^{a}	49.02^{a}	41.40^{abc}
1.0% Seaweed Extract	1.03^{ab}	0.71^{f}	0.710^{f}	0.490^{efg}	0.062^{abc}	0.114^{ab}	50.03a	48.53^{bcde}
1.5 % Seaweed Extract	1.03^{ab}	0.57^{cdf}	0.711^{f}	0.498^{fg}	0.031^{a}	0.102^{a}	55.42^{b}	45.63^{bcde}
0.5% *Cyperus rotundus* Extract	1.24^{cde}	0.62^{ef}	0.659^{e}	0.451^{d}	0.104^{cd}	0.130^{b}	60.93^{c}	51.47^{cde}
1.0% *Cyperus rotundus* Extract	0.95^{a}	0.36^{abe}	0.669^{e}	0.468^{def}	0.102^{cd}	0.131^{b}	61.90^{c}	34.81^{ab}
1.5 % *Cyperus rotundus* Extract	1.33^{de}	0.47^{bcd}	0.577^{c}	0.363^{b}	0.052^{ab}	0.168^{c}	62.39^{c}	59.13^{e}
0.5% *Echinochloa colona* Extract	1.18^{bcd}	0.39^{ab}	0.648^{de}	0.399^{c}	0.103^{cd}	0.248^{d}	67.52^{de}	55.40^{de}
1.0% *Echinochloa colona* Extract	1.32^{de}	0.32^{a}	0.645^{de}	0.458^{de}	0.116^{d}	0.189^{c}	70.50^{ef}	42.9^{abcd}
1.5% *Echinochloa colona* Extract	1.33^{de}	0.49^{bcde}	0.610^{cd}	0.367^{bc}	0.092^{d}	0.176^{c}	73.38^{f}	37.96^{abc}
0.5% *Moringa oleifera* Extract	1.04^{abc}	0.55^{cde}	0.483^{a}	0.258^{a}	0.134^{de}	0.177^{c}	79.88^{g}	40.82^{abc}
1.0% *Moringa oleifera* Extract	1.43^{e}	0.59^{cdef}	0.640^{de}	0.433^{d}	0.118^{d}	0.170^{c}	61.36^{c}	55.27^{d}
1.5 % *Moringa oleifera* Extract	1.31^{de}	0.45^{bcd}	0.719^{f}	0.505^{g}	0.040^{a}	0.092^{a}	63.72^{cd}	37.99^{abce}

Note: No significant difference between the treatments when same letter follows the mean in the same column (DMRT)

