

Research Article

Foliar application of *Ascophyllum nodosum* on improvement of photosynthesis, fruit setting percentage, yield and quality of tomato (*Solanum lycopersicum* L.)

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Abstract

In recent days, liquid formulations of brown seaweed extract, *Ascophyllum nodosum* used as a biostimulant in agriculture. Various studies suggest that *A. nodosum* enhanced the growth and yield of agriculturally important crops, but still, there is a lack of information about the biostimulation effects on photosynthesis, flowering and fruit setting of tomato. Hence, the present study aimed to know the effect of foliar application of *A. nodosum* on photosynthesis, flowering, fruit setting, yield and quality of tomato. A biostimulant product, MC Set with *A. nodosum* extract applied to tomato as a foliar spray at rates of three different concentrations such as 1.0 L ha⁻¹ (MS 1), 2.0 L ha⁻¹ (MS 2), 3.0 L ha⁻¹ (MS 3) for six times during flowering of 2nd (30 Days after transplanting – DAT), 3rd (40 DAT) and 4th (50 DAT) cluster and fruit setting of 2nd (60 DAT), 3rd (70 DAT) and 4th (80 DAT) cluster respectively. The MC Set treatments enhanced the plant photosynthesis, flower number and fruit number per cluster, yield and quality traits of tomato. However, the middle concentration MS 2 showed highest photosynthetic rate, stomatal conductance, SPAD value, flower and fruit in 2nd, 3rd and 4th cluster. It also had better average fruit weight and yield per plant and hectare and enhanced the quality parameters such as total soluble solids, ascorbic acid content, lycopene and total sugars compared to control and other two concentrations of MS Set. Hence, using *A. nodosum* extract on tomato growth could be a better sustainable crop production method.

Keywords: *Ascophyllum nodosum*, Flowering, Fruit setting, MC set, Photosynthesis, Quality, Yield

INTRODUCTION

Tomato is the world's second most important vegetable crop grown for its dietary and commercial importance and provides a good amount of vitamins A and C, minerals and antioxidants to the human diet (Farooq *et al.*, 2020). Due to the limited amount of cultivable land and the dramatic rise in global population (Rodriguez and Sanders, 2015), there is a need for higher crop production with better crop management approaches (Godfray *et al.*, 2010). By 2050, global food production must double to meet the constantly rising demands of the expanding world population (Voss-Fels and Snowdon, 2016). To meet this demand for higher crop production, the growers use more quantity of synthetic agrochemicals *viz.* inorganic fertilizers and chemical pesticides which are harmful to the environment, soil and human health (Damalas and Koutroubas, 2016), so it is challenging to increase agricultural productivity by minimizing environmental damage. Hence, a productive biological substitute is needed to minimize the use of synthetic agrochemicals.

In this context, plant and microbial based biostimulants are better ways to replace the fertilizers and pesticides, which could enhance crop production without affecting the environment (Van Oosten *et al.*, 2017; Yakhin *et al.*, 2017). Plant biostimulants are microorganisms or/and substances. It may contain seaweed extracts, amino acids, yeast extracts, vitamins, protein hydrolysates, *etc.*; which are given to the crop as foliar or to the rhizosphere, triggering the metabolic activities to increase nutrient uptake, enhance nutrient use efficiency and crop quality and it also imparts tolerance against many abiotic stresses and they are receiving greater awareness in global market (Calvo *et al.*, 2014; Halpern *et al.*, 2015; Du Jardin *et al.*, 2015; Koleska *et al.*, 2017; Rouphael *et al.*, 2018).

Nowadays seaweeds are effectively utilized as biostimulants, seaweeds are macroscopic, multi-cellular organisms that live in coastal habitats and they contain a substantial number of alginates, polysaccharides, enzymes, biologically active peptides and some polyunsaturated fatty acids (PUFAs) (Meng *et al.*, 2023; Ahmadi *et al.*, 2015; Yakhin *et al.*, 2017; Ali *et al.*, 2021; Battacharyya *et al.*, 2015; Khan *et al.*, 2009; Okolie *et al.*, 2018; Shukla *et al.*, 2019). The brown seaweed extract *Ascophyllum nodosum* had enhanced the growth, nutrient use, yield and crop quality of various crops (Shukla *et al.*, 2019; Khan *et al.*, 2009; Ali *et al.*, 2016; Ali *et al.*, 2016; Campobenedetto *et al.*, 2021). Some researches explained the potential Biostimulation effects of *A. nodosum* on numerous horticultural and crops, including tomato (Subramaniyan *et al.*, 2023; Di Mola *et al.*, 2023; Dookie *et al.*, 2020; Ahmed *et al.*, 2022; Ikuyinminu *et al.*, 2022; Ali *et al.*, 2022), maize (Shukla *et al.*, 2021; Basavaraja *et al.*, 2018), broccoli

(Kaluzewicz *et al.*, 2017), spinach (Castronuovo *et al.*, 2023), avocado (Arioli *et al.*, 2023), okra (Ali *et al.*, 2022), pea (Rashad *et al.*, 2022), wheat (Langowski *et al.*, 2022), soybean (Repke *et al.*, 2022). The micronutrients boron and zinc play important roles in improving fertilization, flower and fruit setting in crops such as tomato (Ali *et al.*, 2015; Francesca *et al.*, 2020), okra (Rahman *et al.*, 2020).