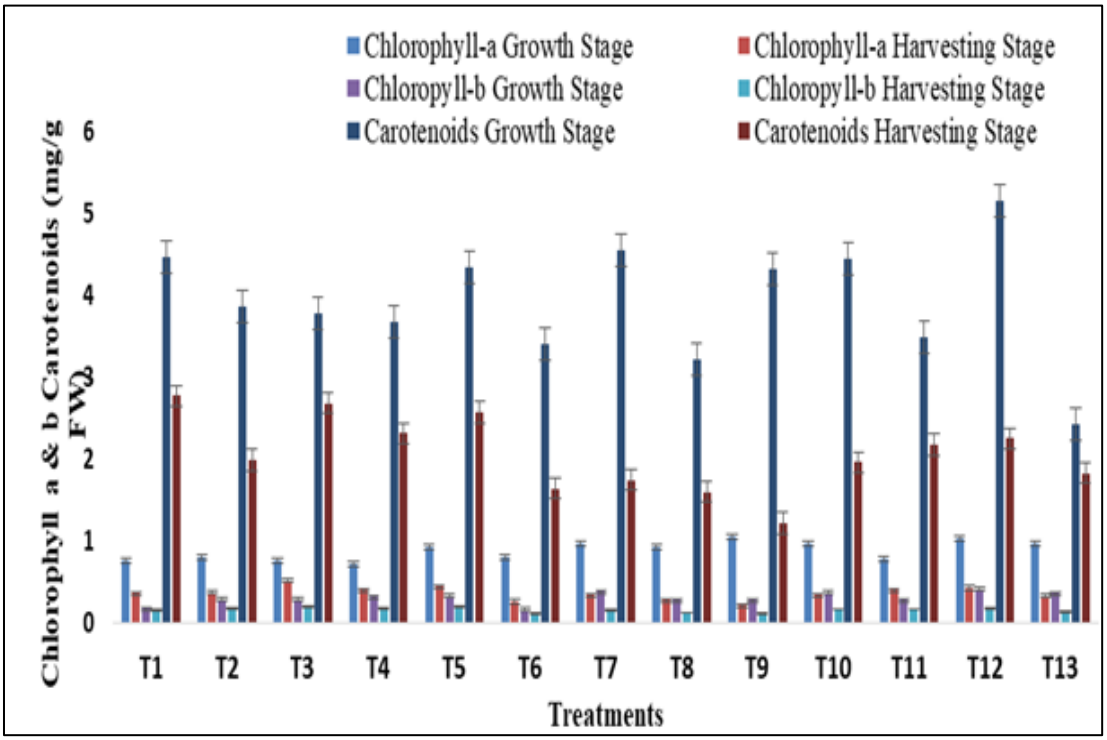

Figure 2. Chlorophyll-a and b (Growth and Harvesting stage) and carotenoids content