Furthermore, a number of findings indicate that foliar application of *A. nodosum* had positive impact on plant growth, yield and quality of tomato, but the effect of foliar application of *A. nodosum* on photosynthesis, flowering, fruit setting of tomato is remained scared and should be explore further. Based on this view, the present hypothesis was fixed, the seaweed extract (*A. nodosum*) containing biostimulant product MC Set might enhance the photosynthesis, flowering and fruit setting of tomato thereby it increasing the yield and quality. The present study used the biostimulant product MC Set as foliar application to assess their biostimulation effects on photosynthesis, flowering, fruit setting, yield and quality of tomato at open field conditions.

MATERIALS AND METHODS

Planting and biostimulant materials

Tomato (Hybrid *Shivam*) seeds were purchased from Rasi Seeds Pvt. Ltd., Salem, Tamil Nadu and the seeds were sowed in portrays containing vermicompost and coir pith media with a ratio of 1:3. After the germination of seeds, young seedlings were watered frequently by using rose can and the water-soluble nutrients (19:19:19 NPK) were sprayed on leaves twice in a week. Then, 23 days old seedlings with 4 to 5 leaves were transplanted into the main field at 60 x 45 cm spacing. The world's leading biostimulant company M/s. Valagro BioSciences Ltd., Hyderabad, provided the biostimulant product MC Set.

Experimental design and treatment details

The experiment was conducted at Eastern Block Farm, Department of Agronomy, Tamil Agricultural University, Coimbatore, from February 2023 - June 2023. The field trial was conducted to assess the effect of foliar application of MC Set on physiology, fruit setting, yield and quality of tomato. The field is situated at an elevation of 426.7 m above mean sea level and 110° N latitude and 770° E longitude. The experiment was conducted with Randomized Block Design (RBD) with six replications and four treatments. Physio-chemical properties of the experimental soils were analysed and the results are given in Table 1. About 45-50 tomato plants were maintained in each replication with a plot size of 15 m². Irrigation was given at weekly intervals based on the requirements and NPK @ 50:250:100 kg ha⁻¹ and zinc sulphate 50 kg ha⁻¹ were applied as basal dose. Then,

Table 1. Physio-chemical properties of experimental field soil

Soil properties	Range
Texture	Clay loam
pH	7.57
Electrical conductivity (ds m ⁻¹)	0.66
Soil organic carbon (%)	0.69
Available Nitrogen (Kg ha ⁻¹)	275
Available phosphorous (Kg ha ⁻¹)	38
Available potassium (Kg ha ⁻¹)	975

a top dressing of 50 kg ha⁻¹ of each N and K was applied at 30, 45, and 60 days after transplanting (DAT). For weed control, 25 mL 10⁻¹ L of pre-emergence herbicide pendimethalin was sprayed at two DAT.

The composition of biostimulant product MC Set with seaweed, *Ascophyllum nodosum* extract supplemented with boron (B) and zinc (Zn). The experiment has four treatments, including control (CT), and three different concentrations of MC Set biostimulant included MC Set 1.0 L ha⁻¹ (MS 1), MC Set 2.0 L ha⁻¹ (MS 2), and MC Set 3.0 L ha⁻¹ (MS 3). The MC Set biostimulant solution was prepared by mixing 1.5 mL (MS 1), 3.0 mL (MS 2) and 4.5 mL (MS 3) of MC Set and 750 mL of water and uniformly sprayed on the leaf surface. The physiological parameters *viz.*, SPAD value, photosynthetic rate, and stomatal conductance were measured 5 days after the first (35 DAT), second (45 DAT) and third spray (55 DAT) at the flowering of 2nd, 3rd and 4th clusters, respectively. The physiological parameters were taken from three plants (3 leaves from each plant) in each replication. The flower and fruit numbers per cluster were counted at the flowering and fruit-setting stages. The yield and quality parameters, *viz.*, total soluble solids, ascorbic acid, lycopene content and total sugars of tomato were also measured from 3 plants in each replication.

Physiological parameters

Portable chlorophyll meter, SPAD (Soil-plant analytical development) Model 5020 Minolta (Konica Minolta, INC, Tokyo, Japan), was used to measure the chlorophyll index of tomato plants of physiologically active leaves. Portable photosynthesis system (PPS; LI-6400 XT, Licor Inc., Lincoln, NE, USA) was used to measure the photosynthetic rate (Pn) and stomatal conductance (gs) of tomato plants at 5 days after first, second and third spray. These observations were made between 10:00 am to 12:00 noon on a clear sunny day. The plant's third leaf from the top (physiologically active leaf) was selected and inserted in the IRGA (Infra Red Gas Analyser) chamber to determine the gas exchange traits. Before inserting the leaf, photosynthetically active radiation was set to greater than 1000 μmol photons m⁻² s⁻¹ and matched the CO₂ concentration inside the chamber with the ambient CO₂ concentration. Then,

relative humidity was set at a stable level equal to the ambient relative humidity to reproduce a state which is similar to that of ambient air. The photosynthetic rate and stomatal conductance were expressed as μmol CO₂ m⁻² s⁻¹ and mol H₂O m⁻² s⁻¹, respectively.