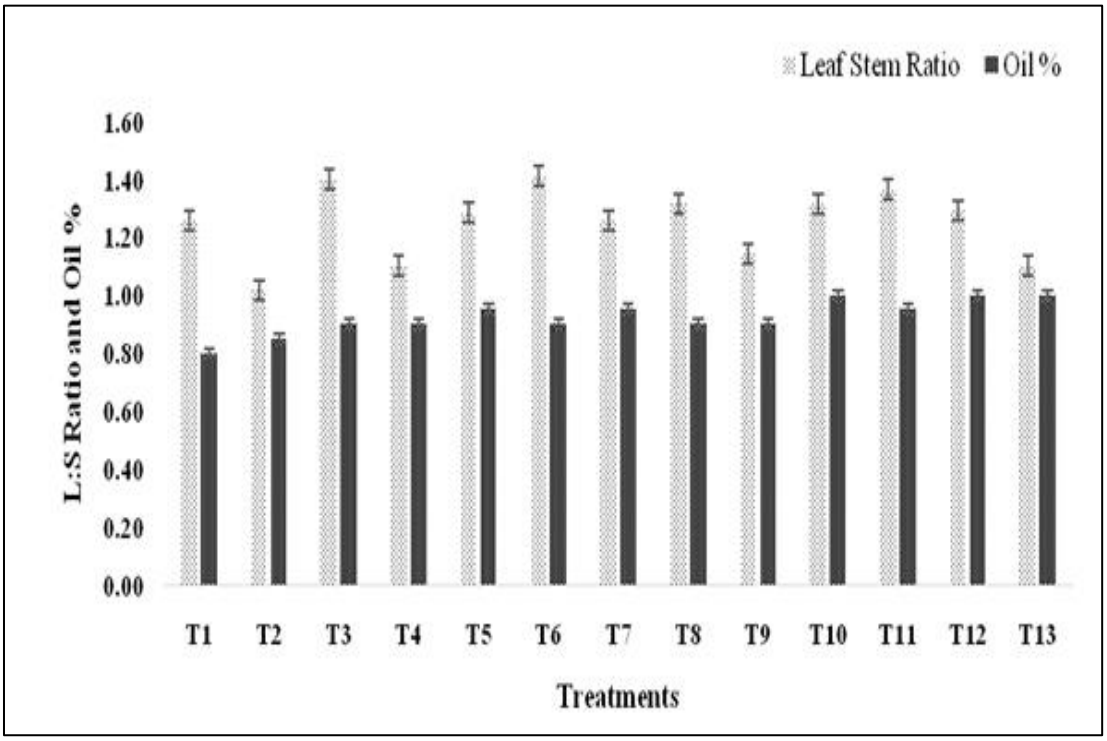

Figure 3. Leaf stem ratio and oil content

Influence of bio-stimulants on yield attributes and essential oil yield of basil

Different biostimulants substantially increased basil fresh herb and essential oil yields. All the treatments gave higher essential oil yield in comparison to the control (Table 3). Similarly leaf to stem (L:S) ratio and content of essential oil were also displayed significant enhancement over control due to the foliar application of botanical biostimulants (Figure 3). Among all the treatments 1.0% *Echinochloa colona* extract gave significantly highest fresh biomass (266 grams per pot), dry biomass (89.7 grams per pot), fresh root (48.5 g per pot), dry root (20.0 gram per pot), and essential oil yield (2.4 ml/pot), while in case of shoot root (S:R) ratio 1.0% *Moringa oleifera* and 1.5% *Moringa oleifera* gave significantly highest value (6.2, 6.3). Control gave lowest fresh biomass (163.5 grams per pot), dry biomass (55.6 grams per pot), fresh root (30 grams per pot), dry root (11.6 grams per pot), and essential oil yield (1.3 ml/pot). In the case of a shoot root (S:R) ratio of 1.0% seaweed extract gave the lowest value (4.4). The results of the present study show that *Echinochloa colona* extract has very high antioxidant activity and is also rich in micro and macro nutrients which may be the reason for to enhanced biomass yield significantly. Increased crop biomass under different biostimulant treatments is one of the reasons for higher essential oil yields. Similar findings were previously reported by Mazeed et al., [19].

Table 3. Influence of foliar spray of bio-stimulants on yield attributes of Tulsi variety CIM-Shisir

Treatments	Fresh biomass g/pot	Dry biomass g/pot	Fresh root g/pot	Dry root g/pot	Shoot root (S:R) ratio	Oil yield ml/pot
Control	163.5^{a}	55.6^{a}	30.0^{a}	11.6^{a}	5.6^{abc}	1.3^{a}
0.5% Seaweed Extract	216.0^{g}	71.3^{e}	49.0^{d}	19.2^{d}	4.6^{ab}	1.8^{c}
1.0% Seaweed Extract	174.5^{abc}	57.1^{ab}	40.0^{abcd}	16.3^{bcd}	4.4^{a}	1.6^{b}
1.5 % Seaweed Extract	209.8^{f}	69.7^{e}	47.0^{cd}	18.7^{d}	4.6^{ab}	1.9^{c}
0.5% *Cyperus rotundus* Extract	190.8^{d}	64.3^{cd}	32.5^{ab}	13.5^{ab}	6.0^{cd}	1.8^{c}
1.0% *Cyperus rotundus* Extract	180.0^{bcd}	60.7^{bc}	32.5^{ab}	12.9^{ab}	5.7^{abc}	1.6^{b}
1.5 % *Cyperus rotundus* Extract	203.0^{ef}	67.7^{de}	36.3^{ab}	13.5^{ab}	5.7^{abc}	1.9^{c}
0.5% Echinochloa colona Extract	186.5^{cd}	62.8^{c}	38.0^{abc}	15.7^{bcd}	4.9^{abc}	1.7^{b}
1.0% *Echinochloa colona* Extract	266.0^{h}	89.7^{f}	48.5^{d}	20.0^{e}	5.6^{abc}	2.4^{e}
1.5 % *Echinochloa colona* Extract	206.0^{fg}	69.4^{e}	43.0^{bcd}	17.5^{cd}	5.0^{abc}	2.1^{d}
0.5% *Moringa oleifera* Extract	172.5^{ab}	57.2^{ab}	35.3^{ab}	14.6^{abc}	5.1^{abc}	1.6^{b}
1.0% *Moringa oleifera* Extract	187.0^{d}	63.0^{c}	30.3^{a}	12.5^{ab}	6.2^{c}	1.9^{c}
1.5 % *Moringa oleifera* Extract	192.5^{d}	63.6^{cd}	31.3^{a}	13.2^{ab}	6.3^{c}	1.9^{c}