Flower number, fruit number and fruit setting percentage

Flower and fruit numbers in 2nd, 3rd and 4th clusters were counted manually and the fruit setting percentage was computed and expressed as a percentage (%).

$$\text{Fruit setting percentage (\%)} = \frac{\text{Fruit number}}{\text{Flower number}} \times 100 \quad \text{Eq. 1}$$

Yield parameters

Tomato fruits were manually harvested weekly twice by handpicking uniformly ripened red-coloured fruits. The first picking was started from 70 DAT. The number of fruits per plant, average fruit weight (g), and yield per plant (kg) were measured at each harvest and expressed in the cumulative value of all the harvests and it was used to calculate yield per hectare (t ha⁻¹).

Quality parameters

The tomato fruit quality traits were analysed using physiologically ripened three equal-sized fruits from each plant. ERMA hand refractometer (0-32 °C) was used to record the total soluble solids (TSS) of tomato fruits by following the protocol of Tigist *et al.* 2013. It was determined by placing a few drops of tomato juice on prism of the Refractometer and it was expressed as °Brix.

Ascorbic acid content was analysed by titration method using the procedure of Ikewuchi *et al.* 2011. About 10 mL of 4% oxalic acid and 5 mL of working standard (0.1% ascorbic acid) were taken together and titrated against the 2,6-dichlorophenol indophenol dye until the appearance of pink color as an endpoint, noted the amount of dye consumed as V1. Then, 500 mg of macerated tomato fruit sample mixed with 4% oxalic acid and the volume made up to 100 mL by using distilled water. The above mixture was centrifuged at 3000 rpm for 15 min. After that, 5 mL of supernatant and 10 mL of 4% oxalic acid were mixed and titrated against the dye and noted the volume as V2 at the appearance of pink color. It was denoted as mg 100 g⁻¹ fruit.

$$\text{Amount of ascorbic acid in the sample} = \frac{0.5}{V1} \times \frac{V2}{5} \times \frac{100}{0.5} \times 100 \quad \text{Eq. 2}$$

Lycopene content of tomato fruit was analysed by following the procedure of Ranganna, 1986. About 1 g of fruit sample was macerated with 5 mL of acetone and extracted by adding 20 mL of 5% sodium sulphate and 20 mL of petroleum ether to sample in a separating funnel. The mixture was shaken well and separate the petroleum ether layer after incubation. Again, re-

extracted the lower aqueous phase with 20 mL of petroleum ether. Then, extract was mixed with 10 g of anhydrous sodium sulphate and kept for 30 min. The petroleum ether extract was collected in a 25 mL volumetric flask and the absorbance was measured at 503 nm in Spectrophotometer. Lycopene content was denoted as mg 100 g⁻¹.

$$\text{Lycopene content} = \frac{3.12 \times \text{Absorbance value} \times \text{Total volume} \times 100}{\text{Weight of sample (g)} \times 1000}$$

Eq. 3

Absorbance (1 unit) = 3.12 mg lycopene/mL

Total sugars in tomato fruits were quantified by following the Anthrone method of Hedge and Hofreiter (1962). About 250 mg of fruit sample was extracted with 10 mL of 80% ethanol and centrifuged, the content at 6000 rpm for 10 min. 0.5 mL of supernatant was collected and incubated in Water bath for 30 min. Then, 4 mL of anthrone reagent and 1 mL of distilled water were added into a test tube, kept in a water bath for 10 min, and read the absorbance at 630 nm in Spectrophotometer. It was expressed in mg 100 g⁻¹ of the sample.

$$\text{Total sugars present in sample} = \frac{X}{0.5} \times \frac{10}{0.25} \times 1$$

Eq. 4

X = Absorbance at 630 nm

Statistical analysis

The statistical analysis for experimental data was performed separately for each stage of observation by using SPSS software (version 16.0). One-way ANOVA (Analysis of variance) was done for all the parameters and the results were presented as mean with standard error. The mean values of four treatments were ranked using Duncan's multiple range test (DMRT) at $p = 0.05$. GraphPad Prism (version 8.2.0) was used for data visualization of the recorded traits. The experimental data was subjected to SPSS software for Pearson correlation analysis to know the relationship between MC Set bio-stimulant treatments and physiological traits, fruit setting, yield and quality of tomato plants.

RESULTS AND DISCUSSION

Effect of MC Set on physiological traits of tomato plants

The chlorophyll is the primary pigment involved in photosynthesis and the pigment accumulation in the leaf is represented as SPAD value. The foliar spray of three doses of MC Set treatments had a significant effect on the chlorophyll index of tomato plants (Fig.1). Among the doses, the MS 2 (MC Set 2.0 L ha⁻¹) efficiently increased the chlorophyll index by 14.36%, 16.21% and 17.77% at 5 days after first, second and third spray respectively (flowering of 1st, 2nd, and 3rd cluster). The MC Set biostimulant significantly increased the SPAD value of tomato plants in all three stages and similar

results were suggested by Subramaniyan *et al.* (2023) for an increase in SPAD by soil application of *A. nodosum* and plant extract-derived product (Kendal root) in tomato. The *A. nodosum* extract increases the SPAD value of tomato plants (Hussain *et al.*, 2021; Ali *et al.*, 2019), it is because of reduced senescence of leaf, higher uptake of nitrogen, and reduced activity of chlorophyllase, a chlorophyll degrading enzyme (Lucia *et al.*, 2022).