Note: No significant difference between the treatments when same letter follows the mean in the same column (DMRT)

Influence of biostimulants on the volatile compounds of basil

The principal volatile components of basil cultivar CIM-Shishir are Linalool, camphor, and 1,8 Cineole. Linalool content varies from 63.2% to 68.1%, camphor varies from 9.5% to 10.3%, and 1,8 Cineole varies from 7.1% to 8.5% under different biostimulant treatments (Figure 4). Moringa leaf extract gave higher results for these volatile compounds. In part, these results may be explained by the phytohormones, amino acids, and nutrients present in *Moringa oleifera* leaves, which enable the aggregation of specialized metabolites, consisting of volatile components of the oil. These findings were similar to those of Hazzoumi et al., [23], and Aftab et al., [24].

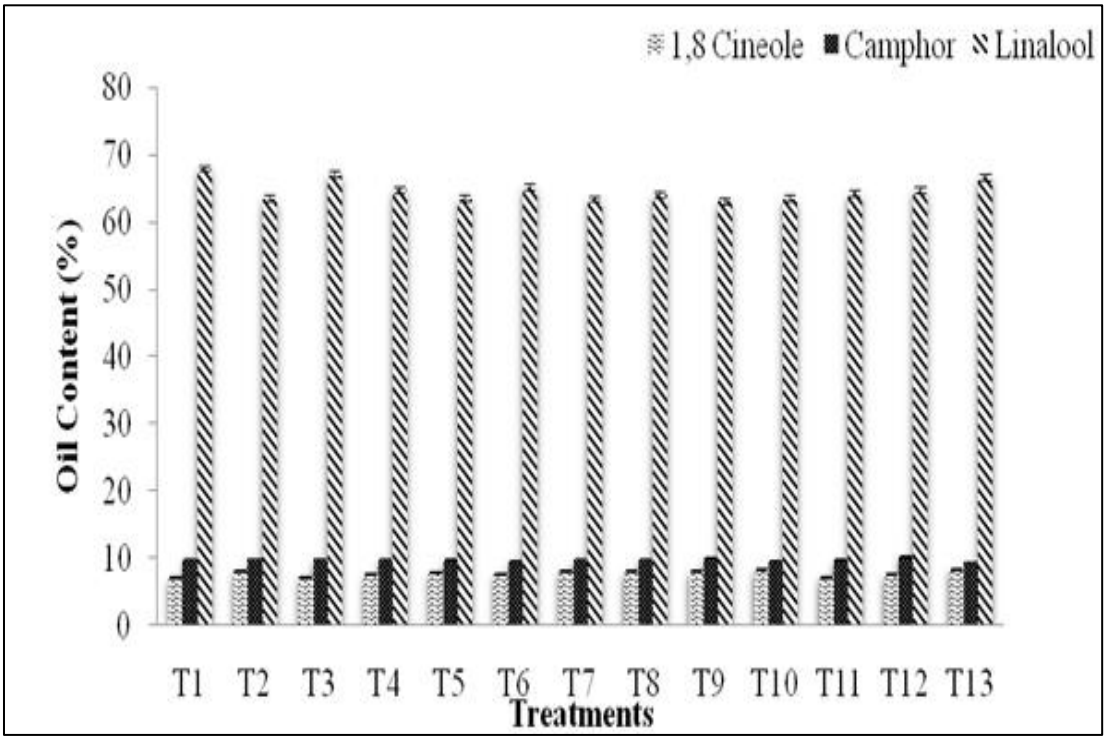

Figure 4. Major constituents of *Ocimum basilicum* essential oil as influenced by the treatments

Conclusion

It can be concluded from the present study that as impactful as Moringa Leaf Extract is and has been found in various of the previous studies, *Echinochloa colona* extract gave the highest oil yield with a good amount of volatile compounds in it, while other weed leaf extracts at different concentration have also positively influenced the growth and yield of basil. Not much work has been done on these weed leaf extracts previously therefore further research needs to be conducted to observe the impacts of these weed leaf extracts on basil as well as other crops to confirm these findings.

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