Photosynthesis efficiency determines the growth and yield potential of the crops. In the current study three doses of MC Set biostimulant significantly enhanced the photosynthetic performance of tomato plants compared to the control (Fig. 1). However, the treatment MS 2 (MC Set 2.0 L ha⁻¹) recorded a maximum photosynthetic rate of 28.72, 30.47 and 31.13 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ at 5 days after first, second and third spray respectively. Similar results were found by Subramaniyan *et al.* (2023), who suggested the soil drenching of Kendal root biostimulant containing *A. nodosum* significantly raised the photosynthetic rate of tomato plants. The *A. nodosum* based biostimulant increases the photosynthetic activity of spinach (Castronuovo *et al.*, 2023) and broccoli (Kaluzewicz *et al.*, 2017). The *A. nodosum* extract prevents the photosynthetic machinery from photo-damage, dissipates the excess energy in the PS II, and increases the intrinsic water use efficiency, stomatal modulation, and activation of antioxidant system, which leads to a higher photosynthetic rate (Santaniello *et al.*, 2017).

Stomatal conductance (gs) is directly related to the size of the stomatal aperture. In the present study, the stomatal conductance of tomato plants also positively improved by foliar application of MC Set biostimulants (Fig. 1). The treatment MS 2 (MC Set 2.0 L ha⁻¹) had more impact on stomatal conductance of 0.70, 0.77 and 0.81 mol H₂O m⁻² s⁻¹ at 5 days after first, second and third spray respectively. Pearson correlation analysis confirmed (Table 2) that foliar spraying of MC Set biostimulant positively influenced the chlorophyll index (SPAD; $r^2 = 0.874$), photosynthetic rate (Pn; $r^2 = 0.791$) and stomatal conductance (gs; $r^2 = 0.762$). The higher stomatal conductance of tomato plants treated with *A. nodosum* containing biostimulant was also found by Subramaniyan *et al.*, 2023 and it is due to the *A. nodosum* extract enhanced plant water relations, which leads to high turgor pressure and decreased stomatal closure and higher photosynthesis (Urban *et al.*, 2017; Kaluzewicz *et al.*, 2017). Similarly, the *A. nodosum* filtrate enhances the stomatal conductance even under drought conditions in broccoli (Kaluzewicz *et al.*, 2017).

Influence of MC Set on flowering, fruit setting of tomato plants

The effect of foliar spray of three doses of MC Set biostimulant on flowering, fruit setting and fruit setting

Table 2. Pearson correlation between MC Set biostimulant and physiological and yield related variables of tomato

Variables	BS	Pn	gs	SPAD	FNPC	FrNPC	FSP	FrNPP	AFW	YieldP P	YieldP H	TSS	AA	Lycopene	TS
BS	1	0.791**	0.762**	0.874**	0.654**	0.766**	0.679**	0.682**	0.804**	0.663**	0.702**	0.763**	0.683**	0.683**	0.781**
Pn		1	0.967**	0.974**	0.684**	0.969**	0.870**	0.766**	0.893**	0.776**	0.791**	0.896**	0.854**	0.854**	0.923**
Gs			1	0.948**	0.727**	0.921**	0.793**	0.736**	0.846**	0.727**	0.770**	0.832**	0.760**	0.760**	0.882**
SPAD				1	0.685**	0.960**	0.876**	0.811**	0.912**	0.787**	0.833**	0.878**	0.828**	0.828**	0.936**
FNPC					1	0.651**	0.416*	0.557**	0.680**	0.447*	0.630**	0.560**	0.625**	0.625**	0.712**
FrNPC						1	0.941**	0.808**	0.892**	0.809**	0.780**	0.860**	0.868**	0.868**	0.938**
FSP							1	0.758**	0.822**	0.827**	0.708**	0.804**	0.812**	0.812**	0.861**
FrNPP								1	0.836**	0.645**	0.751**	0.731**	0.671**	0.671**	0.717**
AFW									1	0.753**	0.793**	0.799**	0.836**	0.836**	0.853**
YieldPP										1	0.592**	0.724**	0.745**	0.745**	0.765**
YieldPH											1	0.674**	0.641**	0.641**	0.748**
TSS												1	0.849**	0.849**	0.849**
AA													1	1.000**	0.855**
Lycopene														1	0.855**
TS															1

** Correlation is significant at the 0.01 level (2-tailed); * Correlation is significant at the 0.05 level (2-tailed); biostimulant (bs), photosynthetic rate (Pn), stomatal conductance (gs), chlorophyll index (SPAD), flower number per cluster FNPC), fruit number per cluster (FrNPC), fruit setting percentage (FSP), fruit number per plant (FrNPP), average fruit weight (AFG), yield per plant (YieldPP), yield per hectare (YieldPH), total soluble solids (TSS), ascorbic acid (AA), lycopene (Lycopene), total sugars (TS).

percentage of 2nd, 3rd and 4th clusters of tomato plants is shown in Fig. 2. The MC Set treatments did not have any significant effect on flowering of 2nd cluster, since the spraying was started at the flowering of 2nd cluster, but the MC set treatments significantly increased the flower number in 3rd and 4th cluster. The treatment MS 2 (MC Set 2.0 L ha⁻¹) recorder higher flower number in both 3rd cluster (13.00) and 4th cluster (12.83). MC Set treatments enhanced the fruit number per cluster. However, the MS 2 (MC Set 2.0 L ha⁻¹) concentration had a more positive impact on fruit number per cluster at 2nd (10.83 fruits), 3rd (11.00 fruits) and 4th (11.00 fruits) clusters.

The foliar spray of three MC Set treatments significantly improved the fruit setting percentage of tomato plants over a control. The middle concentration MS 2 (MC Set 2.0 L ha⁻¹) had the highest fruit setting percentage of 84.87%, 84.98% and 85.96% at 2nd, 3rd and 4th cluster, respectively. The MC Set biostimulant treatment positively correlated with flower number per cluster (FNPC; $r^2 = 0.654$), fruit number per cluster (FrNPC; $r^2 = 0.766$) and fruit setting percentage (FSP; $r^2 = 0.679$; Table 2). The application of *A. nodosum* extract on tomato plants increased the flower number and fruit number per cluster, and thereby it increases the fruit setting percentage (Hussain *et al.*, 2021; Ali *et al.*, 2019; Dookie *et al.*, 2020; Ali *et al.*, 2016; Renaut *et al.*, 2019)). The *A. nodosum* extract stimulates flowering by enhancing plant growth and it promotes early reproductive growth

by switching from vegetative growth and increasing flower number (Ali *et al.*, 2016).

A substantial quantity of growth-promoting hormones like auxin, gibberellin and cytokinin is present in seaweed extracts that stimulate plant growth and increase tomato's flowering and fruit setting (Ali *et al.*, 2016). This increased flowering and fruit setting may be due to increased photosynthesis, delaying of senescence, enhanced nutrient uptake and phytohormone levels (Khan *et al.*, 2009). Dookie *et al.* (2020) found that foliar application of brown seaweed extract *A. nodosum* directly or indirectly enhanced the expression levels of major flowering genes *viz*, Constans -1 (CO), Single Flower Truss (SFT), Anantha (AN), Self – Pruing (SP), Jointless (J) and Falsiflora (FA) in flower buds and apical meristems of tomato which can trigger the flowering. The micronutrients boron and zinc present in the MC Set enhances the flowering and fruit setting by improving fertilization (Rahman *et al.*, 2020). The boron and zinc increased the flowering and fruit setting of mandrins; zinc plays a vital role in pollen tube growth and boron is involved in pollen germination and also in pollen tube growth which results in enhanced flowering and fruit setting (Ruchal *et al.*, 2020). The boron and zinc nutrients delay the abscission of flowers, thereby it improves the fruit setting and provides the required quantity of carbohydrates needed for flowering (Ullah *et al.*, 2015).

Effect of MC Set on yield of tomato plants

Yield is a polygenic factor influenced by many external and internal traits. It is the complexation of plant growth, biochemical and physiological traits; yield is not solely dependent on photosynthetic rate but also on the assimilate Partitioning efficiency. The positive impact of MC Set biostimulant spray on yield parameters of tomato is presented in Fig. 3. The fruit number per plant

was significantly increased in MC Set treatments compared to control. The highest number of fruits per plant was observed in MS 2 (MC Set 2.0 L/ha) with 21.17 fruits and MS 3 (MC Set 3.0 L/ha) with 20.50 fruits. The foliar application of MC Set treatments significantly influenced average fruit weight. The maximum average fruit weight (83.72g) was observed in MS 2 (MC Set 2.0 L/ha). The yield per plant and yield per hectare were

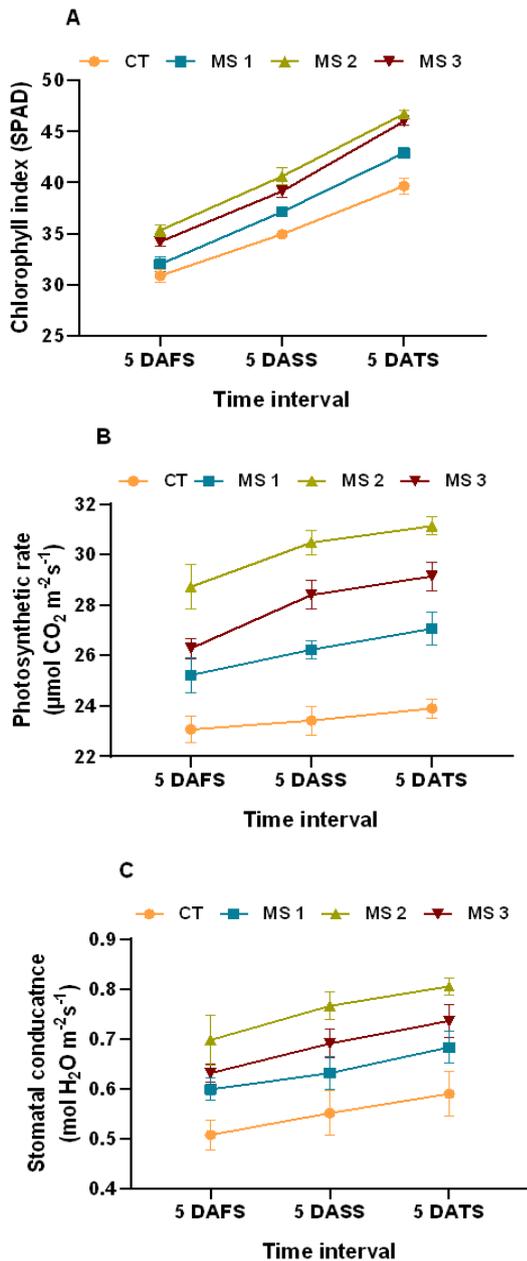


Fig. 1. Effect of MC Set on gas exchange and chlorophyll index of tomato at 5 days after first, second and third spray A) Photosynthetic rate, B) Stomatal conductance, C) Chlorophyll index (SPAD); DAFS – Days after first spray, DASS- Days after Second Spray, DATS- Days after third spray. Data in the figure are expressed as mean \pm SE

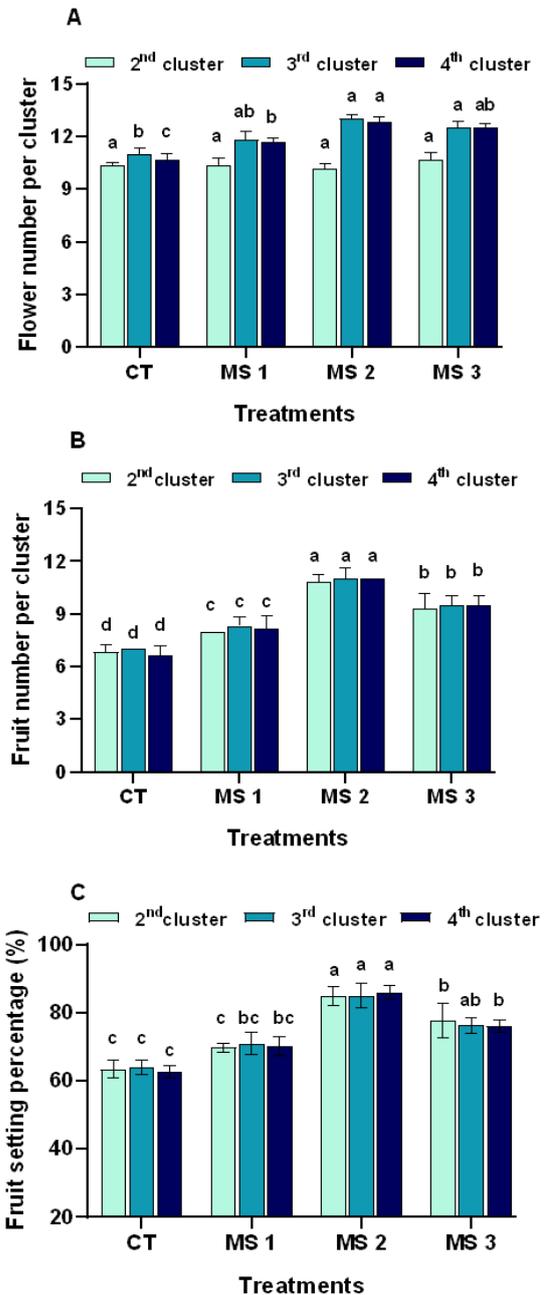


Fig. 2. Effect of MC Set on flowering and fruit setting percentage of 2nd, 3rd and 4th cluster. A) Flower number per cluster, B) Fruit number per cluster, C) Fruit setting percentage; Data in the figure are expressed as mean \pm SE. Mean values followed by the same letter do not differ significantly at P \leq 0.05 by DMRT

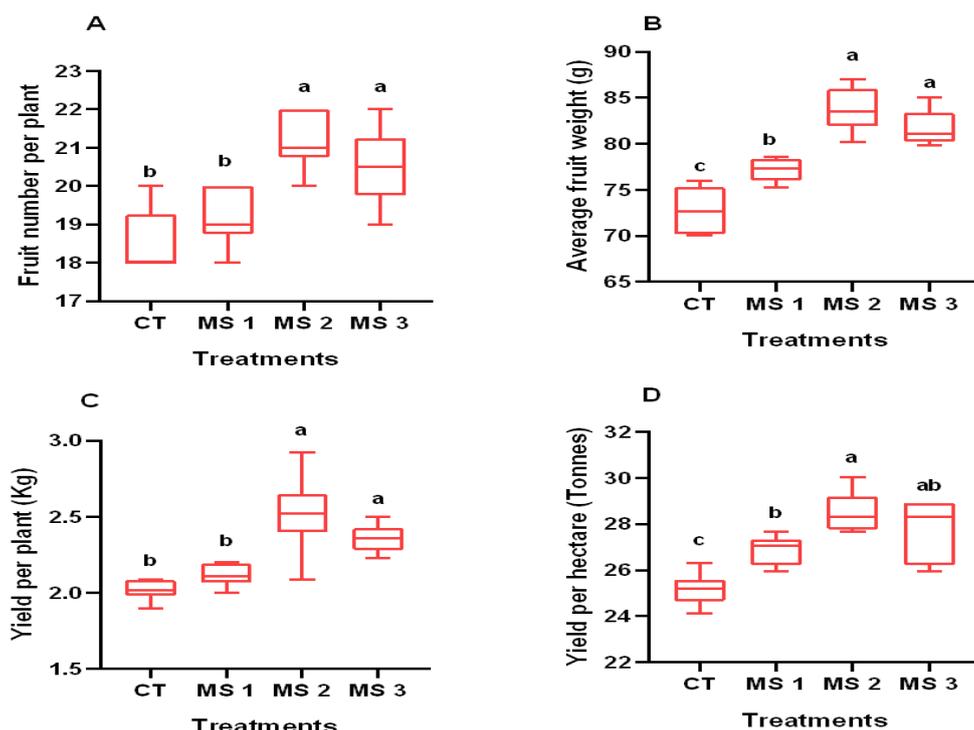


Fig. 3. Effect of MC Set on yield and yield traits of tomato. A) Fruit number per plant, B) Average fruit weight, C) Yield per plant, D) Yield per hectare (Tonnes); Data in the figure are expressed as mean \pm SE. Mean values followed by the same letter do not differ significantly at $P \leq 0.05$ by DMRT

significantly increased by foliar spraying of MC Set treatments and the greater yield per plant (2.52 kg) and yield per hectare (28.51 tonnes) were observed in MS 2 (MC Set 2.0 L ha⁻¹).

The correlation analysis assured that foliar application of MC Set positively improved tomato yield parameters ($r^2 = 0.791$) by enhancing the photosynthetic rate and fruit setting percentage (Table 2). The similar results were found by Subramaniyan *et al.*, 2023; Hussain *et al.*, 2021; Di Mola *et al.*, 2023; Ali *et al.*, 2016; Di Stasio *et al.*, 2020; Murtic *et al.*, 2018; Mannino *et al.*, 2020; Colla *et al.*, 2017. The brown seaweed *A. nodosum* contains alginates, polysaccharides which have functional molecules that leads to trigger plant signalling, increases nutrient uptake and nitrogen metabolism resulting in better plant growth and yield of tomato (Ahmed *et al.*, 2019; Subramaniyan *et al.*, 2023; Khan *et al.*, 2009). The seaweed extracts promote the beneficial microbial colonization in the root, which solubilizes the nutrients and increases the nutrient uptake by plant root, thereby it enhances the assimilate production and yield (Sani *et al.*, 2020) and this increased yield is also due to increased hormone activity in plants (Colla *et al.*, 2017). The boron and zinc nutrients present in the MC Set biostimulant may also have a stimulatory effect on yield and yield parameters of tomato; Ali *et al.* (2015) reported that boron and zinc nutrient improved the fruits per cluster and per plant, single fruit weight, fruit length, diameter and yield. Zinc and boron significantly im-

proved fruit growth by promoting the synthesis of tryptophan and auxin and boron has a crucial role in photosynthates accumulation which correlated the higher yield (Ali *et al.*, 2015). According to Dookie *et al.* (2020), yield is directly influenced by flowering; foliar application of *A. nodosum* extract enhanced the expression levels of six key flowering genes in tomato, increasing the yield (Dookie *et al.*, 2020).

Effect of MC Set on quality traits of tomato fruits

The foliar application of three different doses of MC Set treatments significantly enhanced the fruit quality parameters over a control (Fig. 4). Among the three doses of MC Set biostimulant, the highest total soluble solids (5.51°brix), ascorbic acid (35.00 mg100g⁻¹), lycopene content (3.44 mg g⁻¹) and total sugars (5.03 mg100g⁻¹) were found in MS 2 (MC Set 2.0 L ha⁻¹). Positive correlation was observed between MC Set treatment and quality parameters of tomato (Table 2). The different products derived from *A. nodosum* seaweed represent an increase in fruit quality parameters of tomato like total soluble solids, ascorbic acid, lycopene and total sugars (Sani *et al.*, 2020; Colla *et al.*, 2017; Rouphael *et al.*, 2021; Mannino *et al.*, 2020; Murtic *et al.*, 2018; Stasio *et al.*, 2017; Mzibra *et al.*, 2021; Mola *et al.*, 2023). The more TSS content of tomato may be due to the higher metabolic activities of fruits that result in sugar metabolite accumulation (Mzibra *et al.*, 2021; Hussain *et al.*, 2021). The application of 0.2% and

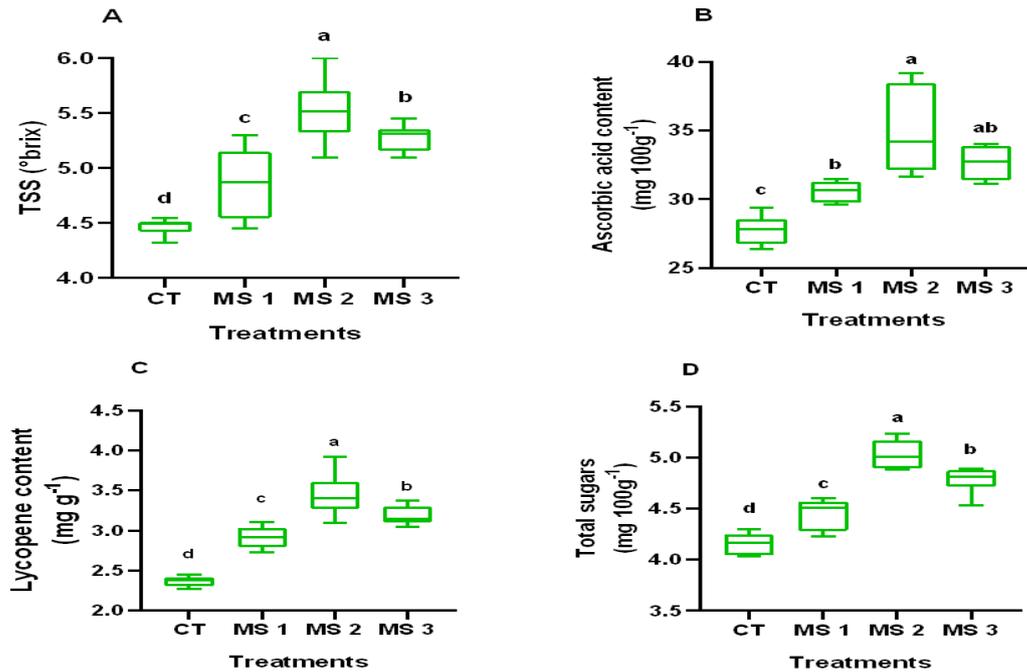


Fig. 4. Effect of MC Set on quality parameters of tomato. A) TSS (Total soluble solids), B) Ascorbic acid content, C) Lycopene content, D) Total sugars; Data in the figure are expressed as mean \pm SE. Mean values followed by the same letter do not differ significantly at $P \leq 0.05$ by DMRT

0.5% of *A. nodosum* extract significantly improved the TSS content of tomato fruits and they suggest that increasing concentrations of seaweed extract increased the TSS content of fruit (Ali *et al.*, 2016).

Ascorbic acid serves as a crucial antioxidant that effectively scavenges hydrogen peroxide, superoxide, and singlet oxygen. The antioxidant capacity of ascorbic acid plays a significant role in determining the nutritional quality of fruits (Di Mola *et al.*, 2023). In the present study, application *A. nodosum* along with boron and zinc improved the ascorbic acid content of tomato fruits. The brown seaweed has growth-enhancing stuff which enhances the nutritional quality and vitamin content of tomato, and thereby it increases the ascorbic acid (vitamin C) content of fruits (Stasio *et al.*, 2018; Mannino *et al.*, 2020; Di Mola *et al.*, 2023). The pigment present in tomato fruit is lycopene, an effective antioxidant that gives red colour to the fruit. Application of *A. nodosum* positively enhances the lycopene of tomato fruits which may be due to the presence of bioactive compounds, nutrient present in seaweed may stimulate specific metabolic pathways (Mannino *et al.*, 2020). *A. nodosum* enhances nutrient uptake in plants, particularly potassium (K), which activates enzymes like phosphofructokinase and pyruvate involved in carbohydrate metabolism and carotenoid biosynthesis. This mechanism leads to an increase in lycopene content in tomato (Colla *et al.*, 2017). Total sugars in tomato fruits are also enhanced by the foliar application of seaweed extract, which represents the taste of fruit. The rise in sugar content in biostimulant treatments can be attribut-

ed to the quick conversion of acids into sugars, enhanced photosynthetates assimilation, and the allocation of assimilates into sink organs such as fruits (Sani *et al.*, 2020; Subramaniyan *et al.*, 2023).

Plethora of reports are confirmed that application of *A. nodosum* improved the growth, nutrient uptake, yield and quality, and stress tolerance of the tomato. However, the effects of *A. nodosum* on tomato crops were well studied by supplementation of formulation contain solely *A. nodosum* based extract. Therefore, the combination effects of *A. nodosum* based extract and micro-nutrients (Boron and zinc) on tomato remained scarce and needed to study. Our results are confirmed that foliar application of MC Set, combination of *A. nodosum* extract and boron and zinc, improved growth, flowering, fruit setting, yield and quality traits of tomato, however, significant effects were observed in the middle dose (MC Set 2.0 L ha⁻¹). Moreover, the effects of biostimulants on tomato growth and yield depend on the application method, concentration, environmental conditions, and tomato variety. The present study confirmed that among three different dosages MC set, middle dose (2.0 L ha⁻¹) significantly improved tomato's physiological, yield and quality traits.

Conclusion

Results from this study suggest that foliar application of *A. nodosum* containing bio stimulant MC Set significantly improved the plant photosynthetic performance, flowering number, fruit setting percentage, yield and

quality of tomato. The three different concentrations of MC Set viz, MS 1 (MC Set 1.0 L ha⁻¹), MS 2 (MC Set 2.0 L ha⁻¹) and MS 3 (MC Set 3.0 L ha⁻¹) doses enhanced the plant performance compared to the control, but the treatment MS 2 (MC Set 2.0 L ha⁻¹) had performed better while comparing other two treatments and control. The foliar application of *A. nodosum* biostimulant product significantly raised the physiological traits like photosynthetic rate, stomatal conductance and chlorophyll index, and it also increased the number of flowers per cluster, number of fruits per cluster and fruit setting percentage of tomato. The MC Set treatments had significantly increased yield parameters viz, number of fruits per plant, average fruit weight, yield per plant and per hectare and it enhanced the fruit quality by improving TSS, ascorbic acid content, lycopene and total sugars. However, molecular approaches will need to study the effect of *A. nodosum* on enhancing tomato flowering. Hence, using MC set biostimulant (*A. nodosum* extract) could be the best way for sustainable tomato production.

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Conflict of interest

The authors declare that they have no conflict of interest.

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